

## APPENDICES

*The following appendix contains (1) a short presentation of species (using lists and figures) identified after the new species described in Carter (1966) which were found in moss-habitats of Gough Island, (2) a list of all diatom taxa and their authorities identified from the moss material during this survey, (3) abstracts of two posters that were presented on international conferences during my master studies dealing with the topic, and finally (4) a paper published in the second year of my master studies about diatom communities from Svalbard (High Arctic), therefore not concerning the topic of this thesis, but answering a similar ecological dispersal-related question, as to what extent adjacent habitats are responsible for structuring diatom assemblages.*

## APPENDIX 1

Diatom species identified after Carter (1966)

List of species and formas described in Carter (1966) from Tristan da Cunha and Gough Islands. Current names (if changed) or corrected names (if wrongly identified). Species in bold are in following figures. Distribution: T – Tristan da Cunha Island, G – Gough Island, N – Nightingale Island, I – Inaccessible Island, C – reported also outside the archipelago.  
 × - sp. was found during counting, (×) – species has been observed only outside the counts.

TAXON NAME IN CARTER (1966)	Current name	Distribution	If found
<b>Achnanthes atalanta</b>	<b>Psammothidium atalanta</b> (J.R.Carter) L.Bukhtiyarova	T, G	×
<b>Achnanthes investians</b>	<b>Psammothidium investians</b> (J.R.Carter) L.Bukhtiyarova	T, G	×
<i>Achnanthes lanceolata</i> f. <i>rhombica</i>		T	
<i>Achnanthes natrata</i>		T	
<b>Achnanthes pericava</b>	<b>Planothidium pericavum</b> (J.R.Carter) Lange-Bertalot	T, G, C	×
<i>Achnanthes sumara</i>	(As <i>Achnanthes petersenii</i> Hust. in Lange-Bertalot & Krammer (1989: 117))	T	
<b>Amphora incurvata</b>		T	
<i>Caloneis liophilla</i>		G	
<i>Caloneis toxa</i>		T	
<i>Caloneis transepta</i>		T	
<i>Cyclotella stelligeroides</i> f. <i>irregularis</i>		G	
<b>Eunotia ambigua</b>		T, G	×
<b>Eunotia diverta</b>		G	×
<i>Eunotia incurva</i>		T	
<b>Eunotia morbida</b>		G	×
<b>Eunotia pectinoides</b>	<b>Eunotia johncarteri</b> (Carter) D.M.Williams	G	×
<i>Fragilaria brevistriata</i> f. <i>australis</i>		T	
<i>Gomphonema asymmetricum</i>	<i>Gomphonema johncarteri</i> (' <i>johncarterii</i> ') (Carter) D.M.Williams	T	
<b>Gomphonema spatulum</b>		T, G	×
<i>Melosira dispersa</i>			
<i>Melosira incorrupta</i>			
<b>Melosira setosa</b>	<b>Melosira johncarteri</b> (Carter) D.M.Williams	G	×
<i>Navicula abica</i>		G	
<i>Navicula amerinda</i>		T	
<i>Navicula brockmanii</i> f. <i>concave</i>		T	
<b>Navicula decissa</b>	<b>Humidophila decissa</b>	T, G	(×)
<b>Navicula frisca</b>	[as <i>Chamaepinnularia</i> sp2, this thesis]	T, G	×
<b>Navicula nienta</b>	<b>Humidophila nienta</b> (Carter) Lowe, Kociolek, Johansen, Van de Vijver, Lange-Bertalot & Kopalová	T, G, C	×
<i>Navicula orbita</i>		T	
<i>Navicula riparta</i>		G	
<i>Navicula scalpia</i>		G	
<i>Nitzschia epressa</i>		T	
<b>Nitzschia navia</b>		T, G	×
<b>Nitzschia oscilla</b>		T	(×)
<i>Nitzschia palmida</i>		G	
<b>Nitzschia petulla</b>		T, G	×
<i>Nitzschia pistilla</i>		T	
<b>Nitzschia rotula</b>	<b>Nitzschia terrestris</b> (J.B.Petersen) Hustedt	T, G, C	×
<i>Nitzschia salta</i>		T	
<i>Nitzschia serpentiformis</i>		G	
<i>Nitzschia sonora</i>		G	×
<b>Pinnularia dispersa</b>	<b>Pinnularia johncarteri</b> (Carter) D.M.Williams	T, G	×
<i>Pinnularia extenda</i>		T	
<i>Pinnularia impassa</i>		T	

TAXON NAME IN CARTER (1966)	Current name	Distribution	If found
<i>Pinnularia leptosomiformis</i>		G	
<b><i>Pinnularia posita</i></b>		G	×
<i>Pinnularia redunda</i>		G	
<b><i>Pinnularia restituta</i></b>		G	×
<i>Pinnularia ritarda</i>		T	
<i>Pinnularia seriata</i>		G	(×)
<b><i>Pinnularia sistassa</i></b>		T, G	×
<i>Pinnularia subcapitata</i> f. <i>plana</i>		T	
<b><i>Pinnularia vapilla</i></b>		T, G	×
<b><i>Pseudoeunotia linearis</i></b>	<b><i>Eunotia linearis</i></b> (Carter) Vinsová, Kopalová & Van de Vijver	G	×
<b><i>Pseudoeunotia duplex</i></b>		G	×

In the following part, each species identified during this study after what Carter (1966) described as new to the science, is shortly presented following the order of original taxon names as listed in the previous list. Nomenclature changes or corrected names are specified together with the island where the species was originally identified from. If not specified otherwise, the species has been observed and identified from Gough material during this study. Most of the species are also accompanied in respective figures of paired Carter's original drawings and pictures of light microscope (LM). Some of them are represented also by scanning electron microscope (SEM) pictures. Notes regarding morphological features are added when appropriate. One species, *Eunotia (Pseudoeunotia) linearis* is presented as a separate (published) paper in **Chapter 2**.

***Psammothidium atalanta*** (Carter) Bukhtiyarova (Bukhtiyarova et al. 2013)  
 Basionym: ***Achnanthes atalanta*** Carter, 1966: 444, pl. 1, figs. 15, 16, 19, 20.  
 Locality. Tristan da Cunha (Carter 1966).  
**Figures A1.19, A1.38.**

***Psammothidium investians*** (J.R.Carter) L.Bukhtiyarova in Bukhtiyarova & Round 1996: 26  
 (as '*P. investians*'). Basionym: ***Achnanthes investians*** J.R. Carter 1966: 445, pl. 1, figs. 23-28.  
 Locality. Tristan da Cunha (Carter 1966).  
**Figure A1.22.**

***Planothidium pericavum*** (J.R.Carter) Lange-Bertalot 1999: 278.  
 Basionym: ***Achnanthes pericava*** J.R. Carter 1966: 447, pl. 1, figs. 5-8.  
 Locality. Tristan da Cunha (Carter 1966).  
**Figures A1.21.**

*Amphora incurvata* J.R. Carter 1966: 448, pl. 3, figs. 29-31.

Locality. Tristan da Cunha (Carter 1966).

**Figure A1.30.** SEM photo from Carter's original material.

*Eunotia ambigua* J.R. Carter 1966: 451, pl. 2, figs. 1 1-15, 20.

Locality. Tristan da Cunha (Carter 1966).

**Figure A1.1.** Upper LM pictures present what has been identified as this species and below, the three valves which were identified as *E. cf. ambigua*.

*Eunotia diverta* J.R. Carter 1966: 452, pl. 9, figs. 6-7.

Locality. Gough Island (Carter 1966).

**Figures A1.6, A1.25–A1.26.**

*Eunotia morbida* J.R. Carter 1966: 453, pl. 9, figs. 23-24.

Locality. Gough Island (Carter 1966).

**Figure A1.3, A1.23–A1.24.**

*Eunotia johncarteri* D.M. Williams in D.M. Williams & G. Reid 2002: 142 (as '*E. johncarterii*')

Heterotypic synonym: *Eunotia pectinoides* J.R. Carter 1966: 454, pl. 9, figs. 4-5, 22.

Locality. Gough Island (Carter 1966).

**Figure A1.4.**

Note. Two forms of *E. johncarteri* have been identified and separated during the analysis of samples from Gough. However, in sample 2000-195 both of them, *E. johncarteri* forma thin and *E. johncarteri* forma thick, have been found together quite inseparably, which implies that it might be one species with a wider morphological variability (phenotypic plasticity).

*Gomphonema spatulum* J.R. Carter 1966: 456, pl. 6, fig. 12, pl. 7, fig. 23.

Locality. Tristan da Cunha (Carter 1966).

**Figures A1.2, A1.27–A1.28.**

*Melosira johncarteri* D.M. Williams 2001: 143.

Heterotypic synonym: *Melosira setosa* J.R. Carter 1966: 458, pl. 9, figs. 12-14.

Locality. Gough Island (Carter 1966).

**Figure A1.5.** Very rarely seen.

*Navicula decissa* J.R. Carter 1966: 461, pl. 3, fig 11.

Locality. Tristan da Cunha (Carter 1966).

**Figure A1.14.**

***Navicula friska*** J.R. Carter 1966: 462, pl. 3, figs. 7-10.

Locality. Tristan da Cunha (Carter 1966).

**Figure A1.7, A1.29.**

Note. *Navicula friska* is most probably the same as species identified under *Chamaepinnularia* sp2. Absence of gaps in the middle valve face area firstly refused pairing of these two, but it seems that the species might possess both features. Also the identity of *Ch.* sp1 in comparison to *Ch.* sp2 and the original species of Carter should be made prior the formal change.

***Humidophila nienta*** (Carter) Lowe, Kociolek, Johansen, Van de Vijver, Lange-Bertalot &

Kopalová 2014: 353. Basionym: ***Navicula nienta*** J.R. Carter 1966: 464, pl. 3, fig. 17.

Heterotypic Synonyms: *Diadasmus langebertalotii* Le Cohu & Van de Vijver 2002; *Diadasmus pseudolangebertalotii* Metzeltin, Lange-Bertalot & García-Rodríguez 2005

Locality. Tristan da Cunha (Carter 1966).

**Figure A1.8, A1.31.**

***Nitzschia navia*** J.R. Carter 1966: 468, pl. 7, figs 1, 7.

Locality. Tristan da Cunha (Carter 1966).

**Figure A1.11.**

Note. Identified as 'cf. *navia*' due to only solely observed valves.

***Nitzschia oscilla*** J.R. Carter 1966: 468, pl. 7, figs. 21-22.

Locality. Tristan da Cunha (Carter 1966).

**Figure A1.10.**

Note. Identified as 'cf. *oscilla*' due to only solely observed valves.

***Nitzschia petulla*** J.R. Carter 1966: 469, pl. 6, figs. 18-23.

Locality. Tristan da Cunha (Carter 1966).

***Nitzschia sonora*** J.R. Carter 1966: 470, pl. 8, figs. 21-22.

Locality. Gough Island (Carter 1966).

Note. Identified as 'cf. *sonora*' due to only solely observed valves.

***Nitzschia terrestris*** (J.B.Petersen) Hustedt 1934: 396.

Erroneously described as new, ***Nitzschia rotula*** J.R. Carter 1966: 470, pl. 7, figs. 19, 26.

Locality. Tristan da Cunha (Carter 1966).

**Figure A1.12.**

***Pinnularia johncarteri*** (Carter) D.M.Williams 2001: 149.

Heterotypic synonym: ***Pinnularia dispersa*** J.R. Carter 1966: 472, pl. 5, figs. 7-9.

Locality. Tristan da Cunha and Gough Island rarely (Carter 1966).

**Figures A1.13, A1.32–A1.33.**

*Pinnularia posita* J.R. Carter 1966: 475, pl. 8, figs. 7-8.

Locality. Gough Island (Carter 1966).

**Figures A1.15, A1.35.**

Note. Abundant species, the size range varies in assemblage (grouped photos in **Figure A1.15**).

*Pinnularia restituta* J.R. Carter 1966: 475, pl. 8, figs 3-5.

Locality. Gough Island (Carter 1966).

**Figures A1.18, A1.34.**

Note. Very abundant and apparently quite morphologically variable species.

*Pinnularia seriata* J.R. Carter 1966: 476, pl. 8, fig. 12.

Locality. Gough Island (Carter 1966).

*Pinnularia sistassa* J.R. Carter 1966: 477, pl. 5, figs. 3-6.

Locality. Tristan da Cunha (Carter 1966).

**Figures A1.17, A1.36–A1.37.**

Note. Abundant species, costae may or may not be present along the entire valve (**Figure A1.17**).

*Pinnularia vapilla* J.R. Carter 1966: 478, pl. 4, figs. 1-2.

Locality. Tristan da Cunha (Carter 1966).

**Figure A1.20.**

*Eunotia linearis* (Carter) Vinsová, Kopalová & Van de Vijver (2016: 122).

Basionym/Homotypic synonym: *Pseudoeunotia linearis* J.R. Carter 1966: 479, pl. 9, figs. 15-19.

Locality. Gough Island (Carter 1966).

