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REGULAR PAPER

On the identity of the rare *Fragilaria subconstricta* (Fragilariaceae), with *Fragilaria* species forming ribbon-like colonies shortly reconsidered

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Background and aims – During phytoplankton surveys of a freshwater pond in Northeastern France, *Fragilaria subconstricta* (Bacillariophyceae) was observed. This rare diatom was present in large numbers in several plankton samples from Lachaussée Pond in the French Grand Est region.
Methods – The morphology of *Fragilaria subconstricta* was examined in detail using light and scanning electron microscopy. Its identity is discussed and it is compared to the morphologically most similar species, especially *Fragilaria capucina*, *F. mesolepta* Rabenhorst, *F. neointermedia* and *F. tenuistriata*.
Key results – *Fragilaria subconstricta* produces ribbon-like colonies and has narrow linear-lanceolate valves with slightly rostrate ends. The central area is rectangular, usually small or even absent. Spatulate linking spines are present all along the valve margins and a single rimoportula is located near one apex at the valve face-mantle junction. An amended diagnosis and new synonyms are proposed for this species. Additionally, the type material of *Fragilaria mesolepta* and a population of the very rare species *Fragilaria capucina* have been investigated and illustrated.
Conclusions – *Fragilaria subconstricta* and *F. tenuistriata* are synonyms. *Fragilaria capucina* s. str. is a rare species in Europe, and the majority of records for this species should be considered part of the *Fragilaria capucina* "Sippenkomplex" *sensu* Krammer & Lange-Bertalot (1991).

Key words – Bacillariophyta, *Fragilaria subconstricta*, *Fragilaria capucina*, taxonomy, scanning electron microscopy.

INTRODUCTION

The diatom genus *Fragilaria* Lyngbye (1819) was established for species forming linear, flat and articulate colonies, with corner joints alternately loosened and not adhering to each other. Hustedt (1930: 132) stated that *Fragilaria* formed very long ribbon colonies and that *Synedra* Ehrenb. cells could live individually or forming star-shaped colonies. One year later (Hustedt 1931: 138), he stated that "The genus *Fragilaria* is so closely related to the other genera of this group that a systematic separation seems hardly justified. In particular, the relationship with the genus *Synedra* is so narrow that isolated individuals are indistinguishable at all, also some *Synedra* species occur in ribbon-like colonies". However, he still separated these two genera (Hustedt 1931: 115–117). Lange-Bertalot (1980) combined all the common freshwater species of *Synedra* with *Fragilaria*, which resulted in a more broadly defined genus with twelve morphologically dis-

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Table 1 – Values of physical and chemical parameters from Lachaussée Pond on 20 Jul. 2016.
DOC: Dissolved Organic Carbon; COD: Chemical Oxygen Demand; BOD_5: Biochemical Oxygen Demand after 5 days.

	Lachaussée Pond		Lachaussée Pond
NH_4^+ (mg.l^{-1})	0.059	Pheopigments (µg.l^{-1})	0.6
NO_2^- (mg.l^{-1})	0.01	Ca^{2+} (mg.l^{-1})	47
NO_3^- (mg.l^{-1})	0.5	Cl^- (mg.l^{-1})	5.6
Kjeldhal Nitrogen (mg.l^{-1})	1.1	Mg^{2+} (mg.l^{-1})	4.3
Total Nitrogen (mg.l^{-1})	1.1	K^+ (mg.l^{-1})	3.6
PO_4^{3-} (mg.l^{-1})	0.015	SiO_2 (mg.l^{-1})	0.9
Total Phosphorus (mg.l^{-1})	0.02	Na^+ (mg.l^{-1})	4.2
SO_4^{2-} (mg.l^{-1})	10.4	Conductivity (µS.cm^{-1})	289
DOC (mg.l^{-1})	11.5	Suspended Matter (mg.l^{-1})	4
COD (mg.l^{-1})	37	pH	8.42
BOD_5 (mg.l^{-1})	2.4	Dissolved Oxygen (mg.l^{-1})	9.9
Chlorophyll *a* (µg.l^{-1})	12.3	Dissolved Oxygen saturation (%)	120

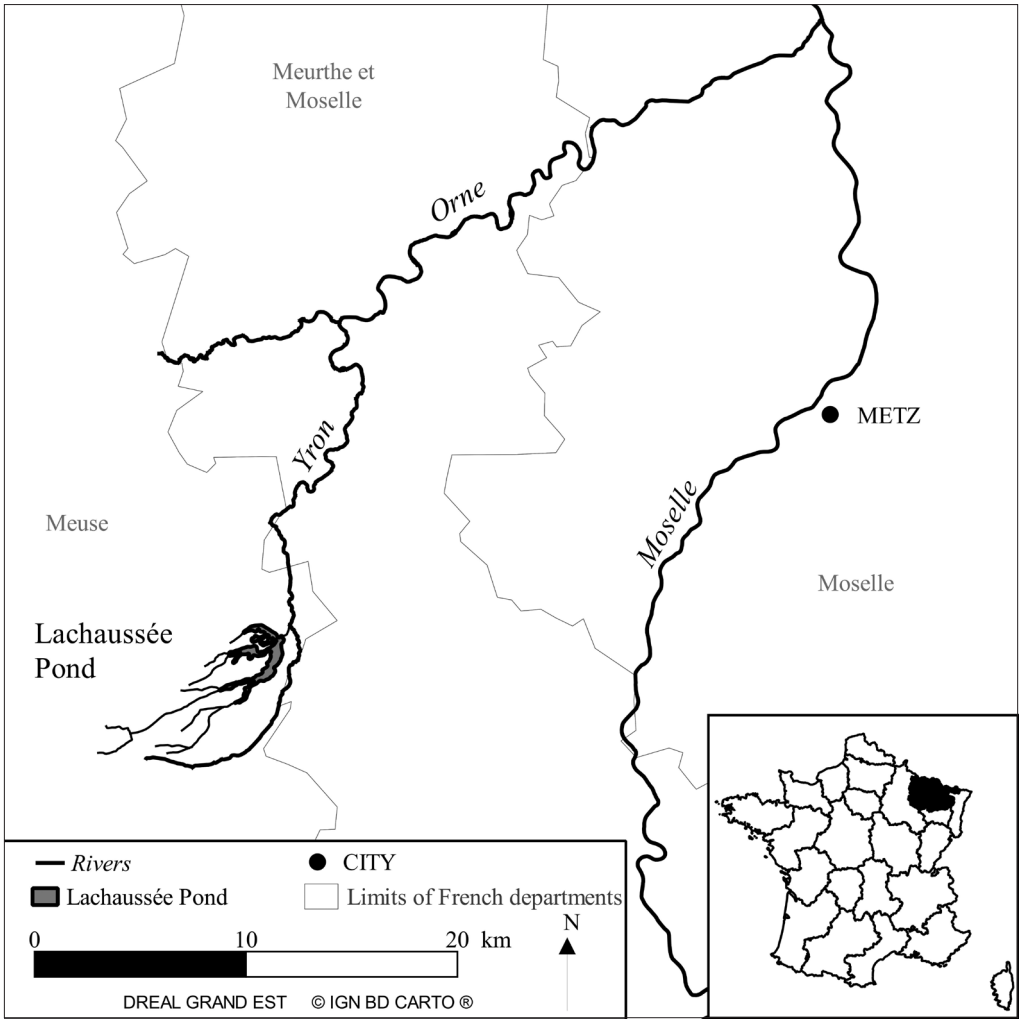

Figure 1 – Location of Lachaussée Pond in the Northeastern France. Map produced using QGIS (QGIS Development Team 2019) and BD Carthage (2019).

