Observations on and typification of *Navicula fontinalis* Grunow (*Naviculaceae, Bacillariophyta*)

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Navicula fontinalis Grunow (in Van Heurck 1880, pl. 12: fig. 33, Van Heurck 1885: 103) was originally described and illustrated as "*Navicula (bacillaris* var.?) *fontinalis*". Van Heurck (1885) added the following description (translated from French): "Valve linear, with rounded apices. Raphe bordered by a lanceolate hyaline zone, widened in the middle of the valve forming a very large pseudo-stauros. Striae weakly radiate, 24–26 in 10 μ m, crossed by an almost marginal longitudinal groove. Length approx. 25 μ m, width 5 μ m" (Fig. 2). A (type) locality was added: "Fresh waters. – Brussels (Delogne)". A search of the Grunow collection (**W**) in Vienna, Austria did not reveal any record of where exactly the material was collected (Walter, pers. comm.), but in the Van Heurck collection in Meise Botanic Garden (**BR**), Belgium, a slide containing *Navicula fontinalis* (VII-19-C5, Fig. 1), was discovered, made from a sample collected by Charles-Henri Delogne (1834–1901). The sample, labelled *Diadesmis gallica* W.Smith, was collected in the Geranium Greenhouse of the former Jardin Botanique de Bruxelles on 3rd of May 1877.

According to the Van Heurck catalogue cards, Grunow identified the species in the slide adding a handwritten list on the slide label of taxa present (Fig. 1), viz, Diadesmis gallica (?), Nitzschia amphibia Grunow, Nitzschia denticula Grunow, Achnanthes lanceolata, Navicula appendiculata, Navicula fontinalis Grunow. Unfortunately, no sample number was provided on the slide. Meise Botanic Garden (**BR**) houses all samples collected by Delogne during his life. Part of these samples were for instance later used to make a set of 100 exsiccatae for the Diatomées de Belgique. A handwritten, incomplete list of samples accompanies the set. Based on this list, the number of possible samples from which the slide was made was reduced to three by selecting all samples for which the genus *Diadesmis* was listed amongst the genera present in the sample (Fig. 3). One of the samples (No. 142) was not collected in the Jardin Botanique de Bruxelles and hence is omitted from the list of possible sources for the slide containing *Navicula fontinalis*. Simultaneously, other slides containing *Diadesmis gallica* in the Van Heurck collection were checked. One of the slides, also based on a sample collected in the Jardin Botanique de Bruxelles, likewise contained valves that could be identified as N. fontinalis. The slide was labelled '165', made from one of the above selected samples in the Delogne sample set. All selected samples were processed for LM and SEM analysis. Only samples 137 and 165 showed a similar flora with the slide labelled N. fontinalis. As sample No. 165 also contained Sellaphora saugeresii (Desmazières) C.E.Wetzel & D.G.Mann, absent in sample No. 137, the latter is selected as the most likely source for the N. fontinalis slide, as on the original slide, S. saugeresii was also absent.

Navicula fontinalis Grunow was later transferred to the genus *Caloneis*, first as a variety of *C. lacunarum* (Grunow) Hustedt, *C. lacunarum* var. *fontinalis* (Grunow) Hustedt (1909: 437), then as a variety of *C. fasciata* (Lagerstedt) Cleve, *C. fasciata* var. *fontinalis* (Grunow) Østrup (1910: 11, 12). Subsequently, Cleve-Euler (1932) treated it as species, *Caloneis fontinalis* (Grunow) Cleve-Euler (1932: 130). The same combination was introduced in 1996 by Lange-Bertalot & Reichardt

(in Lange-Bertalot & Metzeltin 1996: 29), but the Cleve-Euler combination has priority, although in later publications, the 1996 combination is erroneously maintained (e.g., Lange-Bertalot *et al.* 2017).

Here, using light and scanning electron microscopy, we detail observations on specimens from a slide prepared from Delogne samples 137 and 165. Sample 137 is designated as lectotype for the species.

Caloneis fontinalis (Grunow) Cleve-Euler (Figs 3-53)

- Basionym: *Navicula fontinalis* Grunow in Van Heurck, *Synopsis des Diatomées de Belgique*, p. 103, 1885.
- Homotypic synonyms: *Caloneis lacunarum* var. *fontinalis* (Grunow) Hustedt (Hustedt 1909: 437), *Caloneis fasciata* var. *fontinalis* (Grunow) Østrup (Østrup 1910: 11, 12), *Navicula fasciata* var. *fontinalis* (Grunow) Frenguelli (Frenguelli 1924: 64, pl. 6: fig. 6).
- Lectotype (here designated): slide VII-19-C5 (**BR**), slide prepared from Delogne sample 137, Jardin botanique de Bruxelles, serre géraniums (coll. date 3.v.1877), material present in the Van Heurck collection (**BR**). The lectotype specimen is here represented by Fig. 16.
- Description: Frustules in girdle view rectangular, occasionally found in pairs. Valves linear with parallel margins and broadly rounded, weakly protracted apices. Mantle broad with mantle striae terminating in the upper 1/4 of the mantle. Rest of the mantle up to the mantle edge hyaline. Valve dimensions (n=50): valve length 11–30 μ m, valve width 3.5–5 μ m. Axial area broad, lanceolate, slightly widening from the apices towards the central area. Near the apices, axial area abruptly narrowing. Small, shallow depressions scattered all along the axial area. Central area forming a large hyaline fascia. Raphe filiform, straight to weakly undulating. Central raphe endings widely spaced, extending beyond the last central striae, weakly deflected and slightly enlarged. Terminal raphe fissures hooked, continuing onto the valve mantle reaching the edge of the mantle striae. Striae parallel to weakly radiate, 24–26 in 10 μ m. Striae short, almost of equal length throughout the entire valve except at the apices, multiseriate, composed of 3 rows of parallel, small, rounded areolae. Internally, alveoli very small, only marginally opening into the valve interior. Irregular series of small silica outgrowth present between the alveoli (Fig. 52). Internal raphe branches straight with indistinct, simple, weakly deflected central raphe endings and terminal endings terminating onto very small helictoglossae.
- Note: The valve dimensions indicated in the above description are based on a random analysis of the entire population. One possible initial valve was, however, observed with a valve length of $36 \mu m$ (Fig. 45).