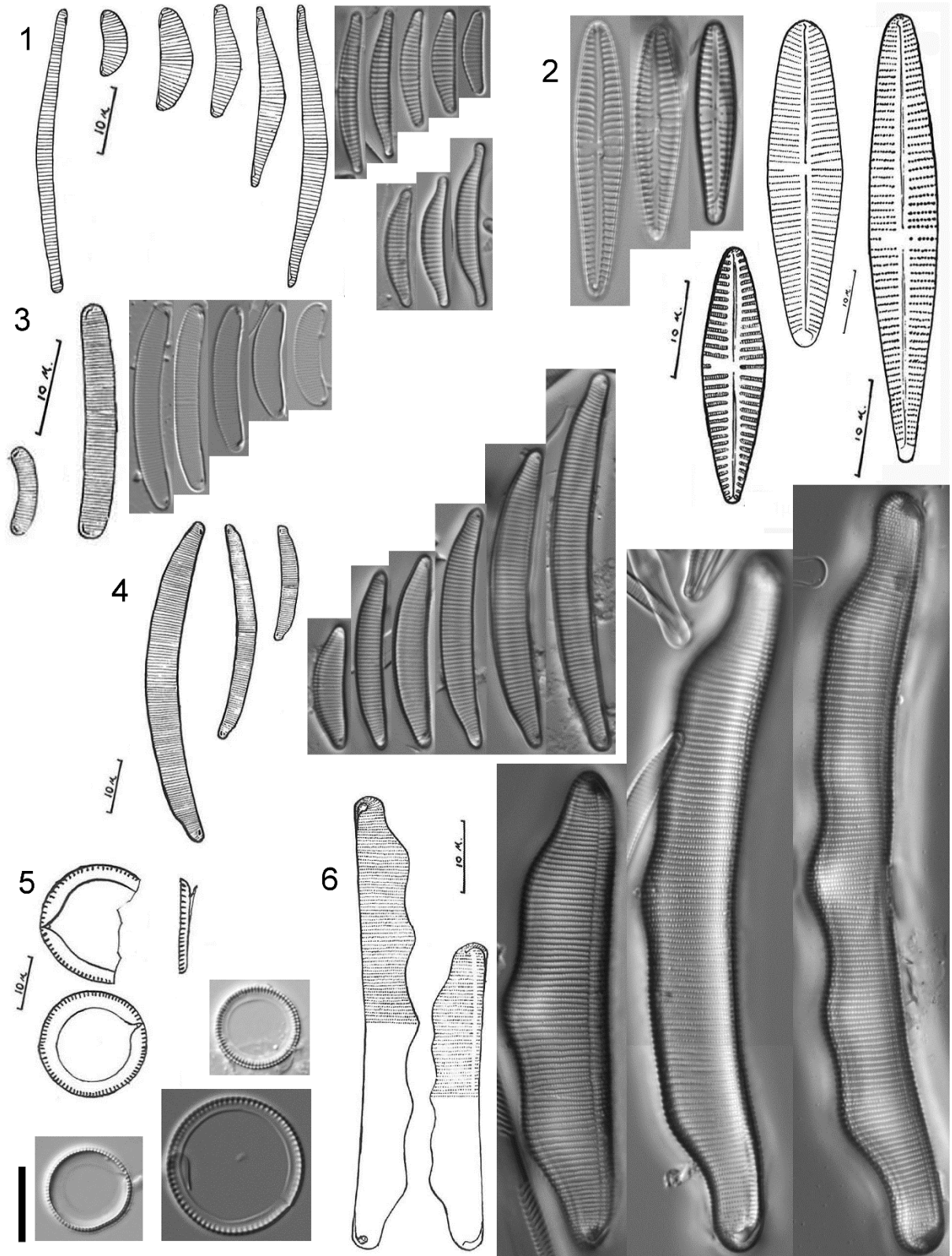
**Figures** can be found in **Chapter 2** of the thesis.

*Pseudoeunotia duplex* J.R. Carter 1966: 479, pl. 9, fig. 29.

Locality. Gough Island (Carter 1966).

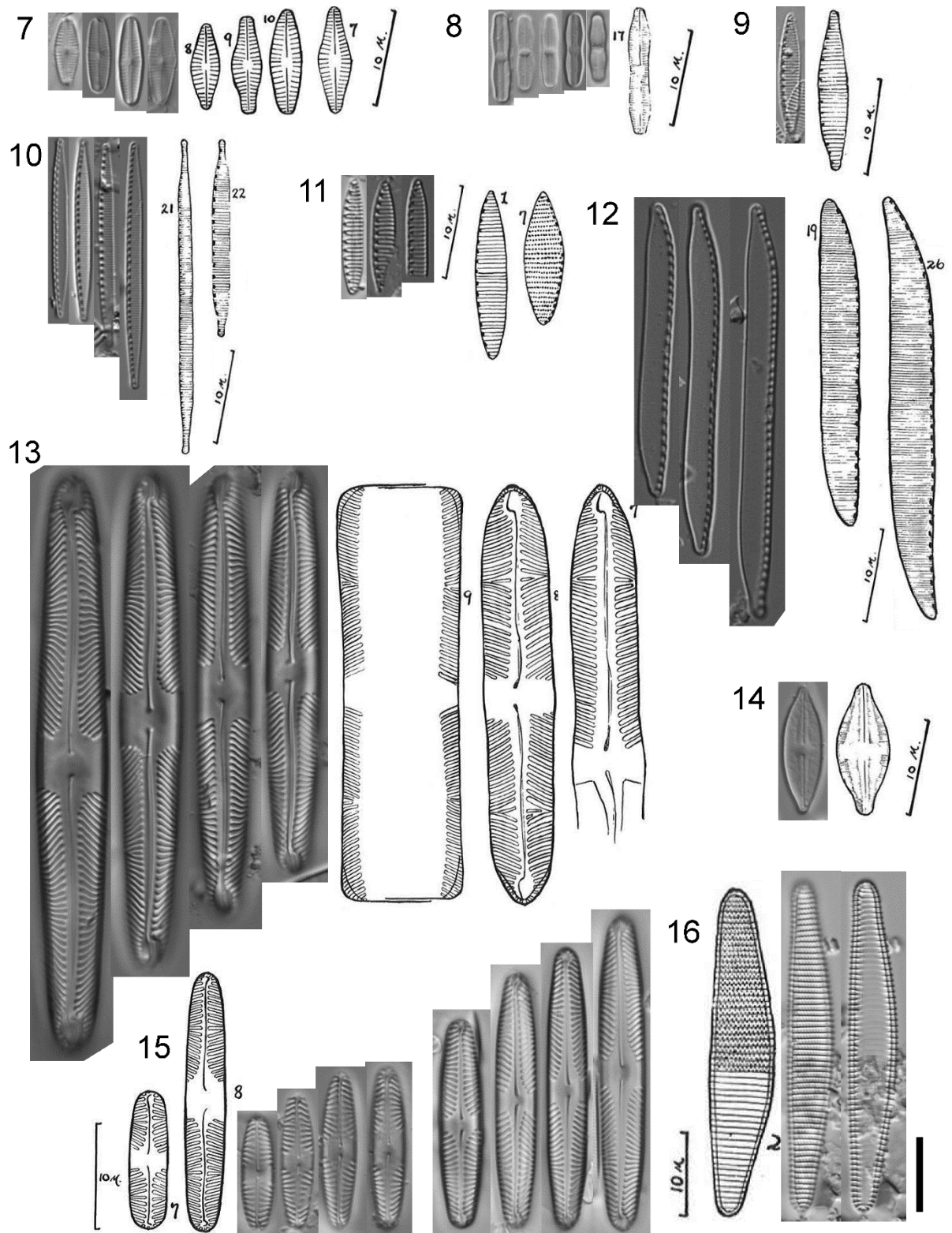
**Figure A1.16.** LM photos of one valve under different focus.

Note. Marine species. Only one entire valve was found, and numerous fragments which suggests that the species is not native to moss habitats.

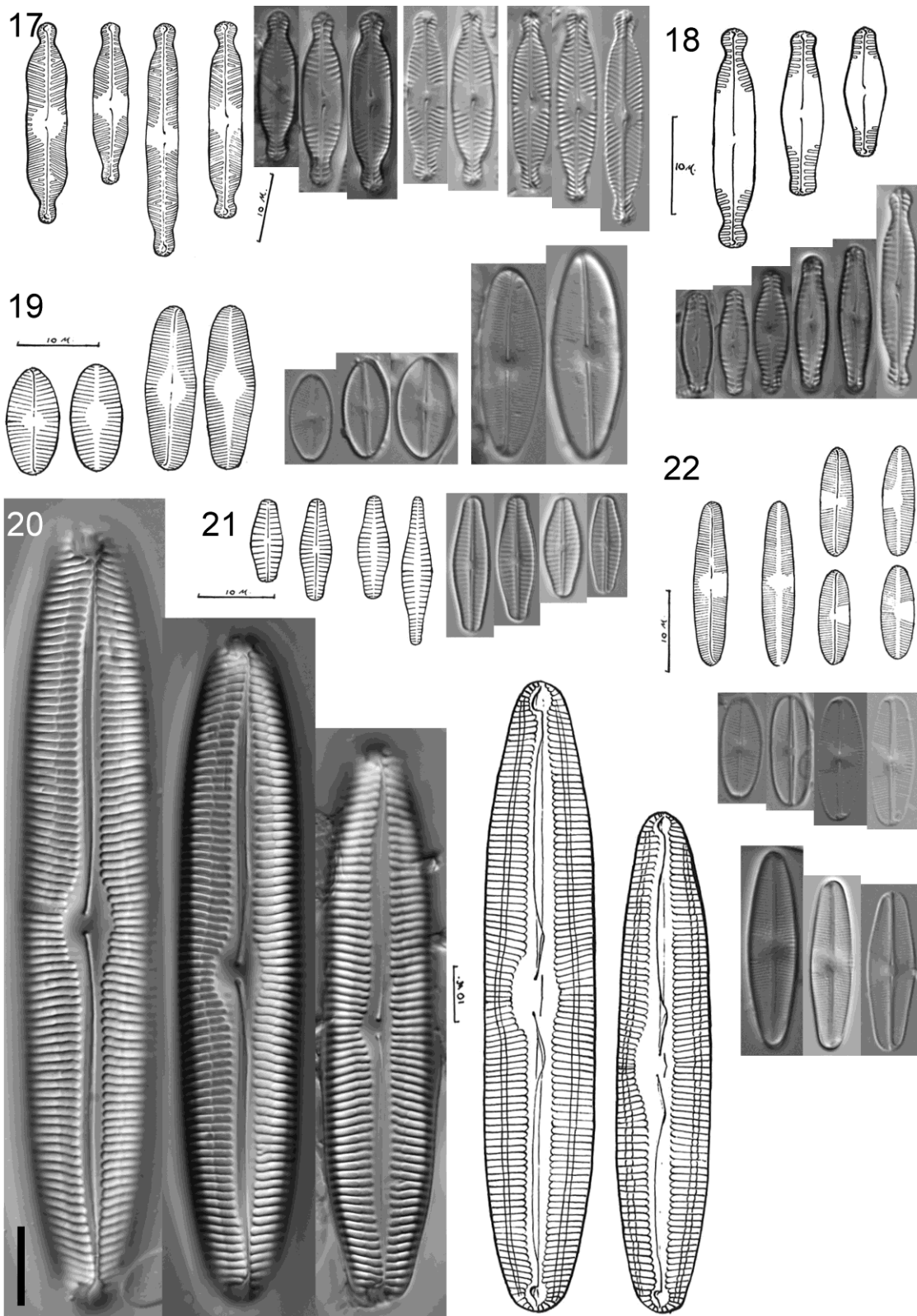


Figures A1.1–A1.6. (1) *Eunotia ambigua*, (2) *Gomphonema spatulum*, (3) *Eunotia morbida*, (4) *Eunotia johncarteri*, (5) *Melosira johncarteri*, (6) *Eunotia diverta*. Scale bars represent 10  $\mu\text{m}$ , drawn ones correspond to each drawing.