tinct sub-groups. Williams & Round (1987, 1988) resurrected *Staurosira* Ehrenb., and broadened the taxonomic discussion on the genus *Fragilaria* by adding several other morphological features such as the structure of the apical pore fields, the presence of only one apical rimoportula and uniseriate striae. Additionally, four new genera were described: *Staurosirella* D.M.Williams & Round, *Pseudostaurosira* D.M.Williams & Round, *Punctastriata* D.M.Williams & Round (1987) and *Fragilariforma* D.M.Williams & Round. Round et al. (1990) further restricted the circumscription of the genus *Fragilaria* to only include species with linear, linear-lanceolate to elliptical, usually capitate valves. Recently, further developments in the taxonomy and morphology of the *Fragilaria* genus definition were necessary to account for species with two rimoportulae such as *F. capucina* (Tuji & Williams 2006a). Even though the original definition of this genus would only include colonial species, currently two groups can be separated within *Fragilaria*: colonial and non-colonial species. However, it can be difficult to assert whether a species forms colonies or not, because it depends on characters like spines or apical pore fields. Moreover, untreated material is not always available for examination. *Fragilaria capucina* Desm., although rarely truly observed (i.e. *sensu* Tuji & Williams 2006a), is by far the most notorious *Fragilaria* species producing ribbon-like colonies and commonly, but often incorrectly (Ector et al. 2016) recorded in both present-day and historical literature worldwide. Other frequently recorded colonial species include *Fragilaria mesolepta* Rabenh., *Fragilaria neointermedia* Tuji & D.M.Williams, *Fragilaria nevadensis* Linares-Cuesta & Sánchez-Castillo, *Fragilaria pararumpens* Lange-Bert. et al. in Hofmann et al. (2011) and *Fragilaria rumpens* (Kütz.) G.W.F.Carlson.

This paper describes a population of *Fragilaria subconstricta* Østrup that was observed in plankton samples from Lachaussée Pond (Eastern France) in 2016. Morphological comparisons are made with similar species of *Fragilaria* showing comparable ribbon-like colonies. The identity of *F. subconstricta* is also discussed as there was a mismatch of name when it was epitypified. Additionally, the type material of *Fragilaria mesolepta* has been investigated and illustrated using SEM. Finally, in this paper a population of the very rare species *Fragilaria capucina*, collected in a small river in Luxembourg, was analysed, discussed and illustrated using LM and SEM.

MATERIAL AND METHODS

Study area

Lachaussée Pond is situated about 40 km to the West of the city of Metz (France), in the Woëvre plain located in the middle of the western part of the Lorraine Regional Natural Park (fig. 1). Its origins date back to the Middle Ages. In 1273, a dam was built to clean up and drain the nearby farmland. The pond thus created also made extensive fish farming possible (Conservatoire d'Espaces Naturels Lorraine 2012). With an elevation of 219 m, the pond has a surface area of 2.58 km² and a total volume capacity of 5.8×10^6 m³. The maximal

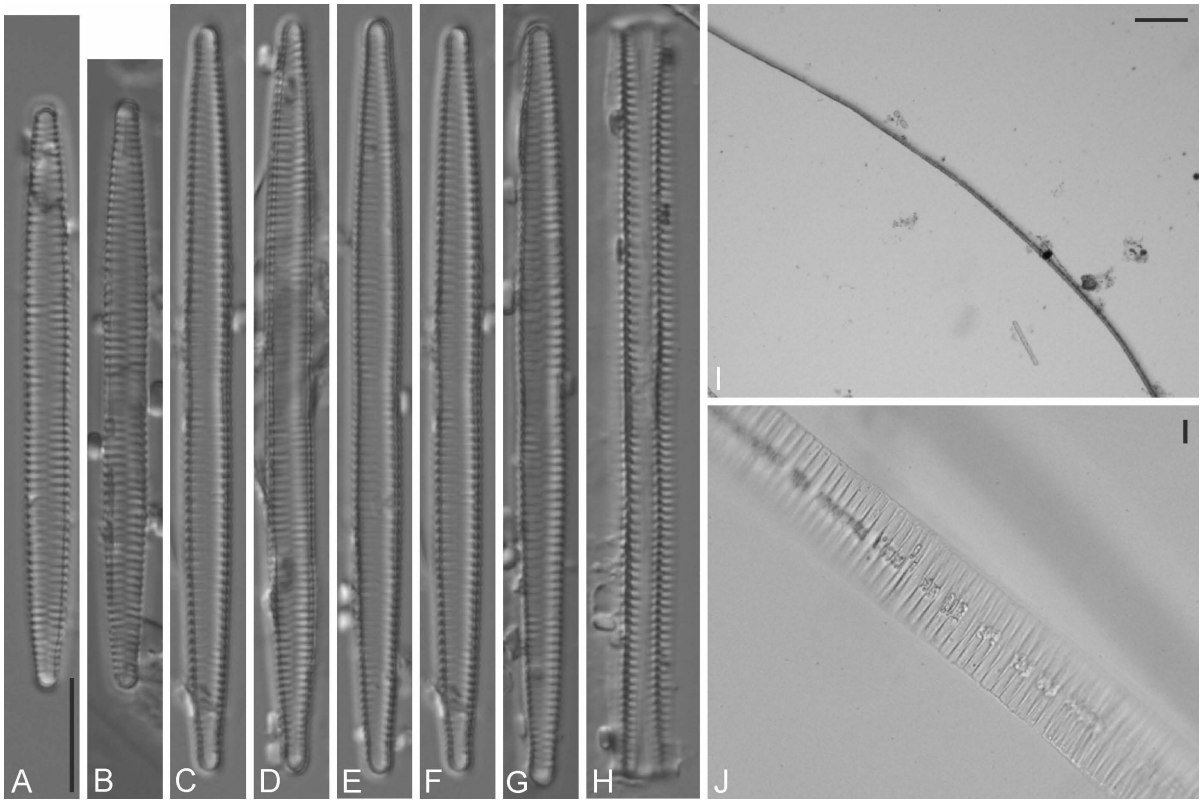

Figure 2 – *Fragilaria subconstricta* LM valve (A–G) and girdle (H–J) views (material from Lachaussée Pond at Lachaussée): A–H, treated material from Lachaussée Pond (France); I & J, live material from Lachaussée Pond (France) fixed with Lugol's solution. Scale bars: A–H & J = 10 µm; I = 100 µm.