Caloneis fontinalis is often reported from spring areas worldwide (e.g., Angeli *et al.* 2010, Lai *et al.* 2016). Published records unfortunately are rarely illustrated and their correct taxonomic identity is hence difficult to verify. Moreover, the species is most likely often reported as *Caloneis bacillum* (Grunow) Cleve. The latter, however, is a typical marine species with an entirely different morphology (Zidarova *et al.* 2016). A literature search revealed a large number of records of *Caloneis fontinalis*. Ideally these records should be reassessed but the slides upon which the records are based are often not conserved making it impossible to verify them. For all subsequent records, even if based on the illustrations, it is very likely that they represent a different species. Nevertheless, most of them show the typical features of *Caloneis fontinalis*. Important features that separate *C. fontinalis* from similar *Caloneis* species include the weakly, still visibly protracted valve apices, the striae that are not shortening towards the central area, the high stria density of 24–26 striae in 10 μ m (erroneously reported in Lange-Bertalot *et al.* 2017 being 21–23 in 10 μ m) and the weakly deflected central raphe endings.

Apart from Belgium (Denys & Oosterlynck 2015, this study), the species was then most likely correctly reported from Argentina (Frenguelli 1924), Sweden (Cleve-Euler 1932), Finland (Lange-

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Bertalot & Metzeltin 1996), Italy (Cantonati 1998, Lai *et al.* 2015), North Macedonia (Levkov *et al.* 2007), Japan (Nakajima *et al.* 2007, Hirota *et al.* 2013), Germany (Reichardt 2011), France (Ector *et al.* 2015), Poland (Peszek *et al.* 2015), United States (Bishop *et al.* 2017), Iran (Kheiri *et al.* 2018) and Serbia (Jakovljević 2019, erroneously reported as *C. bacillum* (plate 3, figs 12–14); the valves shown for *C. fontinalis* (plate 3, figs 15–18) represent a different species).

Some published illustrations of valves named as *C. fontinalis* are most likely not based, for instance, on their gradually shortening striae. These valves probably belong to a very similar, probably unknown species (Veselá & Johansen 2009, Solak & Wojtal 2012, Wojtal 2013, Mogna *et al.* 2015, Vidaković *et al.* 2016, Miho *et al.* 2018, Peeters & Ector 2020). Bąk *et al.* (2012) depict several valves from Poland named as *C. fontinalis* but most likely only one of them (Bąk *et al.* 2012: pl. 46, 4th figure from the left) could be identified as such. Further investigation of these *Caloneis* populations will, however, be necessary to establish their correct identity.

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Figs 1–47. **Fig. 1.** Delogne slide VII-19-C5 containing *Navicula fontinalis* Grunow (see label). **Fig. 2**. Original drawing of *Navicula fontinalis* Grunow in Van Heurck (1880, plate 12, fig. 33). **Fig. 3**. Extract from the handwritten catalogue of the Delogne sample collection. **Figs 4–22**. Cell cycle of *Caloneis fontinalis* (Grunow) Cleve-Euler taken from the lectotype material (Jardin botanique de Bruxelles, serre géraniums, sample 137). Figs 21–22 show frustules in girdle view. Fig. 16 represents the lectotype specimen. **Figs 23–46**. Cell cycle of *Caloneis fontinalis* (Grunow) Cleve-Euler taken from Delogne slide 165 (Jardin botanique de Bruxelles). Fig. 45 most likely represents an initial frustule in girdle view. Fig. 46 shows a frustule in girdle view. Fig. 35 represents the epitype specimen. **Fig. 47**. SEM view of an entire frustule taken from the lectotype material. Note the perforated girdle bands. Scale bars represent 10 μm.

6



Figs 48–53. *Caloneis fontinalis* (Grunow) Cleve-Euler. SEM micrographs taken from the lectotype material (Jardin botanique de Bruxelles, serre géraniums, sample 137). Fig. 48. SEM external view of an entire valve. Note the length of the striae and the shallow depressions in the axial area. Fig. 49. SEM external detail of a valve apex in girdle view showing the terminal raphe fissure and the girdle bands. Fig. 50. SEM external detail of a valve apex in valve face view. Fig. 51. SEM external girdle view of an entire frustule. Fig. 52. SEM internal detail of the siliceous outgrowths present between the alveoli. Fig. 53. SEM internal view of an entire valve. Scale bars represent 10 μ m except for figs 49, 50 & 52 where scale bar = 1 μ m.