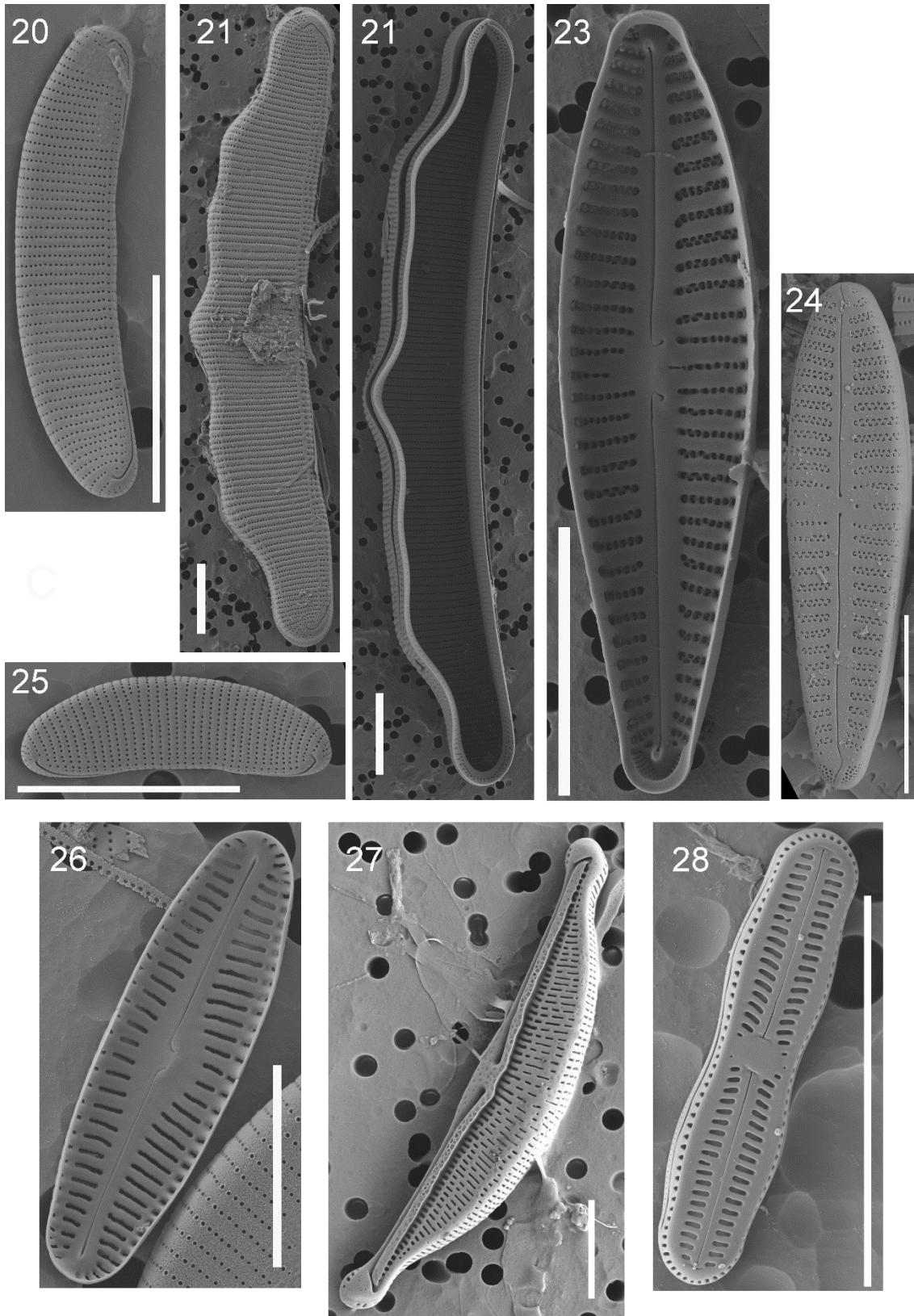




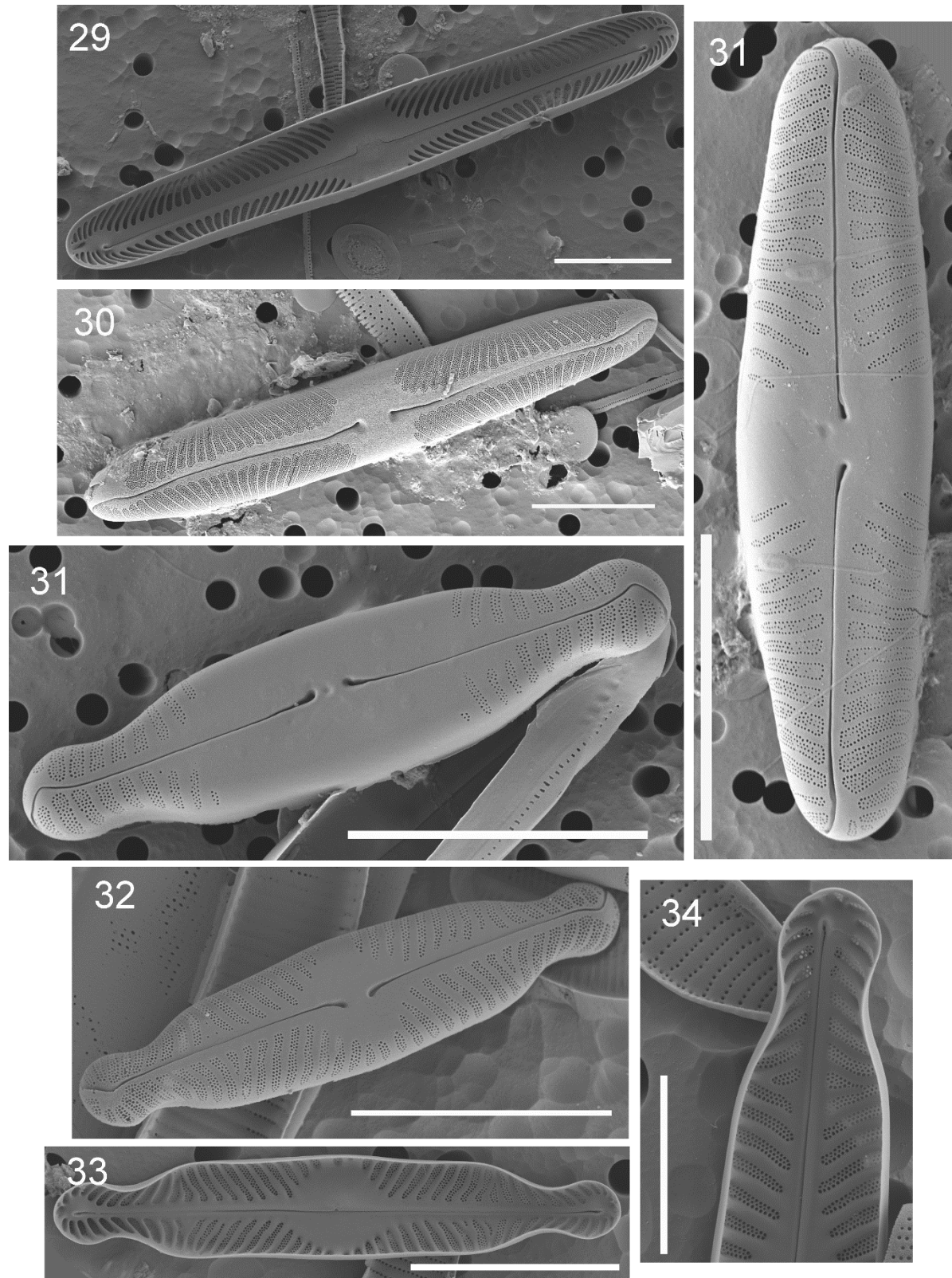
**Figures A1.7–A1.16.** (7) *Chamaepinnularia frisca*, (8) *Humidophila nienta*, (9) *Nitzschia pettula*, (10) *Nitzschia oscilla*, (11) *Nitzschia navia*, (12) *Nitzschia terrestris*, (13) *Pinnularia johncarteri*, (14) *Navicula decissa*, (15) *Pinnularia posita*, (16) *Pseudoeunotia duplex*. Scale bars represent 10  $\mu$ m, drawn ones correspond to each drawing.



Figures A1.17–A1.22. (17) *Pinnularia sistassa*, (18) *Pinnularia restituta*, (19) *Psammothidium atalanta*, (20) *Pinnularia vapilla*, (21) *Planothidium pericavum*, (22) *Psammothidium investians*. Scale bars represent 10 μm, drawn ones correspond to each drawing.



Figures A1.23–A1.31. (23–24) *Eunotia morbida*, (25–26) *Eunotia diverta*, (27–28) *Gomphonema spatulum*, (29) *Chamaepinnularia frigida*, (30) *Halamphora incurvata*, (31) *Humidophila nienta*. Scale bars represent 10  $\mu\text{m}$ .



Figures A1.33–A1.38. (32–33) *Pinnularia johncarteri*, (34) *Pinnularia restituta*, (35) *Pinnularia posita*, (36–37) *Pinnularia sistassa*, (38) *Psammothidium atalanta*. Scale bars represent 10  $\mu\text{m}$ .

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## APPENDIX 2

List of species identified from the moss material of Gough Island

APPENDIX 2. Taxonomic list of all diatom taxa with authorities, observed in the present study. Taxa unidentified to the species level are given a proper provisional code.

TAXON NAME	Abbreviations	If only outside of the counts
<i>Achnanthes coarctata</i> (Brébisson ex W.Smith) Grunow in Cleve & Grunow		×
<i>Achnanthes muelleri</i> G.W.F.Carlson		×
<i>Achnanthes okamuræ</i> Skvortzov	Ach	
<i>Achnanthes</i> sp		×
<i>Achnanthidium minutissimum</i> (Kütz.) Czarnecki	Achn	
<i>Achnanthidium</i> cf. <i>modestiforme</i> (Lange-Bertalot) Van de Vijver	Achn1	
<i>Adlafia bryophila</i> (J.B.Petersen) Lange-Bertalot	Adl	
<i>Amphora incurvata</i> J.R.Carter		×
<i>Aulacoseira ambigua</i> (Grunow) Simonsen	Aul	
<i>Brachysira</i> sp	Bra	
<i>Caloneis</i> cf. <i>bacillum</i> (Grunow) Cleve	Cal	
<i>Caloneis</i> sp1	Cal1	
<i>Caloneis</i> sp2	Cal2	
<i>Cocconeis placentula</i> Ehrenberg	Coc	
<i>Chamaepinnularia</i> cf. <i>begeri</i> (Krasske) Lange-Bertalot	Cha	
<i>Chamaepinnularia evanida</i> (Hustedt) Lange-Bertalot	Cha1	
<i>Chamaepinnularia</i> sp1	Cha2	
<i>Chamaepinnularia</i> sp2 [ <i>Navicula frisca</i> Carter]	Cha3	
<i>Craticula vixneglidenda</i> Lange-Bertalot	Cra	
<i>Cymbella gracilis</i> (Rabenhorst) Cleve	Cym	
<i>Denticula</i> sp	Den	
<i>Distrionella germainii</i> (Reichardt & Lange-Bertalot) Morales, Bahls & Cody	Dis	
<i>Distrionella germainii</i> f. <i>acostata</i> Lange-Bertalot & Rumrich	Dis1	
<i>Distrionella husvikensis</i> (Van de Vijver, Denys & Beyens) Morales, Bahls & Cody	Dis2	
<i>Encyonema</i> sp	Enc	
<i>Encyonopsis</i> sp	Encs	
<i>Eunotia ambigua</i> J.R.Carter	Eun	
<i>Eunotia</i> cf. <i>ambigua</i> J.R.Carter [spX]	Eun1	
<i>Eunotia diverta</i> J.R.Carter	Eun2	
<i>Eunotia elegans</i> Østrup	Eun3	
<i>Eunotia</i> cf. <i>fallax</i> A.Cleve	Eun4	
<i>Eunotia</i> cf. <i>Iecohui</i> Van de Vijver, Beyens & Lebouvier	Eun5	
<i>Eunotia linearis</i> (Carter) Vinsová, Kopalová & Van de Vijver	Eun6	
<i>Eunotia morbida</i> J.R.Carter	Eun7	
<i>Eunotia</i> cf. <i>naegelii</i> Migula	Eun8	
<i>Eunotia paludosa</i> var. <i>paludosa</i> Grunow group	Eun9	
<i>Eunotia</i> aff. <i>papilio</i> (Ehrenberg) Grunow		×
<i>Eunotia johncarteri</i> (Carter) D.M.Williams [forma <i>thick</i> ]	Eun10	
<i>Eunotia johncarteri</i> (Carter) D.M.Williams [forma <i>thin</i> ]	Eun11	
<i>Eunotia pseudoparalleloides</i> (Grunow) Nörpel-Schempp & Lange-Bertalot	Eun12	
<i>Eunotia</i> sp2	Eun13	
<i>Eunotia</i> sp4	Eun14	
<i>Eunotia</i> sp5	Eun15	
<i>Eunotia</i> cf. <i>superbidens</i> Lange-Bertalot		×
<i>Fragilaria construens</i> (Ehrenberg) Grunow forma s.l. Carter	Fra	
<i>Fragilaria</i> cf. <i>parva</i> (Grunow) A.Tuji & D.M.Williams	Fra1	
<i>Frustulia crassinervia</i> (Brébisson ex W.Smith) Lange-Bertalot & Krammer	Fru	
<i>Frustulia saxonica</i> Rabenhorst	Fru1	
<i>Frustulia</i> spE	Fru2	

<i>Frustulia</i> spH	Fru3	
<i>Frustulia</i> spL	Fru4	
<i>Frustulia</i> spN	Fru5	
<i>Frustulia</i> spP	Fru6	
<i>Frustulia</i> spV	Fru7	
<i>Gomphonema coronatum</i> Ehrenberg	Gom	
<i>Gomphonema</i> cf. <i>longiceps</i> Ehrenberg	Gom1	
<i>Gomphonema longiceps</i> var. <i>subclavata</i> Grunow s.l. Carter	Gom2	
<i>Gomphonema parvulum</i> (Kützing) Kützing group	Gom3	
<i>Gomphonema parvulum</i> var. <i>lagenula</i> (Kützing) Frenguelli	Gom4	
<i>Gomphonema</i> cf. <i>spatulum</i> J.R.Carter	Gom5	
<i>Gomphonema</i> sp3		×
<i>Halamphora</i> sp	Hal	
<i>Hantzschia abundans</i> Lange-Bertalot		×
<i>Hantzschia amphioxys</i> (Ehrenberg) Grunow	Han	
<i>Humidophila arcuata</i> (Lange-Bertalot) Lowe, Kociolek, Johansen, Van de Vijver, Lange-Bertalot & Kopalová	Hum	
<i>Humidophila</i> cf. <i>australis</i> (Van de Vijver & Sabbe) Lowe, Kociolek, Johansen, Van de Vijver, Lange-Bertalot & Kopalová		×
<i>Humidophila contenta</i> (Grunow) Lowe, Kociolek, J.R.Johansen, Van de Vijver, Lange-Bertalot & Kopalová	Hum1	
<i>Humidophila</i> cf. <i>ingae</i> (Van de Vijver ) Lowe, Kociolek, Johansen, Van de Vijver, Lange-Bertalot & Kopalová	Hum2	
<i>Humidophila nienta</i> (Carter) Lowe, Kociolek, Johansen, Van de Vijver, Lange-Bertalot & Kopalová	Hum3	
<i>Humidophila</i> spE	Hum4	
<i>Humidophila</i> sp177	Hum5	
<i>Humidophila</i> sp2	Hum6	
<i>Karayevia oblongella</i> (Østrup) Aboal	Kar	
<i>Luticola</i> cf. <i>beyensii</i> Van de Vijver, Ledeganck & Lebouvier	Lut	
<i>Luticola</i> sp1	Lut1	
<i>Luticola</i> sp2	Lut2	
<i>Luticola</i> sp3	Lut3	
<i>Mayamaea atomus</i> (Kützing) Lange-Bertalot	May	
<i>Mayamaea atomus</i> var. <i>permitis</i> (Hustedt) Lange-Bertalot	May1	
<i>Melosira</i> cf. <i>echinata</i> Manguin	Mel	
<i>Melosira jonhcarteri</i> (Carter) D.M.Williams	Mel1	
<i>Melosira varians</i> C.Agardh	Mel2	
<i>Melosira</i> spX	Mel3	
<i>Melosira</i> spY	Mel4	
<i>Meridion circulare</i> (Greville) C.Agardh	Mer	
<i>Navicula arvensis</i> Hustedt	Nav	
<i>Navicula bicephala</i> Hustedt	Nav1	
<i>Navicula cryptotenelloides</i> Lange-Bertalot	Nav2	
<i>Navicula decissa</i> J.R.Carter		×
<i>Navicula exilis</i> Kützing	Nav3	
<i>Navicula gregaria</i> Donkin	Nav4	
<i>Navicula lapidosa</i> Krasske	Nav5	
<i>Navicula</i> aff. <i>lapidosa</i> Krasske	Nav9	
<i>Navicula pseudoventralis</i> Hustedt	Nav6	
<i>Navicula</i> cf. <i>radiosa</i> Kützing	Nav7	
<i>Navicula veneta</i> Kützing	Nav8	
<i>Navicula vilaplani</i> (Lange-Bertalot & Sabater) Lange-Bertalot & Sabater		×
<i>Neidium</i> sp	Nei	