Table 2 – Comparison of biometric data and morphological features of *Fragilaria subconstricta* with closely related taxa.
If several references are quoted, the symbol * shows the one where the biometric data are taken. The symbol ** indicates that the number of striae was measured on the plate in Tuji & Williams (2013), which is different from the density provided in diagnose (8–10 striae in 10 µm).

	F. capucina type material	*F. capucina* (Grouf stream, Luxembourg)	*F. mesolepta* type material	*F. neointermedia*	*F. nevadensis*	*F. pararumpens*	*F. rumpens*	*F. subconstricta* (Lachaussée Pond)	*F. subconstricta sensu* Østrup	*F. tenuistriata sensu* Østrup
Valve length (µm)	28–47	49–78	20–60	25–35	23.9–47.7	25–50	25–63	44.8–67.6	76	40–50
Valve width (µm)	3.3–4.2	3.5–4.7	3.5–4.5	3.5–4.5	3.2–4.1	2.5–3.0	3.0–4.0	3.2–4.2	3.6	3.6
Valve shape	Linear	Linear	Linear, more or less constricted at the centre	Lanceolate	Lanceolate	Lanceolate	Linear to linear-lanceolate	Linear-lanceolate	Linear-lanceolate	Linear-lanceolate
Apices	Weakly rostrate	Weakly rostrate	Subcapitate	Rostrate and rounded	Subcapitate	Subcapitate	Obtusely rounded	Slightly rostrate	Slightly rostrate	Slightly rostrate
Central area	Rectangular to rhombic	Rectangular to rhombic	Indistinct or absent	Slightly unilateral	Elliptic	Elliptic	Elliptic to rectangular	Small to absent	Rectangular	Rectangular
Number of striae in 10 µm	14–17	14–17	15–18	10–12**	14–18	16–18	18–20	12–15 (15–17 near apices)	20	17
Striation pattern	Alternate, parallel to slightly radiate towards the apices	Alternate, parallel to slightly radiate towards the apices	Alternate, parallel to slightly radiate towards the apices	Alternate, parallel or slightly radiate	Alternate, parallel	Alternate, parallel	Alternate, parallel	Parallel, more or less alternate	Parallel, more or less alternate	Parallel, more or less alternate
Number of rimoportulae per valve	2	2	1	1	1	1	1	1	?	1
Rimoportulae location	Valve face, near the poles	Valve face, near the poles	Valve face, near a pole, close to mantle junction	Valve face–mantle junction	Valve face, near a pole	Valve face, near a pole	Valve face, near a pole	Valve face–mantle junction	Valve face–mantle junction	Valve face–mantle junction
Linking spines	Conical near the apex to triangular in the middle	Conical near the apex to triangular in the middle	Spatulate	Spatulate	Spatulate, but the few closest to the apex are triangular	Conical near the apex to spatulate in the middle	Conical near the apex to triangular in the middle	Spatulate	Spatulate	Spatulate

Table 2 (continued) – **Comparison of biometric data and morphological features of *Fragilaria subconstricta* with closely related taxa.**
If several references are quoted, the symbol * shows the one where the biometric data are taken. The symbol ** indicates that the number of striae was measured on the plate in Tuji & Williams (2013), which is different from the density provided in diagnose (8–10 striae in 10 µm).

	F. capucina type material	*F. capucina* (Grouf stream, Luxembourg)	*F. mesolepta* type material	*F. neointermedia*	*F. nevadensis*	*F. pararumpens*	*F. rumpens*	*F. subconstricta* (Lachaussée Pond)	*F. subconstricta sensu* Østrup	*F. tenuistriata sensu* Østrup
References	Desmazières 1830, Krammer & Lange-Bertalot 1991, Tuji & Williams 2006a, Hofmann et al. 2011, Delgado et al. 2015*, Lange-Bertalot et al. 2017	This paper	Rabenhorst 1861, 1864, Krammer & Lange-Bertalot 1991, Kobayasi et al. 2006, Tuji & Williams 2008, Hofmann et al. 2011, Lange-Bertalot et al. 2017*	Tuji & Williams 2013	Linares-Cuesta & Sánchez-Castillo 2007, Novais et al. 2019*	Hofmann et al. 2011*, Lange-Bertalot et al. 2017	Kützing 1844, Carlson 1913, Lange-Bertalot 1980, Krammer & Lange-Bertalot 1991, Lange-Bertalot & Metzeltin 1996, Tuji & Williams 2006b*, Hofmann et al. 2011, Lange-Bertalot et al. 2017	This paper	Østrup 1910*, Krammer & Lange-Bertalot 1991, Tuji & Williams 2008	Østrup 1910*, Krammer & Lange-Bertalot 1991, Tuji & Williams 2008, Reichardt 2018

depth is about 4 m with an average depth of only c. 0.8 m. Four tributary rivers flow into the pond. The outlet is the Hattonville River, a tributary of the Yron River (http://www.rhin-meuse.eaufrance.fr/). The pond gathers the outlets of seven smaller interconnected ponds. Nowadays, there is ongoing fish farming activity and the pond is therefore drained annually. The site was designated as a wetland of international importance under the Ramsar Convention on Wetlands in 1991.

Processing and observations

For each illustrated species, samples were taken as follow.
Fragilaria capucina: an epiphytic sample on *Ranunculus* sp. from Grouf stream was collected in 2015 in Helfent, near Luxembourg.
Fragilaria neointermedia: the sample was collected from Brenon River in 2013 (Busseaut, Burgundy, France) according to the French diatom protocol (AFNOR 2007).
Fragilaria mesolepta: Rabenhorst type material (1861, n°1041) [Meise Botanic Garden, Belgium] containing *Fragilaria mesolepta* was observed in SEM. The type material was originally collected in 1858 in an artesian spring near Neustadt-Dresden (Germany).
Fragilaria pararumpens: the sample was collected from Beucinière River in 2016 (Lepuix, Territoire de Belfort, France) according to the French diatom protocol (AFNOR 2007).
Fragilaria subconstricta: four phytoplankton samples were collected from Lachaussée Pond from March to September, 2016 by a private organization (GREBE: Groupe de Recherche et d'Etude Biologie et Environnement) in charge of monitoring water quality control networks. Samples were collected in the euphotic zone with a water sampler (Niskin Type) at the deepest location of the pond and fixed in the field with Lugol's solution, according to the French national protocol (Laplace-Treyture et al. 2009).