<i>Nitzschia debilis</i> (Arnott ex O'Meara) Grunow in Cleve & Grunow	Nit	
<i>Nitzschia</i> cf. <i>navia</i> J.R.Carter	Nit1	
<i>Nitzschia</i> cf. <i>oscilla</i> J.R.Carter	Nit2	
<i>Nitzschia palea</i> (Kützing) W.Smith	Nit3	
<i>Nitzschia perspicua</i> Cholnoky	Nit4	
<i>Nitzschia petulla</i> J.R.Carter	Nit5	
<i>Nitzschia</i> cf. <i>sonora</i> J.R.Carter	Nit6	
<i>Nitzschia terrestris</i> (J.B.Petersen) Hustedt	Nit8	
<i>Nitzschia</i> sp675	Nit9	
<i>Nupela</i> cf. <i>acaciensis</i> Vouilloud & Sala		×
<i>Orthoseira gremenii</i> Van de Vijver & Kopalová	Ort	
<i>Orthoseira roseana</i> (Rabenhorst) O'Meara	Ort1	
<i>Pinnularia austroshetlandica</i> (Carlson) A.Cleve	Pin	
<i>Pinnularia borealis</i> Ehrenberg	Pin1	
<i>Pinnularia borealis</i> var. 65A	Pin2	
<i>Pinnularia borealis</i> var. <i>scalaris</i> (Ehrenberg) Rabenhorst	Pin3	
<i>Pinnularia</i> cf. <i>johnccarteri</i> (Carter) D.M.Williams	Pin4	
<i>Pinnularia</i> cf. <i>laucensis</i> Lange-Bertalot	Pin5	
<i>Pinnularia divergentissima</i> (Grunow) Cleve forma <i>elongata</i> A.Cl.		×
<i>Pinnularia lindanedbalovae</i> Van de Vijver & Moravcová	Pin6	
<i>Pinnularia</i> cf. <i>microstauron</i> (Ehrenberg) Cleve s.l. Carter	Pin7	
<i>Pinnularia</i> cf. <i>neomajor</i> Krammer		×
<i>Pinnularia perirrorata</i> Krammer	Pin8	
<i>Pinnularia posita</i> J.R.Carter	Pin9	
<i>Pinnularia restituta</i> J.R.Carter	Pin10	
<i>Pinnularia seriata</i> J.R.Carter		×
<i>Pinnularia sistassa</i> J.R.Carter	Pin11	
<i>Pinnularia</i> cf. <i>shoenfelderi</i> Krammer	Pin12	
<i>Pinnularia</i> sp11	Pin13	
<i>Pinnularia</i> spO	Pin14	
<i>Pinnularia subacoricola</i> Metzeltin, Lange-Bertalot & García-Rodríguez		×
<i>Pinnularia subantarctica</i> var. <i>elongata</i> (Manguin) Van de Vijver & Le Cohu		×
<i>Pinnularia vapilla</i> J.R.Carter	Pin15	
<i>Pinnularia viridis</i> (Nitzsch) Ehrenberg	Pin16	
<i>Planothidium cyclophorum</i> (Heiden) Van de Vijver	Pla	
<i>Planothidium</i> cf. <i>densistriatum</i> Van de Vijver & Beyens	Pla1	
<i>Planothidium frequentissimum</i> (Lange-Bertalot) Lange-Bertalot	Pla2	
<i>Planothidium pericavum</i> (J.R.Carter) Lange-Bertalot	Ach1	
<i>Psammothidium abundans</i> (Manguin) Bukhtiyarova & Round	Psa	
<i>Psammothidium atalanta</i> J.R.Carter	Psa1	
<i>Psammothidium investians</i> J.R.Carter	Psa2	
<i>Pseudoeunotia duplex</i> J.R.Carter	Pse	
<i>Rhopalodia rupestris</i> (W.Smith) Krammer	Rho	
<i>Sellaphora atomoides</i> (Grunow) C.E. Wetzel & Van de Vijver	Sel	
<i>Sellaphora saugerresii</i> (Desm.) C.E. Wetzel & D.G. Mann	Sel1	
<i>Stauroneis</i> sp	Sta	
<i>Staurosira venter</i> (Ehrenberg) Cleve & J.D.Möller	Sta1	
<i>Staurosira</i> sp1	Sta2	
<i>Surirella</i> sp1	Sur	
<i>Tabellaria flocculosa</i> (Roth) Kützing	Tab	
<i>Thalassiosira</i> sp	Tha	

## APPENDIX 3

### Abstracts

THE GENUS *FRUSTULIA* ON GOUGH ISLAND (TRISTAN DA CUNHA ARCHIPELAGO)

Vinšová, Petra<sup>1\*</sup>, Kopalová, Kateřina<sup>1</sup>, Gremmen, Niek J.M.<sup>2</sup>, de Haan, Myriam<sup>3</sup>  
& Van de Vijver, Bart<sup>3,4</sup>

<sup>1</sup>Charles University in Prague, Faculty of Science, Department of Ecology, Viničná 7, CZ-12844  
Prague 2, Czech Republic, \*Petra Vinšová: [vinsova@gmail.com](mailto:vinsova@gmail.com)

<sup>2</sup>Data-Analyse Ecologie, Hesselsstraat 11, 7981 CD Diever, The Netherlands

<sup>3</sup>Botanic Garden Meise, Department of Bryophyta & Thallophyta, Nieuwelaan 38, B-1860  
Belgium

<sup>4</sup>University of Antwerp, Department of Biology-ECOBE, Universiteitsplein 1, B-2610 Wilrijk,  
Belgium

Gough Island (40°21' S, 9°53' W), part of the Tristan da Cunha archipelago, is a small, uninhabited island, situated in the cool-temperate zone of the southern Atlantic Ocean. Large parts of the island are quite mountainous with most of the island above 400 m. This volcanic island is typically oceanic and has never been part of a continental landmass. The climate is cool and wet, with a mean annual temperature (at sea level) of 11° C, a mean annual precipitation in excess of 3000 mm and with frequent gale-force winds.

Despite its very interesting geographical position, its oceanic origin and the number of interesting habitats, the non-marine diatom flora of Gough Island is only poorly known. Carter published in 1966 on the diatom flora from the Tristan da Cunha Island group, reporting the presence of a very diverse diatom flora with a large number of new taxa that so far have been rarely observed elsewhere. Recently, a new survey of the moss-inhabiting diatoms on Gough Island has started. During this survey, several taxa were discovered belonging to the genera *Pinnularia*, *Eunotia* and *Frustulia*, that could not be identified using the currently available literature.

The poster illustrates and discusses the diversity within the genus *Frustulia*. Apart from the presumably cosmopolitan species *F. vulgaris* (Thwaites) De Toni, six unknown *Frustulia* taxa were observed. Based on detailed LM and SEM observations, it is highly likely that these six taxa need to be described as new species. They differ from each other and from already described taxa by a series of morphological features such as the overall valve outline, the shape and structure of the helictoglossa, the structure of the central area and the external proximal and distal raphe endings. All differentiating characters are highlighted on the poster. Brief notes on the ecology of the six taxa are added.

References: Carter, J. (1966) Some freshwater diatoms of Tristan da Cunha and Gough Island. *Nova Hedwigia*, 9: 443-492.

A NEW INTERESTING SPECIES OF *EUNOTIA* (BACILLARIOPHYTA) FROM GOUGH ISLAND  
(TRISTAN DA CUNHA ARCHIPELAGO)

Petra Vinšová<sup>1\*</sup>, Kateřina Kopalová<sup>1</sup>, Bart Van de Vijver<sup>2,3</sup>.

<sup>1</sup> Charles University in Prague, Faculty of Science, Department of Ecology, Viničná 7, CZ-12844 Prague 2, Czech Republic, <sup>2</sup>Botanic Garden Meise, Department of Bryophyta & Thallophyta, Nieuwelaan 38, B-1860 Belgium, <sup>3</sup>University of Antwerp, Department of Biology-ECOB, Universiteitsplein 1, B-2610 Wilrijk, Belgium.

\* Petra Vinšová, [vinsova@gmail.com](mailto:vinsova@gmail.com)

Gough Island (40°21' S, 9°53' W), is a small remote uninhabited island of a volcanic origin, positioned in southern Atlantic Ocean and some 350 km south of the rest islands from Tristan da Cunha archipelago. Despite its very interesting geographical position, its oceanic origin, an isolation and the number of interesting habitats, the non-marine diatom flora of Gough Island is only poorly known. Carter described in 1966 numerous of new diatom species from Tristan da Cunha Island group, reporting the presence of a very diverse diatom flora with a large number of new taxa that so far have been only rarely observed elsewhere. Recently, a new survey of moss-inhabiting diatom flora on Gough Island has started and so far, several taxa were discovered belonging to the genera *Eunotia*, *Frustulia* and *Pinnularia*, that could not be identified using the currently available literature.

One species belonging to the genera *Eunotia* is of our particular interest. This species, which Carter (1966) newly described as *Pseudoeunotia linearis*, has some morphological features that clearly differs it from the other species of the same genera known so far. Based on a detailed LM and SEM observation, we can now present a complex description of this highly interesting species that differs morphologically (i.e., the overall valve shape, the presence of spines on sides, areolae and striae density and their structure based on the position, as well as the absence of a terminal nodules). All differentiating characteristics are highlighted on the poster. The comparison and the unification with the same species found in original material of Carter (1966) is added and also the correction of the species name is suggested. Notes on the ecology are also added.

KEYWORDS: GOUGH ISLAND, DIATOMS, EUNOTIA

REFERENCES: Carter, J. (1966): "Some freshwater diatoms of Tristan da Cunha and Gough Island". Nova Hedwigia 9: 443-492.

## APPENDIX 4

Diatoms in cryoconite holes and adjacent proglacial freshwater sediments,  
Nordenskiöld glacier (Spitsbergen, High Arctic)

## DIATOMS IN CRYOCONITE HOLES AND ADJACENT PROGLACIAL FRESHWATER SEDIMENTS, NORDENSKIÖLD GLACIER (SPITSBERGEN, HIGH ARCTIC).

Petra Vinšová<sup>1\*</sup>, Eveline Pinseel<sup>2,3,4</sup>, Tyler J. Kohler<sup>1</sup>, Bart Van de Vijver<sup>3,4</sup>, Jakub D. Žárský<sup>1</sup>, Jan Kavan<sup>5</sup>, Kateřina Kopalová<sup>1,5,6</sup>

<sup>1</sup>Charles University in Prague, Faculty of Science, Department of Ecology, Viničná 7, CZ-128 44 Prague 2, Czech Republic; <sup>2</sup>Ghent University, Faculty of Science, Department of Biology, Protistology & Aquatic Ecology (PAE), Krijgslaan 281-S8, BE-9000 Ghent, Belgium; <sup>3</sup>Botanic Garden Meise, Department Bryophyta & Thallophyta, Nieuwelaan 38, BE-1860 Meise, Belgium; <sup>4</sup>University of Antwerp, Faculty of Science, Department of Biology, Ecosystem Management Research Group (ECOBE), Universiteitsplein 1, BE-2610 Wilrijk, Belgium; <sup>5</sup>University of South Bohemia, Faculty of Science, Centre for Polar Ecology (CPE), Branišovská 31, CZ-370 05 České Budějovice, Czech Republic; <sup>6</sup>Academy of Science of the Czech Republic, Institute of Botany, Section of Plant Ecology, Dukelská 135, 379 82 Třeboň, Czech Republic

### ABSTRACT

Cryoconite holes are small, extreme habitats, widespread in the ablation zones of glaciers worldwide. They can provide a suitable environment for microorganisms including bacteria, cyanobacteria, algae, fungi, and invertebrates. Diatoms have been previously recovered from cryoconite holes of Greenland and of Svalbard, and recent findings from Antarctica suggest that cryoconite holes may harbor a unique diatom flora distinct from other aquatic habitats nearby. In the present study, we characterize the diatom communities of Nordenskiöld glacier cryoconite holes in Billefjorden (Svalbard, Spitsbergen), and multivariate approaches were used to compare them with three freshwater localities in the immediate vicinity to investigate possible sources of the species pool. We found cryoconite holes to have similar or greater average genus-richness than adjacent lake/ponds habitats, even though lower numbers of valves were recovered. Overall, cryoconite hole diatom communities differed significantly from those observed in lakes, suggesting that other sources actively contribute to these communities than nearby lakes alone. This further suggests that (i) diatoms present in cryoconite might not exclusively originate from aquatic habitats, but also from (semi-)

terrestrial ones; and (ii) that a much wider area than the immediate surroundings should be considered as a possible source for cryoconite diatom flora.

#### KEYWORDS

Ecology, limnology, lakes, cryosphere, Svalbard, polar region

## INTRODUCTION

Glaciers and ice sheets cover almost 10% of the Earth's landmass (Clark 2009), and as a result, their suitability for life is of great importance. Cryoconite holes represent microhabitats formed by dust transported into the ablation zones of the glacial surface, which leaves behind small water-filled depressions during thawing periods. These supraglacial habitats are found worldwide and can persist for days to decades (Hodson et al. 2008). Since the 1930's, cryoconite holes have been recognized as a micro-environment harboring life (Steinbock 1936), and modern investigations have shown that they support a range of micro-organisms including archaea, heterotrophic bacteria, fungi, microalgae, filamentous cyanobacteria, nematodes, rotifers and tardigrades (*e.g.* Wharton et al. 1981 and 1985, Mueller et al. 2001, Edwards et al. 2013a). In contrast to the surrounding glacier surface, cryoconite holes supply liquid water to organisms, can shelter microorganisms from UV light, and are presumably also less susceptible to temperature extremes. Biovectors and wind are thought to serve as dispersal agents for microbial propagules to the glacier surface. Both local and long range aeolian input (Šabacká et al. 2012, Budgeon et al. 2012), as well as debris from avalanches and erosion, can supply the surface with sediment (Hodson et al. 2008) and organic carbon (Stibal et al. 2008).

Antarctic cryoconite holes may be isolated from the atmosphere for multiple melt seasons by a thick ice lid (Fountain et al. 2004), allowing the development of unique habitats that harbor diverse communities (Mueller et al. 2001). In contrast, only a thin ice lid may develop during summer months in the Arctic, resulting in hydrologically connected environments that are

frequently flushed with meltwater, promoting a more homogenous resident community (Edwards et al. 2011). Nevertheless, biological activity of inhabitants (*e.g.* granule formation, darkening processes and photosynthetic activity) remains surprisingly high for such an extreme habitat (Vonnahme 2014), and as known from the Antarctic, cryoconites can serve as refuges for aquatic and terrestrial microorganisms (Foreman et al. 2007, Stanish et al. 2013). Cryoconite communities may also (re-)seed downstream microbial communities residing in proglacial lakes and streams with cells and propagules (Yallop et al. 2010, Stanish et al. 2013) that can stand at the very onset of microbial colonization (Stibal et al. 2006).