Diatom slides were prepared following the methods described in the French diatom protocol (AFNOR 2007). Small aliquots of the samples were treated by oxidation with hot 37% hydrogen peroxide (H_2O_2) and hydrochloric acid (HCl), and rinsed three times with deionized water. For light microscopy (LM) observations, cleaned diatoms were mounted with Naphrax®. LM and morphometric measurements were performed with an OLYMPUS BX53 microscope using a ×100 oil immersion super apochromat Nomarski DIC objective and a Jenoptik ProgRes Speed XT3 camera. Measurements were taken of 25 valves. Mean and standard deviation values are calculated and provided. The stria density in 10 µm was measured with exclusion of the central area. To allow the observation of valve views, colonies were broken using ultrasonic treatment (80 kHz, 2 minutes) of the unmounted samples. For scanning electron microscopy (SEM), parts of the oxidized suspensions were filtered with additional deionized water through a 3-µm Isopore™ polycarbonate membrane filter (Merck Millipore). Filters were mounted on aluminium stubs and coated with platinum using a Modular High Vacuum Coating System BAL–TEC MED 020 (BAL–TEC AG, Balzers, Liechtenstein). An ultra-high-resolution analytical field emission (FE) scanning electron microscope,

Hitachi SU–70 (Hitachi High-Technologies Corporation, Japan) operated at 5 kV and 10 mm working distance was applied. SEM images were taken using the lower (SE-L) detector signal and a tilting of up to 28°. Micrographs were digitally manipulated and plates containing light and scanning electron microscopy images were created using CorelDraw X8®.

Physical and chemical data (table 1) were taken from the "Système d'Information sur l'Eau Rhin-Meuse (S.I.E.R.M.)" (http://www.rhin-meuse.eaufrance.fr/). Measurements were taken of samples collected in the euphotic zone at the deepest point of the Lachaussée Pond on 20 Jul. 2016.

Terminology for the morphological structures of the valves follows Ross et al. (1979), Cox & Ross (1981) (stria and areola structure), and Barber & Haworth (1981) (valve and area morphology).

RESULTS AND DISCUSSION

Fragilaria subconstricta was found in several plankton samples from Lachaussée Lake in 2016 (maximum relative abundance on 20 Jul. 2016 with only 0.6% abundance of all the planktonic algae but 51% of diatom taxa). The largest observed colony had about 600 cells.

Fragilaria subconstricta and *Fragilaria tenuistriata* Østrup are very similar species. They were illustrated in light and scanning electronic microscopy by Krammer & Lange-Bertalot (1991) and Tuji & Williams (2008). Tuji & Williams (2008) proposed an epitype for both taxa assuming that the illustrations of the lectotype (Krammer & Lange-Bertalot 1991: 450, figs 17–21) show individuals with different size than the original drawing of Østrup (1910). Basionyms in the combinations (Tuji & Williams 2008: 509) show the correct figures numbers, that is to say Østrup (1910) fig. 122 for *F.*

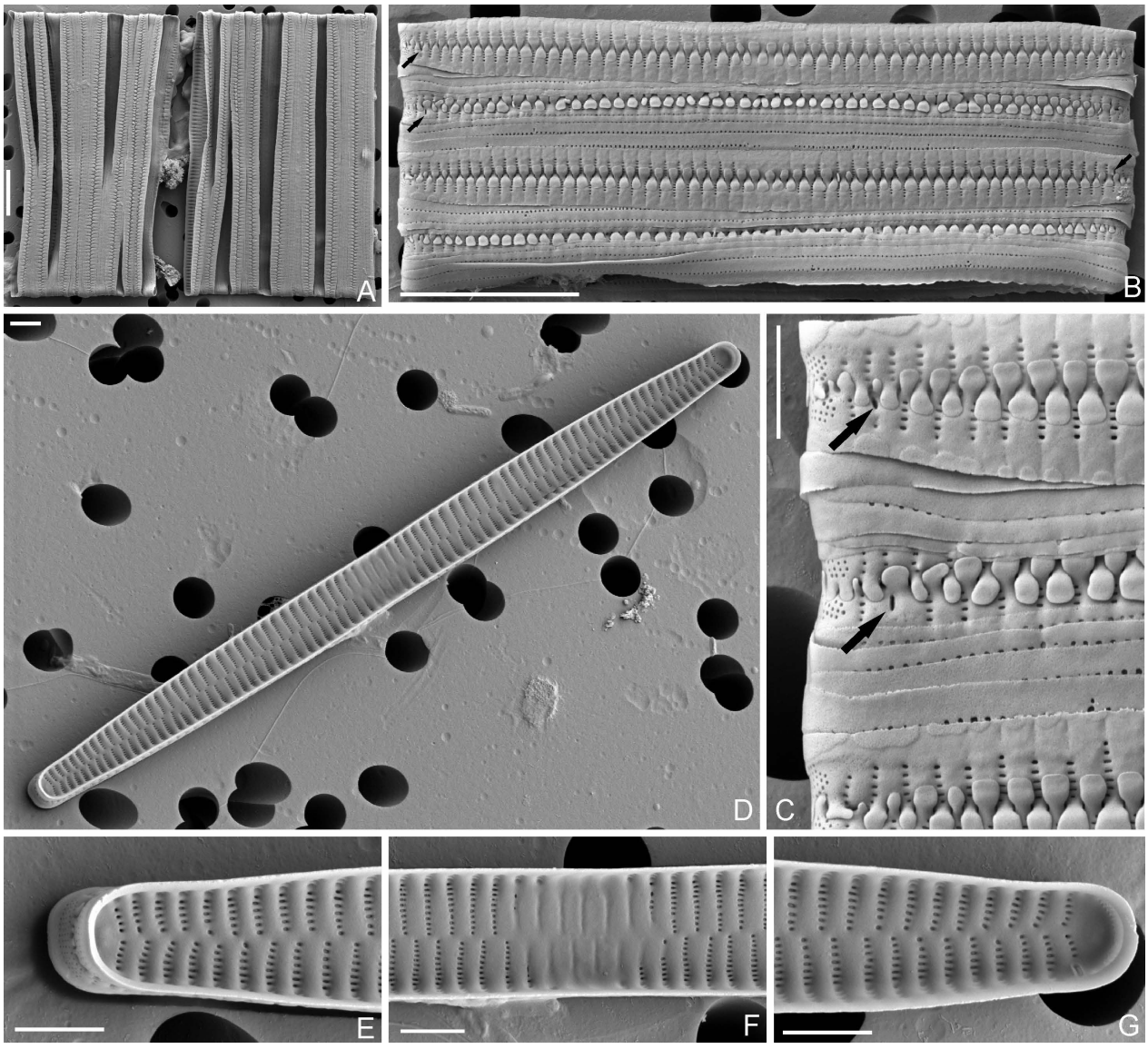

Figure 3 – *Fragilaria subconstricta* SEM views (material from Lachaussée Pond at Lachaussée): A & B, girdle view of colonies, black arrows indicate rimoportulae; C, close up on apices of B showing rimportulae (black arrows) and spatulate linking spines; D, valve in internal view; E–G, close up on apices and central area of D. Scale bars: A & B = 10 µm; C–G = 2 µm.

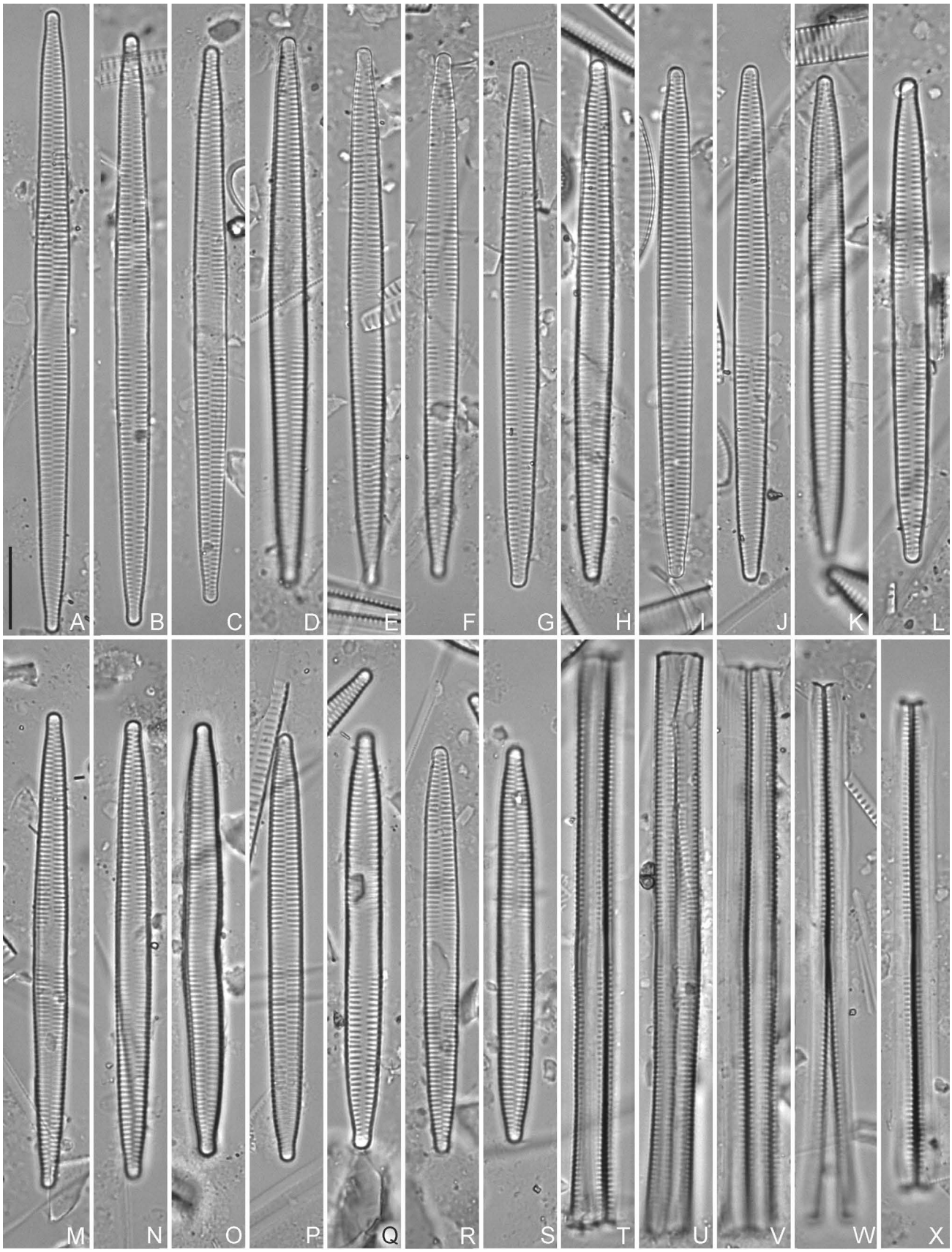

Figure 4 – *Fragilaria capucina* LM valve (A–S) and girdle (T–X) views (material from Grouf stream, Helfent, Luxembourg). Scale bar = 10 µm.

subconstricta and Østrup (1910) fig. 121 for *F. tenuistriata*. However, the conception of Tuji & Williams (2008) of these two species seems inverted in the plates compared to the original publication. The line drawing shown as *F. subconstricta* in Tuji & Williams (2008: fig. 35) is in fact the drawing of *F. tenuistriata* in Østrup (1910: fig. 121). This is confirmed by the identification key and the diagnosis (Østrup 1910: 185, 193) that clearly defined this species as shorter, linear and non-constricted in the central part contrary to *F. subconstricta*. And so, the drawing of *F. tenuistriata* in Tuji & Williams (2008: fig. 47) is the original drawing of *F. subconstricta* in Østrup (1910: fig. 122). This is also supported by the diagnosis of the species (Østrup 1910: 192). This mismatch with the names of these two taxa has been followed in