Most studies of glacial microbial communities have focused on bacteria or cyanobacteria (Mueller et al. 2001, Christner et al. 2003, Cameron et al. 2012, Edwards et al. 2013b), which are the most abundant primary producers in these habitats (Mueller et al. 2001, Porazinska et al. 2004, Stibal et al. 2006, Stibal et al. 2007). Our recent knowledge of other cryoconite hole phototrophs such as diatoms (Bacillariophyceae) is, however, rather poor. It is in spite of the fact that diatoms are one of the most successful groups of unicellular algae worldwide, inhabiting a wide range of aquatic and terrestrial environments including polar regions (Jones 1996, Van de Vijver et al. 1999, Sabbe et al. 2003, Van de Vijver et al. 2005, Antoniadou et al. 2008, 2009). Furthermore, the species-specific characteristics of their outer silica cell-wall, as well as individual responses to the physico-chemical environment make diatoms excellent bio-indicators in applied sciences such as paleo-ecology and biogeography (Spaulding et al. 2010).

While diatoms have been previously recorded from supraglacial habitats (Mueller et al. 2001, Van de Vijver et al. 2010b, Cameron et al. 2012), speculation persists as to how these communities are assembled. It is possible that diatoms, together with other micro-organisms present in microbial mats, can be delivered to the supraglacial habitats from nearby lakes and streams by winds (Nkem et al. 2006) and solely reflect those in transport. On the other hand, they may constitute independently functioning communities with a composition similar, yet distinct from their surroundings. To our best knowledge, only two diatom-focused studies of



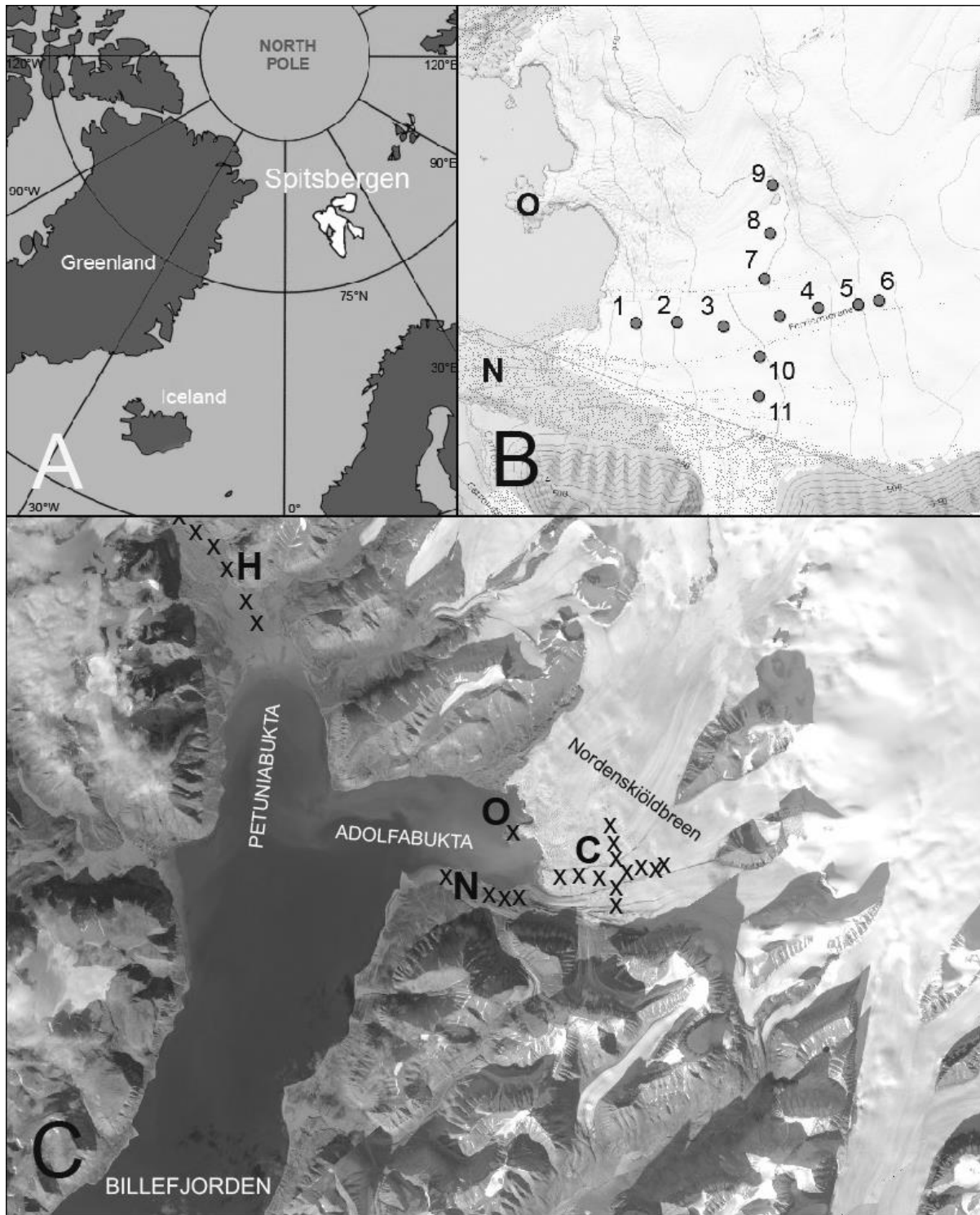
cryoconite holes exist at this time. One relates to the Arctic (Yallop et Anesio 2010), the other one to the Antarctic (Stanish et al. 2013). The former one reports cultured diatoms from cryoconite holes and compares these with database samples. The latter one compares cryoconite hole communities with microbial mats from adjacent streams. Therefore, much work remains for the diatom-related scientific community in order to gain a better understanding of the structure and function of these extreme habitats.

To address such gap in our knowledge, we characterize diatom communities from cryoconite hole sediments of the Nordenskiöld glacier (Adolfbukta, Billefjorden, Svalbard) and compare these results with adjacent aquatic habitats that could potentially serve as a source. We hypothesize that if cryoconite holes are seeded by these adjacent habitats, supraglacial diatom communities should reflect the diatom communities in surrounding lakes and ponds. An alternative to this hypothesis is the potential importance of other nearby habitats such as terrestrial areas and streams, and/ or more long range transport from other sources. To answer this question, a simple genus-based approach was applied to reduce the effects of taxonomic uncertainty, and the results were analyzed using multivariate approaches to determine the similarity of communities between habitats.

## MATERIAL AND METHODS

### Sample collection

Since 2007, summer research campaigns organized by the Centre for Polar Ecology (University of South Bohemia in České Budějovice, Czech Republic) have annually taken place in Petuniabukta (see reports of the research activities at website CPE: <http://polar.prf.jcu.cz/docs.htm> or <http://polar.prf.jcu.cz/index.htm>) located in the central part of Spitsbergen (Svalbard Archipelago) (see Fig. A4.1). The fjord is surrounded by a lowland area of marine terraces, and steep slopes up to 937 m. Its climate is characterized by low precipitation rates (about 200 mm y<sup>-1</sup>), relatively warm winters (-6.7 to -17°C), and wind speed ranging from 2.8 to 23.6 m s<sup>-1</sup> (at 78° 42' N and 16° 27' E) (Rachlewicz et al. 2007, Láška et al. 2012).



*Figure A4.1. Map of the study sites. Sampling points are indicated by crosses. On the last image, C refers to cryoconite holes, H to Hørbye lakes, N to Nordenskiöld lakes, and O to Retrettøya ponds site.*

During the 2014 boreal summer season, 12 samples from cryoconite holes were collected (Fig. A4.2) along two sampling lines on the Nordenskiöld tidewater glacier (glacier description in Rachlewicz et al. 2007). The first line was situated on an altitudinal gradient from the glacier front upwards, and the second one from the side marginal moraine to the glacier center (Fig. A4.1B and Fig. A4.2). These sites were chosen to study possible patterns in aeolian transport of diatoms from the surrounding environments, which should occur from the sea and/or from the marginal zones towards the glacier surface. Sediment samples from small cryoconite holes (3 – 5 cm in diameter, 10 – 15 cm in depth) were collected with pipettes with enlarged openings, stored in 25 mL tubes, and preserved with 96% ethanol. A hand-held GPS was used to determine altitude and location, and pH, temperature and conductivity were measured in the field using a HANNA Instrument HI 98129 Waterproof pH/Conductivity/ TDS Tester (Hanna Instruments Czech s.r.o., Czech Republic). Additionally, the water temperature of one stable cryoconite hole on Nordenskiöld glacier surface was measured every hour for 28 days between the 22nd of July and 25th of August using a temperature datalogger 'minikin T' (EMS Brno, Czech Republic), revealing stable values between  $-1^{\circ}\text{C}$  and  $1^{\circ}\text{C}$ .

During the 2011 and 2013 summer campaigns, benthic epilithon/epipelon samples from the littoral zones of freshwater lakes and ponds were collected for diatom analysis as described above for cryoconite holes. For a full species report of freshwater diatoms from Petuniabukta, see Pinseel (2014). Three of these localities from Pinseel (2014) (Fig. A4.1C) were selected for comparison with the cryoconite samples. The first locality, Retrettøya (O) (known also as 'roche moutonnée' or 'Oblik'), is a peninsula located in front of the tide-water glacier Nordenskiöld. The area is rather freshly deglaciated, being covered by the Nordenskiöld glacier until about 30 years ago, and therefore open to organismal colonization. Several ponds on this peninsula, situated in eroded tectonic faults later remodeled by glacial erosion (Pinseel 2014), are located in close vicinity of the sea, surrounded by a large colony of Arctic terns, *Sterna paradisaea* Pontoppidan. Altogether, glacial influence, the presence of the Arctic terns colony, and possible sea spray influence make this peninsula a unique locality in terms of nutrient sources. The other two localities are comprised of kettle lakes located in the frontal moraines

of the Nordenskiöld (N) and Hørbye (H) glaciers, the latter being located in the northern part of Petuniabukta. All samples used in this study (both lakes and cryoconite holes), together with their parameters, are listed in Table 1.

### **Slide preparation and enumeration**

For light microscopy analysis, subsamples were cleaned by a modified method described in Van der Werff (1955). The sub-samples were added 37% H<sub>2</sub>O<sub>2</sub> and then heated to 80°C for about 1h. The reaction was completed by addition of saturated KMnO<sub>4</sub>. Following digestion and oxidation, samples were rinsed three times with distilled water alternated with centrifugation (10 minutes at 3500 x g). The cleaned diatom material was diluted with distilled water on microscope cover slides, dried, and mounted in Naphrax®.

In each sub-sample, diatom valves were identified to the lowest taxonomic level possible (taxa with uncertain taxonomic status were indicated with 'cf.' (confer: species identification is uncertain) or 'sp.', and sometimes only genus level was possible) and enumerated at 1500 x magnification under immersion oil using an Olympus® BX51 microscope equipped with Differential Interference Contrast (Nomarski) optics. Diatoms were very rare in the cryoconite hole samples, and therefore entire microscope slides were counted for diatom valves. In total, 9 samples were used for the community analysis, with 2 additional samples (C10 and C11, with only a few recovered valves) used in the "inkspot" diagram (for explanation, see below). Of these, total counts ranged between 40 and 216 valves per sample. One sample (C12) did not contain any frustules and was therefore removed from further analysis.

For the identification of diatom species from cryoconite hole material, we primarily consulted Pinseel (2014). Along with diatoms, the presence of Chrysophyte stomatocysts (golden brown algae) was also noted, although abundances were generally very low. Following Yallop et Anesio (2010), count data were combined at the genus level. Due to the widespread practice of "force-fitting" Arctic taxa into their European and North-American relatives (Tyler 1996), or the use of a too broad morphospecies concept (Mann 1999), a substantial number of taxa has

been incorrectly identified in the past. As a consequence, many studies which identified Arctic diatoms to the species level might not be reliable (Pinseel 2014). On the contrary, the genus level is taxonomically more robust and ensures consistency between datasets. Moreover, the

*Table A4.1. List of all samples used in this study and physico-chemical parameters of the study sites.*

Study code (sample code)	Location/ lake type	Temperature (°C)	Conduc- tivity (µS/cm)	pH	Altitude a.s.l. (m)	GPS N	GPS E
<b>Cryoconite holes</b>							
C1 (N002)	Glacier surface	0.5	1	8.30	130	78° 38' 24.7"	16° 58' 37.1"
C2 (N003)	Glacier surface	0.7	1	8.60	201	78° 38' 24.6"	17° 00' 05.1"
C3 (N004)	Glacier surface	0.6	1	8.30	244	78° 38' 22.2"	17° 01' 44.6"
C4 (N006)	Glacier surface	0.5	1	9.10	278	78° 38' 28.3"	17° 05' 11.0"
C5 (N007)	Glacier surface	0.4	7	9.30	361	78° 38' 29.2"	17° 06' 37.6"
C6 (N008)	Glacier surface	0.4	4	9.80	393	78° 38' 30.5"	17° 07' 22.7"
C7 (N009)	Glacier surface	0.3	1	8.50	262	78° 39' 21.4"	17° 03' 40.9"
C8 (N010)	Glacier surface	0.4	2	8.40	263	78° 39' 01.0"	17° 03' 31.9"
C9 (N011)	Glacier surface	0.5	12	10.10	267	78° 38' 41.5"	17° 03' 16.2"
C10 (N012)	Glacier surface	0.5	2	9.20	237	78° 38' 08.6"	17° 03' 01.0"
C11 (N013)	Glacier surface	0.4	11	8.80	233	78° 37' 51.9"	17° 02' 55.3"
C12 (N005)	Glacier surface	0.5	2	9.00	271	78° 38' 25.6"	17° 03' 46.9"
<b>Nordenskiöld lakes (Pinseel 2014)</b>							
N1 (SP20)	Kettle lakes	8.3	443	8.52	28	78° 38' 19.2"	16° 49' 35.8"
N2 (SP21)	Kettle lakes	8.6	329	8.55	24	78° 38' 19.4"	16° 49' 41.1"
N3 (SP22)	Kettle lakes	8.4	658	8.33	29	78° 38' 17.6"	16° 50' 1.5"
N4 (SP23)	Kettle lakes	8.7	566	8.60	26	78° 38' 17.6"	16° 50' 5.1"
<b>Hørbye lakes (Pinseel 2014)</b>							
H1 (SPH1B)	Kettle lakes	ND	ND	ND	37	78° 44' 05.7"	16° 26' 52.5"
H2 (SPH1C)	Kettle lakes	10.9	861	8.18	23	78° 44' 08.1"	16° 26' 51.9"
H3 (SPH2A)	Kettle lakes	11.7	461	8.39	41	78° 44' 13.4"	16° 26' 27.7"
H4 (SPH2B)	Kettle lakes	12.0	760	8.13	43	78° 44' 11.2"	16° 26' 15.2"
H5 (SPH2C)	Kettle lakes	11.6	756	8.20	44	78° 44' 11.8"	16° 26' 10.1"

H6 (SPH3A)	Kettle lakes	13.3	338	8.41	49	78° 44' 22.0"	16° 25' 04.8"
H7 (SPH3B)	Kettle lakes	12.7	306	8.42	50	78° 44' 23.9"	16° 24' 56.3"
H8 (SPH3C)	Kettle lakes	14.2	339	8.54	46	78° 44' 21.4"	16° 25' 08.2"
H9 (SPH4B)	Kettle lakes	11.3	423	8.28	52	78° 44' 34.2"	16° 24' 42.4"
H10 (SPH4C)	Kettle lakes	14.5	655	8.32	50	78° 44' 34.2"	16° 24' 48.7"
H11 (SPH5A)	Kettle lakes	13.9	1428	8.12	54	78° 44' 43.9"	16° 24' 27.4"
H12 (SPH5B)	Kettle lakes	12.5	1805	8.18	57	78° 44' 45.4"	16° 24' 37.0"
H13 (SPH5C)	Kettle lakes	13.3	1208	8.32	57	78° 44' 47.2"	16° 24' 39.5"
H14-15 (SP41-42)	Kettle lakes	11.6	1325	8.18	35	78° 44' 05.0"	16° 26' 34.1"
H16 (SP44)	Kettle lakes	12.7	180	8.45	40	78° 44' 13.0"	16° 26' 14.1"
H17 (SP45)	Kettle lakes	12.2	218	8.54	50	78° 44' 35.4"	16° 24' 44.9"
H18 (SP46)	Kettle lakes	9.2	198	9.20	106	78° 45' 14.0"	16° 21' 46.2"
H19-20 (SP47-48)	Kettle lakes	8.7	182	8.39	107	78° 45' 24.5"	16° 22' 00.8"
<b>Retrettøya ponds (as 'roche moutonnée/Oblík' in Pinseel 2014)</b>							
O1 (SP62)	Tectonic related	8.2	592	8.80	20	78° 39' 24.9"	16° 54' 46.5"
O2 (SP63)	Tectonic related	8.1	256	9.60	7	78° 39' 27.8"	16° 54' 34.3"
O3 (SP64)	Tectonic related	8.5	377	8.90	24	78° 39' 23.0"	16° 54' 44.4"
O4 (SP65)	Tectonic related	8.1	281	9.30	18	78° 39' 19.3"	16° 54' 37.8"
O5 (SP66)	Tectonic related	7.4	510	8.70	20	78° 39' 21.4"	16° 54' 39.9"
O6 (SP67)	Tectonic related	8.0	319	8.70	28	78° 39' 18.6"	16° 54' 38.5"
O7 (SP68)	Tectonic related	8.4	174	9.80	24	78° 39' 16.3"	16° 54' 21.9"
O8 (SP69)	Tectonic related	9.0	347	9.20	7	78° 39' 8.5"	16° 54' 51.4"
O9 (SP70)	Tectonic related	9.0	450	9.40	11	78° 39' 14.6"	16° 55' 28.5"
O10 (SP71-72)	Tectonic related	9.1	147	9.60	26	78° 39' 14.1"	16° 55' 16.4"
O11 (SP73)	Tectonic related	9.7	133	10.90	17	78° 39' 9.0"	16° 55' 8.7"

large amount of debris, together with an association of cells with mineral particles, makes the observation of single small valves rather difficult (Stibal et al. 2015), and sometimes even unfeasible when looking for diatoms in living samples (Vonnahme 2014), impeding identification of diatom valves up to the species level.

## Statistical analyses

To investigate the similarity of the cryoconite diatom flora with those of nearby freshwater habitats, we compared our cryoconite hole dataset with diatom counts from Pinseel (2014), who counted and identified 400 diatom valves in littoral samples from freshwater ponds and lakes in Petuniabukta, using the methods described above. Relative abundances were first calculated from diatom counts from both datasets, and an “inkspot” plot was created using the *rioja* R package (Juggins 2012) to manageably view diatom community structure among samples.

Diatom communities were statistically analyzed using the approaches applied by Stanish et al. (2012, 2013). Briefly, nonmetric multidimensional scaling (NMDS) analyses were performed to visualize relationships between communities from different samples, sites, and habitats. Rare species (< 1.0%) were removed, all data square-root transformed, and a distance matrix was calculated based on Bray–Curtis dissimilarity using the *vegan* R package (Oksanen et al. 2011). From this, a three dimensional model was created, which produced a Kruskal’s “stress” value of 9.88%, and strong nonmetric ( $R^2 = 0.99$ ) and linear fits ( $R^2 = 0.94$ ). To discern which genera drive patterns between samples, a corresponding NMDS figure was produced with genera > 5.0% in relative abundance superimposed.

Lastly, a hierarchical dendrogram was produced by creating a distance matrix as described above, utilizing the “average” clustering method (coeff = 0.81). To test for significant differences between the lake and cryoconite communities in general, the cryoconite hole diatoms communities were tested against the pooled lake diatom community data using permutational multivariate analysis of variance (PERMANOVA), with  $\alpha = 0.05$ . All analyses were performed using the R statistical environment (R Core Team 2014, see References – Other sources).

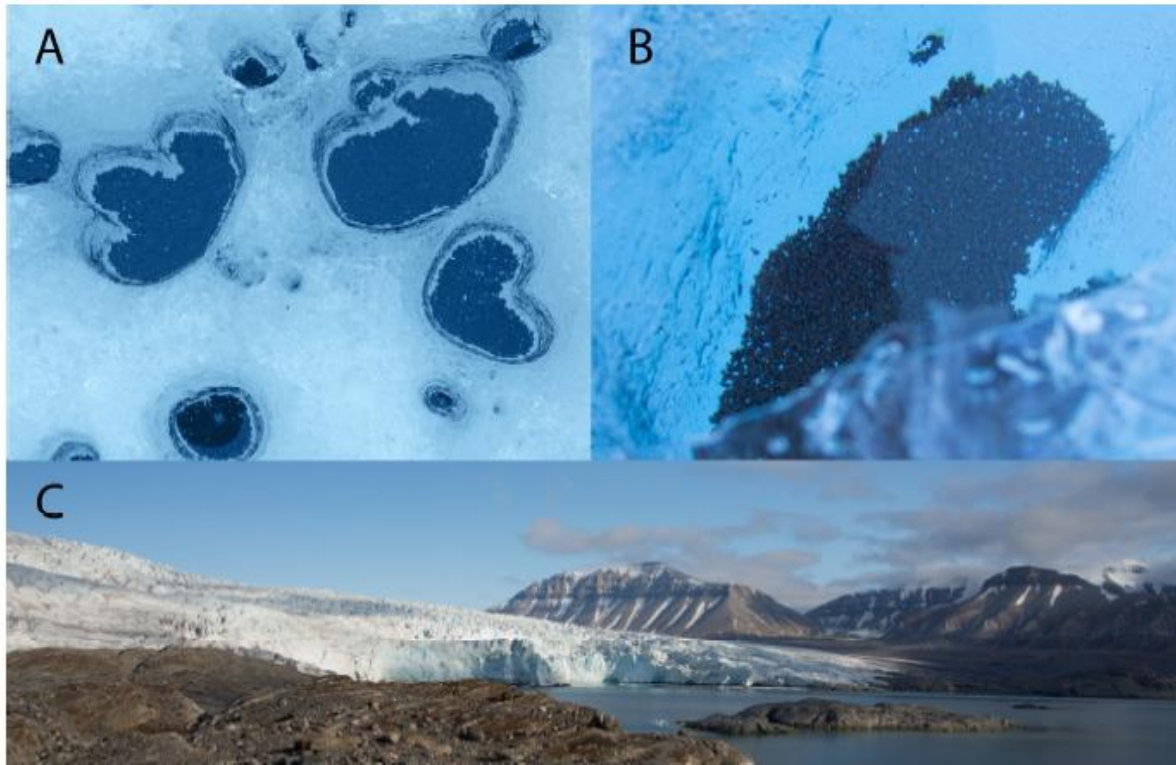


Figure A4.2. Examples of cryoconite holes (A–B), and an overview of the Nordenskiöld tide-water glacier (C).

## RESULTS

### Species composition of the cryoconite holes

A total of 58 diatom taxa (including species, subspecies, varieties and formas) belonging to 46 genera were identified in the cryoconite material (Fig. A4.3). An additional 26 diatom frustules could not be identified below genus level. Genus richness of the cryoconite samples ranged from 9 to 24 with a median of 18. Some genera were common throughout the sampling sites, such as *Pinnularia* Ehrenberg (in all samples), *Nitzschia* Hassall (in 8 out of 9 samples), *Staurosirella* D.M.Williams (8/9), *Gomphonema* Ehrenberg (8/9) and *Luticola* D.G.Mann (8/9). The dominance of these genera are also reflected in the number of counted valves: *Nitzschia* (17% of all counted valves), *Psammothidium* L. Bukhtiyarova (14%), *Pinnularia* (12%), *Staurosirella* (6%), *Gomphonema* (5%) and *Luticola* (5%).



The most species-rich genera were *Pinnularia*, *Nitzschia* and *Eunotia* Ehrenberg. Six *Pinnularia* taxa could be identified – *P. cf. brebisonii* (Kützing) Rabenhorst, *P. obscura* Krasske, *P. intermedia* (Lagestedt) Cleve, *P. schimanskii* Krammer, *P. rabenhorstii* (Grunow) Krammer, and *P. borealis* Ehrenberg (Fig. A4.3). The lattermost actually represents one of the most common species complexes (in 8 out of 9 samples) on the site, together with *P. intermedia* (7/9), *Nitzschia perminuta* complex form 2 (Pinseel 2014; 7/9), *Gomphonema aff. nathorstii* (Pinseel 2014; 8/9), and *Psammothidium* sp1 (Pinseel 2014; 7/9) - see Fig. 3.

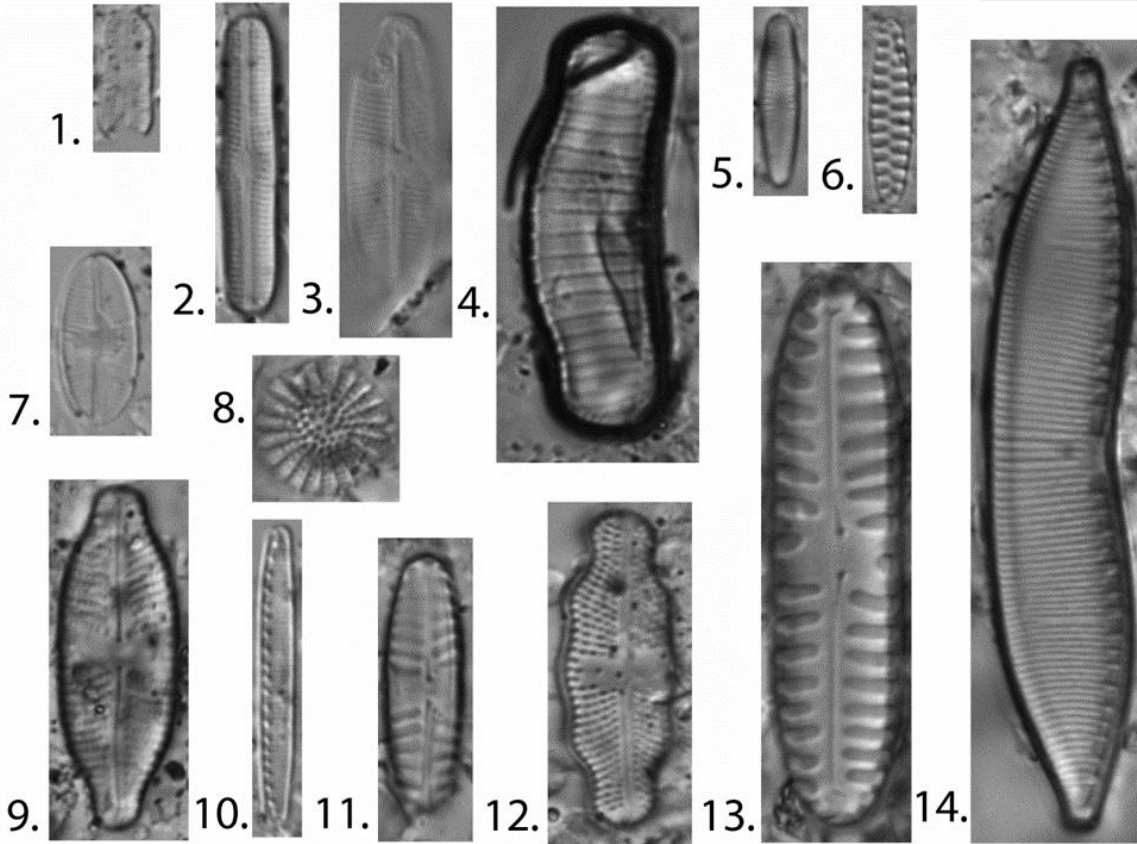
Several other *Nitzschia* taxa, such as *N. amphibia* Grunow, *N. communata* Grunow, *N. flexoides* Geitler, *N. palea* (Kützing) W.Smith and *N. sp8* (Pinseel 2014) have been observed in low numbers (containing less than 2.5 % out of the total species counts). Four *Eunotia* taxa have been identified: *E. cf. ambivalens* Lange-Bertalot & Tagliaventi, *E. curtagrunowii* Nörpel-Schempp & Lange-Bertalot (Fig. A4.3), *E. islandica* Østrup and *E. nymanniana* Grunow. Three species out of the most common taxa also dominated the cryoconite sites in terms of relative abundances: *Nitzschia perminuta* form 2, *Psammothidium* sp1 and *Pinnularia borealis* complex, represented 12%, 10% and 5% respectively.

### Local spatial comparison

To examine diatom community similarity between localities, the genus richness of the cryoconite holes and nearby lake habitats was compared. All observed genera, their abbreviations used for analyses and their distribution in between compared localities, can be found in Table 2. Even though only a low number of diatom valves could be enumerated from the cryoconite samples, overall genus richness was comparable, and sometimes even greater, than some of the lake sites (Fig. A4.4).