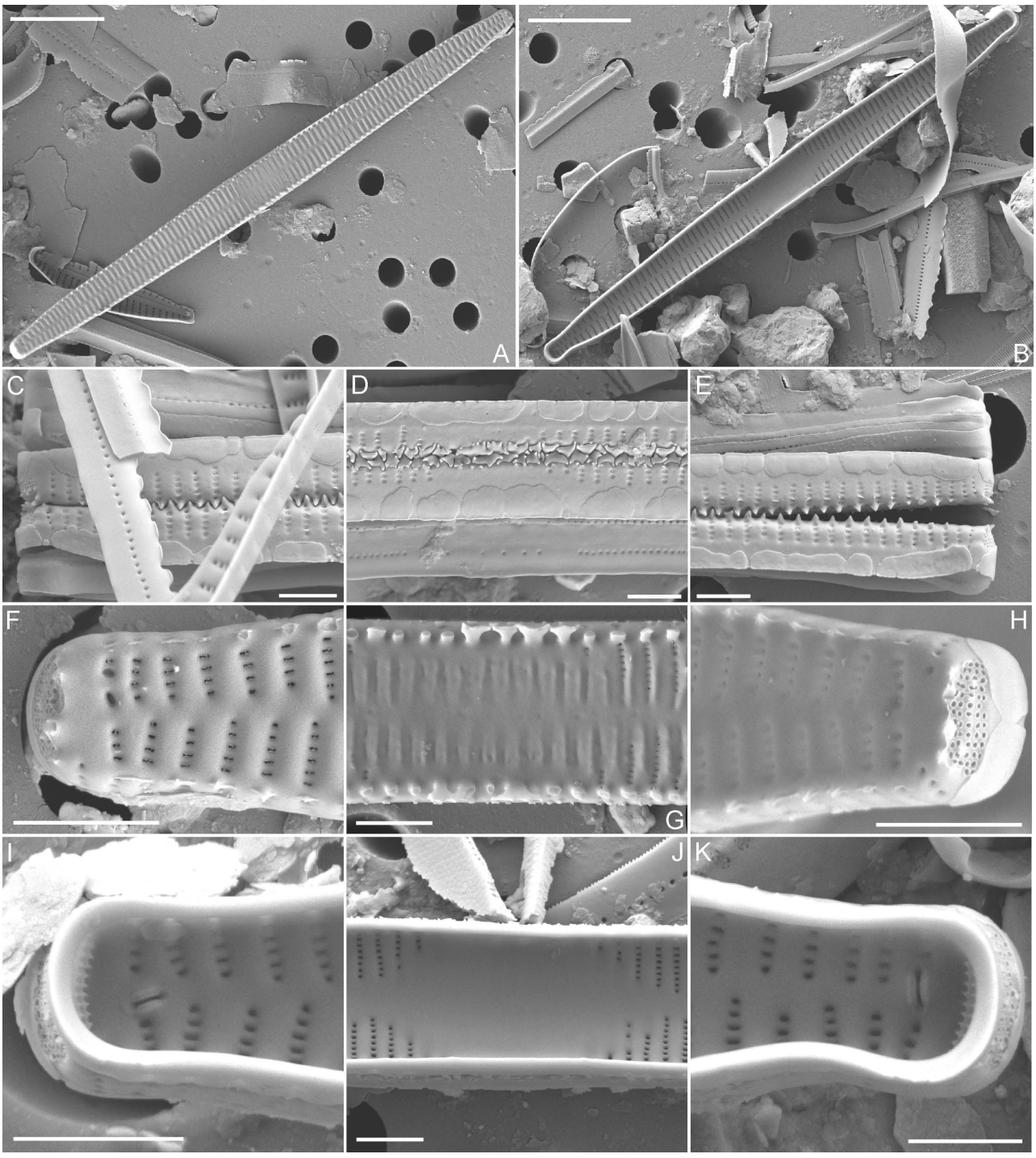

Figure 5 – *Fragilaria capucina* SEM view (material from Grouf stream, Helfent, Luxembourg): A, valve, external view; B, valve, internal view, C–E, close up on apices and central area of two linked valves in girdle view; F–H, close up on apices and central area of a valve in external view; I–K, close up on apices and central area of a valve in internal view. Scale bars: A & B = 10 µm; C–K = 2 µm.

recent literature (Chudaev & Gololobova 2016: pl. 25, figs 1–13, pl. 26, figs 1–5, Kulikovskiy et al. 2016: pl. 16, figs 29–34, Reichardt 2018: pl. 35, figs 20–37).

Our population from Lachaussée Pond is very similar to *F. subconstricta* sensu Østrup and only differs slightly by stria density and the alleged position of a rimoportula on the valve face by Tuji & Williams 2008 (i.e. *F. tenuistriata* sensu Tuji & Williams 2008: 506). They could not find the rimoportula in SEM and so they assumed that it was located on the valve face (Tuji & Williams 2008: fig. 46). This is most likely an artefact as the most similar species (i.e. *F. mesolepta* and *F. tenuistriata*) and our population from Lachaussée pond have a rimoportula on the valve/mantle junction (table 2). Moreover, *F. tenuistriata* specimens are certainly small forms of *F. subconstricta.* They are very similar in terms of valve outline and morphometric data. Small valves from Lachaussée Pond are not constricted in the central zone (fig. 2A & B) making them very similar to *F. tenuistriata*, while long valves are more similar to *F. subconstricta*. Both species were published in the same book (Østrup 1910) and are equally scarcely recorded. We therefore propose to keep the name *Fragilaria subconstricta* because it is the first name that was used, and because the name is more meaningful with respect to the morphology of the valve. We propose the following emended diagnosis.

Fragilaria subconstricta Østrup (Østrup 1910) emend. Heudre

Figs 2 & 3

Synonyms – *Fragilaria tenuistriata* Østrup (Østrup 1910), *Fragilaria tenuistriata* Østrup *sensu* Tuji & Williams (2008), *Fragilaria subconstricta* Østrup *sensu* Tuji & Williams (2008).

Diagnosis – Cells forming long flat ribbon-like colonies (fig. 2I & J). Valves narrow, linear-lanceolate, with rostrate poles. Valve face flat. Occasionally, slight constriction at valve centre (fig. 2E & G). Valve dimensions (n = 25): length 45–68 µm (average 61.4 ± 6.6 µm), width 3–4 µm (average 3.7 ± 0.2 µm). Sternum narrow, central area rectangular, small to absent, often differentiated from axial area only by less noticeable striation. Striae parallel, more or less alternating near central area, 12–15 (average 13.2 ± 0.9) in 10 µm, up to 15–17 (15.5 ± 0.7) in 10 µm near apices, clearly visible on mantle in girdle view (fig. 2H). Striae uniseriate (fig. 3D–G). First areola near axial area rounded, others becoming more elongate towards mantle (fig. 3E–G), 60–70 in 10 µm. Virgae wider than vimines (fig. 3D–G). Areolae continuing from valve face onto mantle (fig. 3B–G). Linking spines spatulate, located on vimines, in most cases, except near poles, when located on, or near, virgae (fig. 3C). One rimoportula present