Nonmetric multidimensional scaling (NMDS) of lake and cryoconite hole communities revealed a strong separation between the two habitat types, both on NMDS axis 1 and 2 (see Fig. A4.5). Cryoconite hole diatom communities had greater proportions of the genera *Eunotia* (not visible on figure), *Aulacoseira* Thwaites, and *Gomphonema*, all of which strongly influenced

sample orientation on both NMDS axes. Lake habitats were strongly influenced by *Adlafia* Moser, Lange-Bertalot and Metzeltin, *Encyonema* Kützing, and *Mayamaea* Lange-Bertalot on



**Figure A4.3.** Selected diatom species observed in cryoconite holes of Nordenskiöld glacier. Scale bar = 10  $\mu$ m. 1. *Humidophila cf. contenta*. 2. *Chamaepinnularia sp.* 3. *Rossithidium petersenii*. 4. *Eunotia curtagrunowii*. 5. *Achnanthisidium minutissimum complex*. 6. *Staurosirella sp.* 7. *Psammothidium sp1*. 8. *Stephanodiscus cf. minutulus*. 9. *Gomphonema aff. nathorstii*. 10. *Nitzschia perminuta forma 1*. 11. *Pinnularia intermedia*. 12. *Luticola nivalis*. 13. *Pinnularia borealis complex*. 14. *Hantzschia amphioxys complex*.

NMDS axis 2. Habitat differences were further influenced by *Staurosira* Ehrenberg, *Staurosirella*, *Luticola*, *Pinnularia* and *Hantzschia* Grunow on axis 1 for the cryoconite holes, and *Encyonema*, *Cymbella* Agardh, *Diatoma* Bory de Saint-Vincent, and *Denticula* Kützing for the lakes. The Bray-Curtis cluster analysis separated communities into several distinct groups, of which, one exclusively consisted of all samples from the cryoconite holes (see Fig. A4.6). When the community data were compared with PERMANOVA, cryoconite hole diatom

communities were significantly different from the pooled lake samples ( $df = 43$ ,  $F = 15.64$ ,  $R^2 = 0.27$ ,  $p = 0.001$ ).

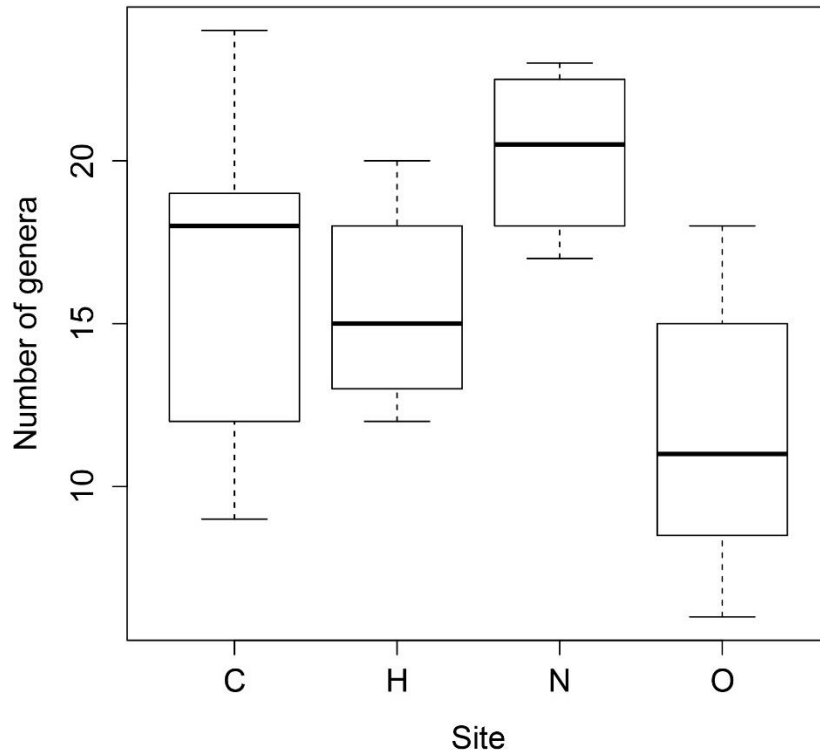


Figure A4.4: Genus richness of cryoconite holes (C), Hørbye lakes (H), Nordenskiöld lakes (N), and Retrettøya (O) sites.

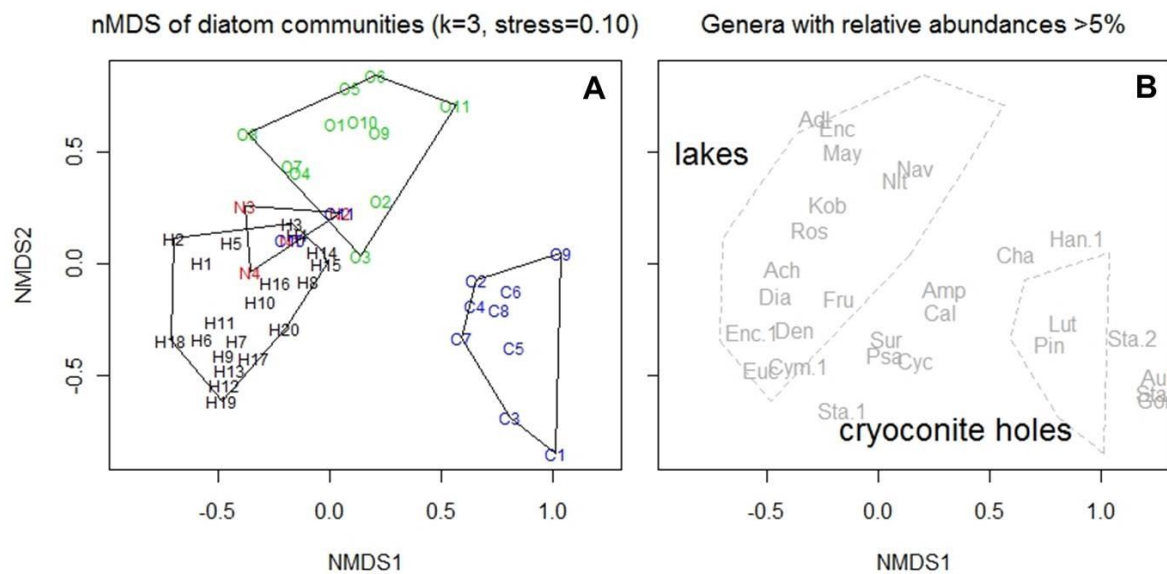


Figure A4.5. Non-metric multidimensional scaling (NMDS) of diatom communities from cryoconite holes and nearby lakes, indicating separation of the two habitats. C1–C9 for cryoconite holes, H1–H20 for Hørbye lakes, N1–N4 for Nordenskiöld lakes, and O1–O11 for Retrettøya. Diatom genus abbreviations are given in Table 2.

**Table A4.2.** List of diatom genera (with abbreviations) from cryoconite holes (C), Hørbye lakes (H), Nordenskiöld lakes (N) and Retrettøya ponds (O).

Genus	C	H	N	O
<i>Achnantheidium (Ach)</i>	x	x	x	x
<i>Adlafia (Adl)</i>		x	x	x
<i>Alveovallum (Alv)</i>	x			
<i>Amphora (Amp)</i>	x	x		
<i>Aulacoseira (Aul)</i>	x			
<i>Brachysira</i>			x	
<i>Caloneis (Cal)</i>	x	x	x	x
<i>Cavinula</i>	x			
<i>Chamaepinnularia (Cha)</i>	x	x		x
<i>Cocconeis (Coc)</i>	x			
<i>Craticula (Cra)</i>		x		x
<i>Cyclotella (Cyc)</i>	x		x	
<i>Cymbella (Cym)</i>		x	x	x
<i>Cymboplectra (Cym.1)</i>	x	x	x	x
<i>Denticula (Den)</i>	x	x	x	
<i>Diatoma (Dia)</i>	x	x	x	x
<i>Diploneis (Dip)</i>	x	x	x	
<i>Encyonema (Enc)</i>	x	x	x	x
<i>Encyonopsis (Enc.1)</i>	x	x	x	
<i>Epithemia (Epi)</i>	x			
<i>Eucocconeis (Euc)</i>	x	x	x	x
<i>Eunotia (Eun)</i>	x			
<i>Fallacia</i>				x
<i>Fistulifera</i>				x
<i>Fragilaria</i>		x	x	
<i>Frustulia (Fru)</i>		x		
<i>Geissleria</i>				
<i>Gomphonema (Gom)</i>	x			
<i>Gyrosigma</i>		x		
<i>Halamphora (Hal)</i>		x	x	x
<i>Hannaea (Han)</i>	x	x		
<i>Hantzschia (Han.1)</i>	x	x	x	x
<i>Humidophila (Hum)</i>	x	x	x	
<i>Hygropetra (Hyg)</i>	x			
<i>Kobayasiella (Kob)</i>		x	x	x
<i>Luticola (Lut)</i>	x	x	x	x
<i>Mayamaea (May)</i>		x	x	x
<i>Melosira</i>				x
<i>Microcostatus</i>				x
<i>Muelleria (Mue)</i>			x	x
<i>Navicula (Nav)</i>			x	x
<i>Neidium (Nei)</i>			x	x
<i>Nitzschia (Nit)</i>			x	x
<i>Orthoseira (Ort)</i>			x	
<i>Pinnularia (Pin)</i>			x	x
<i>Placoneis (Pla)</i>			x	
<i>Planothidium (Pla.1)</i>			x	
<i>Psammothidium (Psa)</i>			x	x
<i>Pseudostaurosira (Pse)</i>			x	
<i>Rossithidium (Ros)</i>			x	x
<i>Sellaphora (Sel)</i>				x
<i>Simonsenia</i>				x
<i>Stauroforma (Sta)</i>			x	
<i>Stauroneis (Sta.1)</i>			x	x
<i>Staurosira (Sta.2)</i>			x	
<i>Staurosirella (Sta.3)</i>			x	
<i>Stephanodiscus (Ste)</i>			x	
<i>Surirella (Sur)</i>			x	x
<i>Tabellaria (Tab)</i>			x	

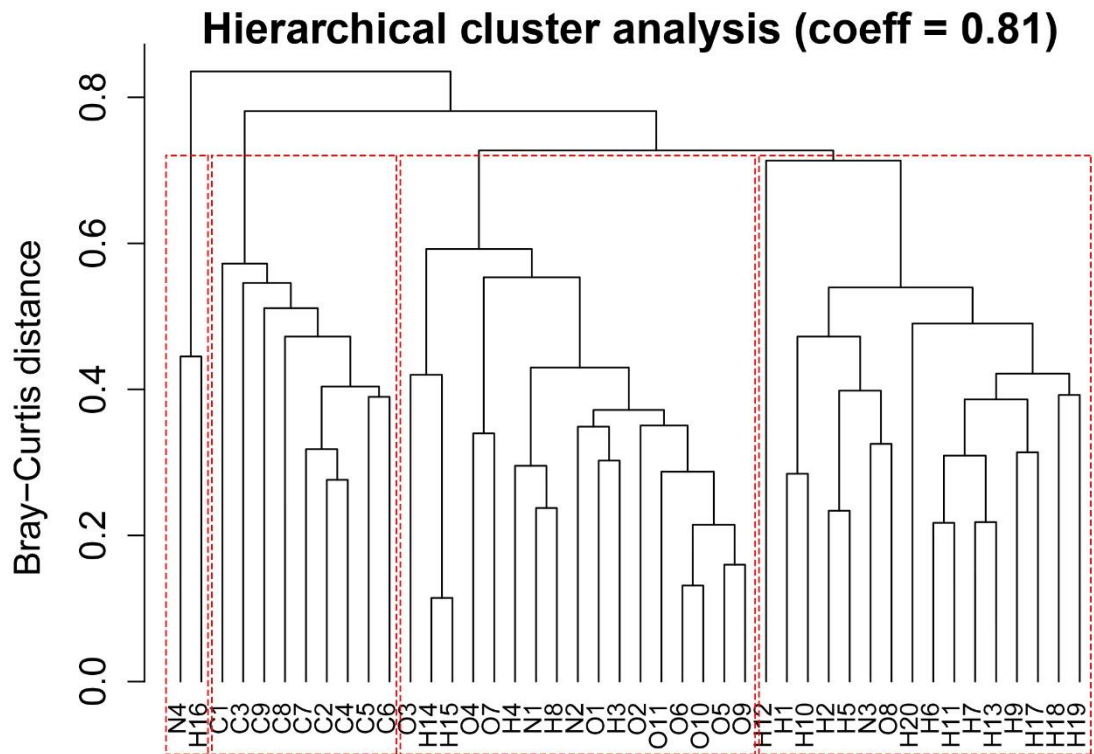
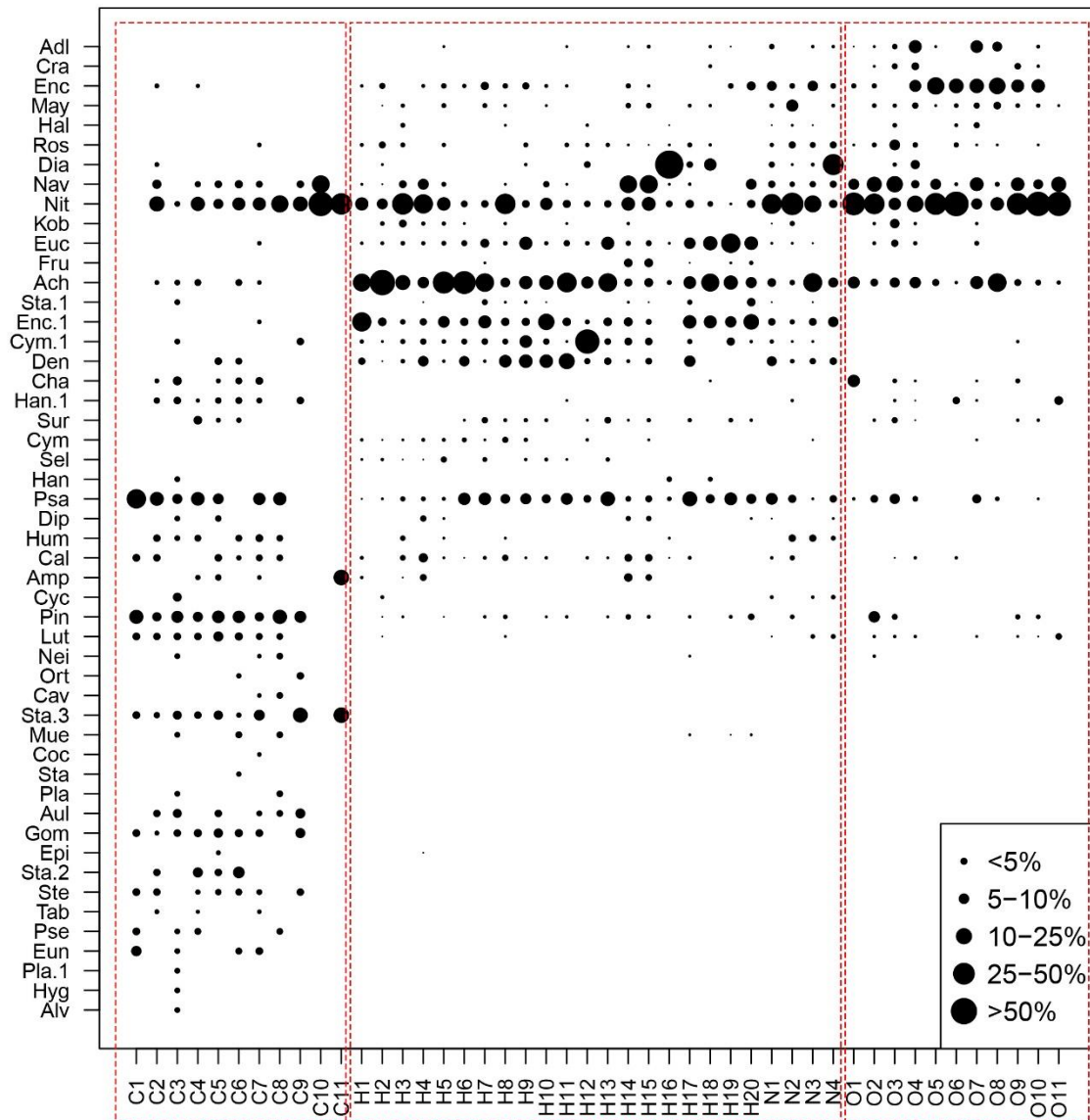