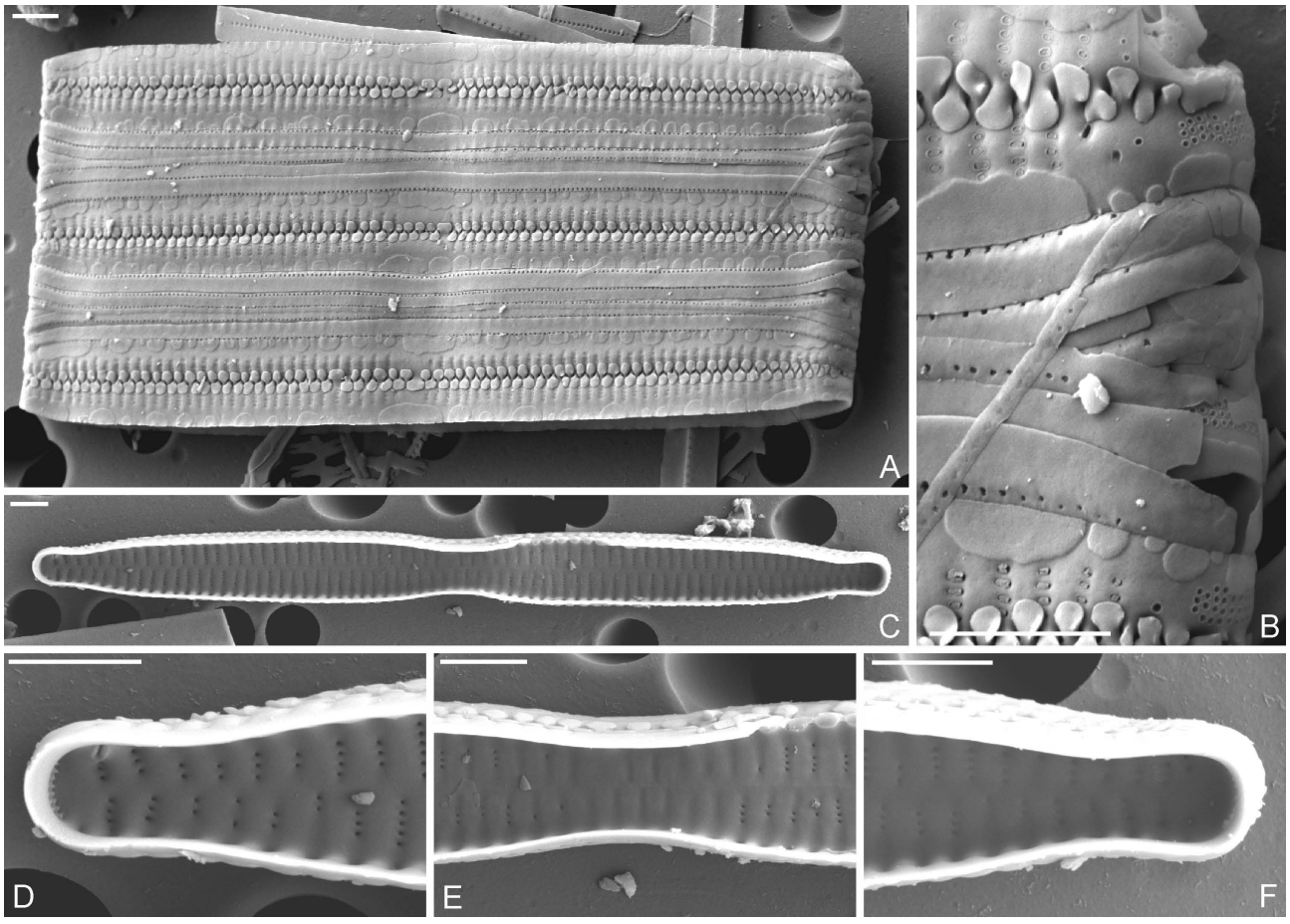

Figure 6 – *Fragilaria mesolepta* SEM view (Rabenhorst material n°1041, 1861, housed at BR): A, girdle view of colonies; B, close up on apices showing open girdle band and apical pore fields; C, valve, internal view; D–F, close up on apices and central area of C. Scale bars = 2 µm.

near apex, situated on valve face-mantle junction (fig. 3B, C & G, arrow). Rectangular apical pore fields present at both poles (fig. 3E & G). Silica plaques visible on valve mantle (fig. 3C). Around 5 open girdle bands with identical structure tapering towards their open ends, a single row of poroids just below the preceding band (fig. 3B & C). Valvocopula with undulated margin on interior part towards the valve (fig. 3B).

Ecology – Large populations of *F. subconstricta* were found in phytoplankton samples (120 cells/ml). Table 1 summarizes the chemical data from the Lachaussée pond where the species was found. This species occurs in a mesotrophic, slightly alkaline environment.

All of the following species also form ribbon-like colonies similar to *Fragilaria subconstricta*. *Fragilaria capucina* (figs 4 & 5) is separated by its more lanceolate valve shape and its large elliptical to rhombic central area (fig. 4A–S), higher stria density in the middle of the valve (14–17 in 10 µm), the presence of two rimoportulae (fig. 5F, H, I & K) (contrary to *F. subconstricta* that has only one rimoportula per valve) and conical to triangular spines (fig. 5C–E) whereas *F. subconstricta* has spatulate spines. The shape of spines and number or location of rimoportulae are admitted discriminating features in other genera (e.g. *Aulacoseira* Thwaites, *Staurosirella* and *Tabellaria* Ehrenb.). *Fragilaria mesolepta* (fig. 6, type material) has a higher stria density in the middle of the valve (15–18 in 10 µm) and subcapitate apices (fig. 6C, D & F) instead of rostrate apices in *F. subconstricta*. A rimoportula is located on the valve face-mantle junction (fig. 6B & D), but its exact position is hard to define because of its variability, it can be below a spine or between two of them. This species is also more clearly constricted in the centre of the valve (fig. 6C & E). Spines are spatulate and their shape does not vary along the valve (fig. 6A) as they do in e.g. *Fragilaria capucina*. *Fragilaria nevadensis* has more lanceolate valves than *F. subconstricta*, slightly bulging in the centre, with subcapitate apices and a large elliptic central area. It is shorter (30–50 µm) and slightly broader (3.5–5.0

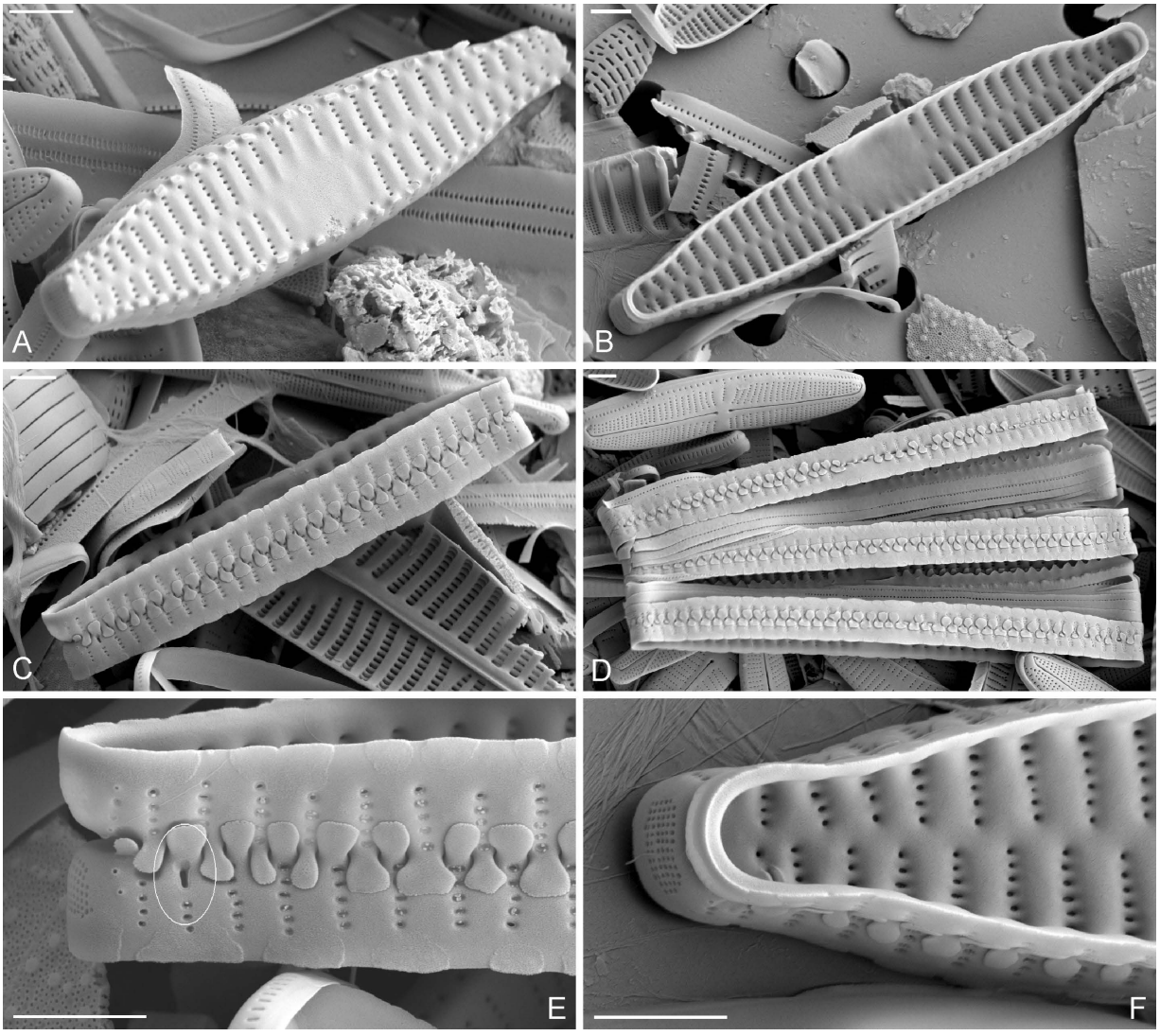

Figure 7 – *Fragilaria neointermedia* SEM view (material from Brenon River, Busseaut, Burgundy, France): A, valve, external view; B, valve, internal view; C, valve, girdle view; D, girdle view of colonies; E, close up on apex of C showing rimoportula on mantle (white circle); F, close up on apex of B. Scale bars = 2 µm.

µm) and has a rimoportula on the valve face (Linares-Cuesta & Sánchez-Castillo 2007: 128, figs 1–9). *Fragilaria pararumpens* (fig. 8) has a shorter (25–50 µm) and narrower (2.5–3.0 µm) lanceolate valves. It also has an elliptic central area (fig. 8A), higher stria density (16–18 in 10 µm) and a rimoportula on the valve face (fig. 8A & D). Finally, *Fragilaria rumpens* has an elliptic to rectangular central area, more striae in 10 µm (18–20), non-rostrated apices and conical to triangular spines (Tuji & Williams 2006b: 101, figs 17 & 18). *Fragilaria neointermedia* (fig. 7) is shorter (25–35 µm), more lanceolate and has a slightly unilateral central area (fig. 7A & B) where *F. subconstricta* is more linear and has an indistinct central area. *Fragilaria neointermedia* also has less striae in 10 µm (10–12) than *F. subconstricta*. It has similar spatulate linking spines and a rimoportula also located on the mantle (fig. 7E & F). It should be mentioned that the stria density given in the diagnose (Tuji & Williams 2013: 7) differs from the one measured on the plate (Tuji & Williams 2013: figs 38–42). Based on past observations of European populations we chose to retain the plate's value as the valid one and so we propose the following emended diagnosis.

Fragilaria neointermedia Tuji & D.M.Williams (Tuji & Williams 2013) emend. Heudre

Fig. 7

Synonym – *Fragilaria intermedia sensu* Grunow in Van Heurck (1881: pl. 45, figs 9, 10, not fig. 11).

Diagnosis – Valves lanceolate, narrowing toward rostrate, rounded apices. Sternum narrow. Central area slightly to one side of valve. Striae parallel or slightly radiate, 10–12 in 10 µm. Valve length 25–35 µm, width 3.5–4.5 µm. Apical pore fields situated at each apex (fig. 7A, B, E & F). Spatulate linking spines present (fig. 7C–E). One rimoportula per frustule, situated on valve face–mantle junction (fig. 7E & F).

Finally, *Fragilaria capucina* s. str. is in fact a rare species in Europe, or at least less frequent than implied by records in the literature. The large majority of the quoted observations of this species are to be considered as belonging to the *Fragilaria capucina* s. lat. as in Krammer & Lange-Bertalot (1991), before the varieties previously belonging to this species were established as distinct species, including those in recent literature such as Reichardt (2018: pl. 32, figs 1–39; pl. 38, figs 1–7). In conclusion, all *Fragilaria* species forming ribbon-like colonies need to be examined in more detail. *Fragilaria subconstricta* could have been identified as *F. capucina* when the colonies were observed using only an inverted microscope (fig. 2I & J). We recommend that samples are treated with ultrasound when colonies of species listed in table 2 are present so that distinctive features (table 2) can be examined in valve view.

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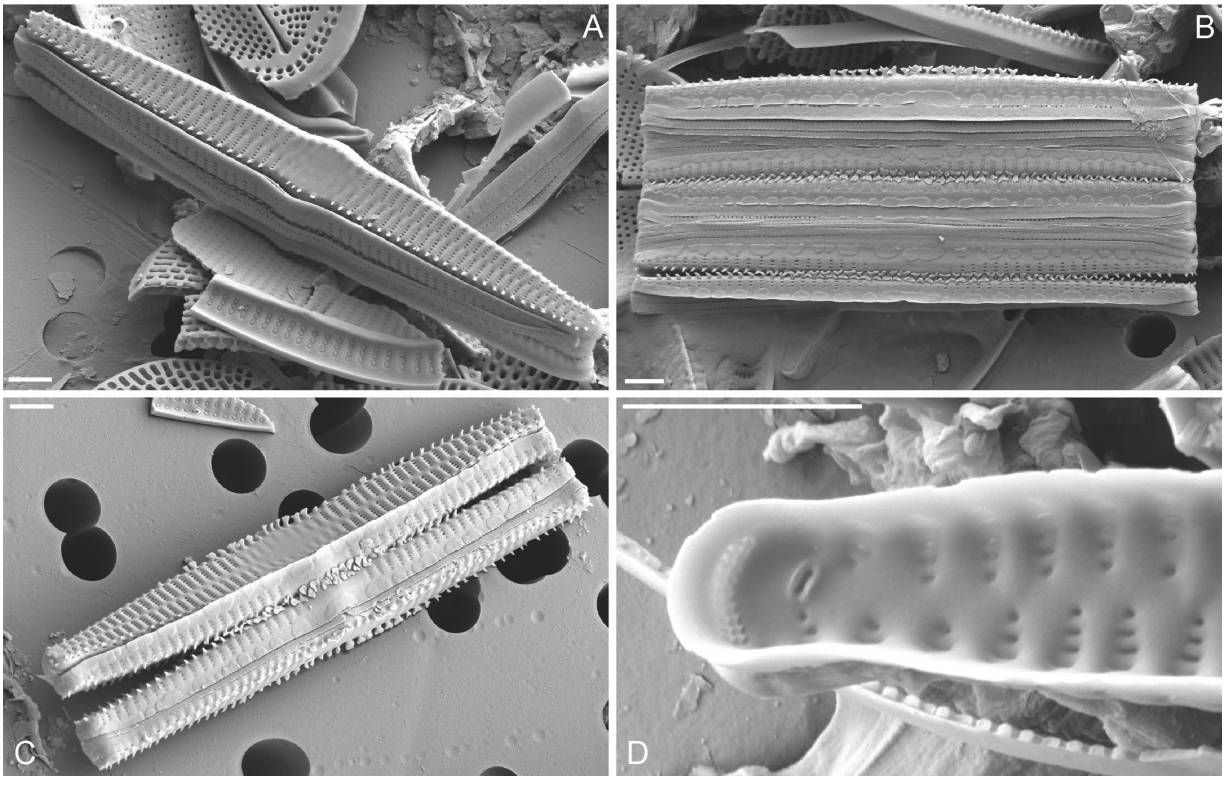

Figure 8 – *Fragilaria pararumpens* SEM view (material from Beucinière River, Lepuix, Territoire de Belfort, France): A, valve, external view; B & C, girdle view of colonies; D, close up on apex in internal view. Scale bars = 2 µm.

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