Figure A4.6. Bray-Curtis cluster analysis. C1–C9 for cryoconite holes, H1–H20 for Hørbye lakes, N1 – N4 for Nordenskiöld lakes, and O1–O11 for Retrettøya.

Despite such clear split of both habitat types, some genera were present between both localities studied as seen in the ‘inkspot’ plot. This diagram, which can be used to visualize the community structure among samples using the raw relative abundance data (Fig. A4.7), resulted in a clear separation of three groups. The first consisted entirely of samples from cryoconite holes. The second group linked both localities of the Hørbye and Nordenskiöld moraine kettle lakes. The third group consisted entirely of samples of Retrettøya. Some diatom genera showed clear differences between these three groups: *e.g.*, *Encyonema* and *Adlafia* were both abundant in the third group. *Nitzschia*, although prevailing in the entire dataset, was clearly less abundant in the samples of the second group. *Psammothidium* was almost equally abundant in the first two groups, and, finally, *Pinnularia*, *Luticola*, *Staurosirella*, together with other genera (lower part of Fig. A4.7), clearly separated the first group from the remaining two.



*Figure A4.7. An 'inkspot' plot visualizing the diatom community structure among samples using relative abundance data. Separation of three groups is shown. C1–C11 for cryoconite holes, H1–H20 for Hørbye lakes, N1 – N4 for Nordenskiöld lakes, and O1–O11 for Retrettøya ponds.*

## DISCUSSION

Although numerous studies have reported the presence of diatoms in cryoconite holes, many questions remain as to their origin, viability, community structure, and assembly. Here, our aim was to describe and compare the diatom assemblages from cryoconite holes of Nordenskiöld glacier to the communities from lake habitats in the immediate vicinity which might serve as a potential source. We hypothesized that if diatoms are being

transported from surrounding aquatic habitats to cryoconite systems, then cryoconite communities should be highly similar to the communities from the habitat of origin.

We found that the diatom communities of the cryoconite hole sediments were distinctly different from those from the adjacent lake habitats. Not only did cryoconite sediment contain different genera not observed in lake habitats, but often a higher number of genera was observed in the cryoconite communities compared to the lake sediment samples, even when only a fraction of the amount of valves was counted. This may suggest that (i) cryoconite hole diatom communities have a unique structure, albeit at low cellular densities, or that (ii) these cryoconite communities are not self-sustaining and are dependent on dispersed cells, but are derived from more varied sources than our surveyed lakes.

If these communities were actively reproducing, it could be argued that this should be reflected in a community dominated by only a few species or genera. On the contrary, our results show the presence of a lot of different genera with a low number of species and valves within the cryoconite holes, which suggest that these assemblages are more likely to be formed as a result of aeolian dispersion and deposition. While there is much left to learn about these extreme environments, our study adds further evidence that the diatom flora from cryoconite holes is unique and distinct from adjacent freshwater habitats. Recently, there is a need for further study that could help in developing a more complete understanding of diatom biogeography, dispersal, and evolution.

### **Ecology of individual diatoms**

Our results showed that the cryoconite diatom community composition exhibited only little similarity to any of the lake diatom communities in the vicinity, despite the fact that 25 genera occur in both habitat types. These shared genera may indicate that these lakes contribute to the diatom community in the cryoconite holes (though investigation at a finer resolution would be necessary to support this claim). On the other hand, it is clear that at least one other source is necessary to explain the higher number of genera in the Nordenskiöld glacier cryoconite holes, a number of which are rare or uncommon to the

surrounding lakes. Similar results have been reported by Stanish et al. (2013), who found that cryoconite hole communities were distinctly different from adjacent stream habitats in the McMurdo Dry Valleys (though they certainly shared more taxa than in our study). Another supporting data come from Edwards et al. (2013c), who found that cryoconite bacterial communities significantly differed from those from the glacier margins in Svalbard.

A number of diatom genera found in cryoconite holes, including *Hantzschia*, *Humidophila* Lowe et al., *Orthoseira* Thwaites, *Pinnularia* and *Luticola*, are typically aerophilic genera, known to thrive mainly in moist terrestrial soil or moss habitats. Diatom cells from soils can be easily transported by wind together A number of diatom genera found in cryoconite holes, including *Hantzschia*, *Humidophila* Lowe et al., *Orthoseira* Thwaites, *Pinnularia* and *Luticola*, are typically aerophilic genera, known to thrive mainly in moist terrestrial soil or moss habitats. Diatom cells from soils can be easily transported by wind together with sediment or volcanic dust (Dagson-Waldhauserová et al. 2015), or be attached to small moss patches able to overgrow small cryoconite holes and thus becoming a 'glacial mouse' (Vonnahme 2014). *Pinnularia borealis* and *Hantzschia amphioxys* (Ehrenberg) Grunow, two species highly abundant in cryoconite samples, are frequently reported from polar soils and also commonly recorded from mosses (Beyens 1989, Van de Vijver et al. 2003, Vinocur et al. 2010). Terrestrial diatoms make good candidates for cryoconite colonization because they are more able to resist freezing and desiccation than freshwater species (Souffreau et al. 2010, Yallop et al. 2010). Abrupt freezing can be lethal for diatom vegetative cells, especially for non-terrestrial species (Souffreau et al. 2010, 2013). Furthermore, some of these diatoms could be present in resting stages, increasing thus the ability to survive freezing stress (Souffreau et al. 2013).

In our study, rather than more, few genera such as *Adlafia*, *Encyonema*, *Navicula* and *Nitzschia*, were present in greater relative abundances from the Retrettøya proglacial site, in contrast to the other two lake localities. According to our field observations, no direct hydrological connection exists between ponds on Retrettøya and the glacier. However, plenty of fresh glacial sediment, including cryoconite material, was present drying on the margin and mobilized by wind, which blows from the glacier towards the fjord.



Consequently, the Retrettøya ponds might be supplied by cryoconite communities, similar as suggested by Vonnahme (2014) for Hørbye glacier and proglacial ponds in its frontal moraine.

Species of the genera *Achnantheidium* Kütz., *Denticula* Kütz., *Encyonema*, and *Eucoconeis* Cleve had relatively high abundances in moraine kettle lakes. These genera were uncommon or rare in the cryoconite samples, most likely suggesting their preference for more established (aquatic) habitats. The *A. minutissimum* (Kütz.) Czarnecki complex reaches high abundances in various freshwater habitats across Petuniabukta and actually presents the most common freshwater diatom complex in the area (Pinseel 2014). Although this genus was rarely found in the cryoconite holes sampled for diatoms (this study), fresh cryoconite material from the same area observed by Vonnahme (2014) in the field laboratory revealed a few viable individuals of *Achnantheidium*. *Psammothidium* is another very interesting genus that connects the cryoconite holes to the moraine kettle lakes of Nordenskiöld and Hørbye. Moreover, species of this genus prefer habitats with sandy bottoms (Round et Bukhtiyarova 1996). This is in contrast with another highly abundant genus, *Nitzschia*, which dominates both the cryoconite habitats and the Retrettøya ponds, but usually occurs in lower abundances in the kettle lakes.

*Luticola* was a rather common genus in cryoconite holes involved into our study. Previously, *L. ventricosa* (Kütz.) Mann and *L. nivalis* (Ehrenberg) Mann have been retrieved alive from frozen cryoconite material (Yallop et Anesio 2010). *Luticola nivalis* was also observed in the present study, together with *L. frequentissima* (see Fig. 3). Species of the genus *Muelleria* (Frenguelli) Frenguelli are not abundant in the Arctic, and only one species resembling the cosmopolitan *M. terrestris* (Petersen) Spaulding & Stoermer was observed in this study. The species was previously reported from cryoconite holes by Yallop et Anesio (2010). *Muelleria* is, however, one of the most dominant genera of Antarctic cryoconite diatom communities, accompanied by species from genera *Humidophila* and *Luticola* (Mueller et al. 2001, Van de Vijver et al. 2010, Stanish et al. 2013, Kohler et al. 2015).

## Insights into microbial dispersal

While we did not check for cell viability in our samples due to the limited amount of material, we can postulate that viability may have been low, as recovered valves were often broken in addition to being sparse. However, earlier studies have suggested that at least some cryoconite diatom cells are viable. Stanish et al. (2013) and Vonnahme (2014) have both reported viable valves by microscopic analysis, and Yallop et al. (2010) were able to culture 27 diatom genera from cryoconite material, even after being frozen for 1-2 years. Vonnahme (2014) analyzed only a few samples ( $n = 3$ ) of fresh material from cryoconite holes of Nordenskiöld and Hørbye glaciers for diatoms content, and reported following genera and species as living, although in low numbers: *Achnanthisidium* sp., *Encyonopsis laevis* Nägeli and *E. subminuta* Krammer & E. Reichardt, *Humidophila* sp., *Pinnularia* cf. *obscura*, and *Psammothidium* cf. *marginulatum* (Grunow) Bukhtiyarova and Round. All of these were also identified in our study of fixed material, albeit present in various abundances.

In Antarctica, some diatom species, such as *Muelleria cryoconicola* Stanish & Spaulding (Van de Vijver et al. 2010), *Luticola bradyi* Kohler, and *L. spainiae* Kohler & Kopalová are thus far found almost exclusively in cryoconite holes, and have been suggested to be endemic to these habitats. According to Kohler et al. (2015), transportation of diatom cells from cryoconite holes to surrounding aquatic habitats might even take place, as suggested for *L. bradyi*, a species found in a large population in cryoconite material, but only very rarely observed in glacial meltwater streams. These above observations, together with the differences in cryoconite diatom communities from marginal habitats, provide evidence that while diatoms may be seeded from adjacent habitats, at least some survive, live, and possibly reproduce while in cryoconite holes. While unique cryoconite diatom communities are thought to develop in stable cryoconite systems found in Antarctica (Stanish et al. 2013), Arctic systems are much more exposed to the outside world and do not persist as long due to glacier hydrology and melting. Despite this, they can still be colonized by living diatoms (Yallop et al. 2010), and these habitats might subsequently select the most adapted species to live in.

In our study, many genera observed in the cryoconite material were rare in the lakes and ponds of Petuniabukta. For example, the genera *Aulacoseira*, *Tabellaria* Ehrenberg, *Melosira* Agardh, *Orthoseira*, and *Stephanodiscus* Ehrenberg were present in the cryoconite holes, but have never been observed in the lakes and ponds of Petuniabukta (> 50 lakes studied, in Pinseel 2014). One *Gomphonema* taxon (*i.e.* *Gomphonema* aff. *nathorstii*, Fig. 3) was recorded as common in almost all (eight out of nine) cryoconite samples but was visually absent from the surrounding lakes. However, the same species has been observed previously in a temporal pond on a mountain top in Mimerdalen (Pinseel, *unpublished data*), suggesting that this taxon prefers ephemeral habitats. Several other species of *Gomphonema* were present within the cryoconite samples, although absent from nearby lakes, and the same is true for species of *Staurosira*, *Staurosirella* and *Eunotia*. Some genera (*e.g.* *Achnanthydium*, *Gomphonema*, *Psammothidium*, *Staurosirella*) may produce (mucilaginous) stalks (Potapova 2009, Gesierich et Rott 2012) so they can be attached to solid objects, hypothetically favoring them in aeolian dispersion. It was striking that no marine species have been identified from the cryoconite material, despite the immediate vicinity of the sea, even though direct evidence of marine diatom deposition on ice sheets was previously reported before from the Antarctic (Budgeon et al. 2012). This is probably because the prevalent wind direction is in the opposite way, which reduces the extent of marine deposition of diatoms compared to those that are limnoterrestrial in origin.

As light microscope studies of cryoconite material are challenging, it is possible that the lack of records of diatom species inhabiting cryoconite holes is a result of under-sampling, as previously hypothesized by Yallop et Anesio (2010). In total, some 84 taxa were identified in our study, and broader sampling efforts would certainly reveal more taxa. Our gradient sampling also revealed that sites closer to the side of the glacier contained less diatom valves than those situated more inside. This could further support the suggestion of aeolian input of cells rather than the cell input from avalanches of eroded material that occurs on glaciers sides (Landford et al. 2011). On the contrary, sites that were situated the most to the center of the Nordenskiöld glacier (C8 and C9) contained less (roughly half) of the average counted valves. It might be interesting to sample across the whole ablation

zone of the glacier by adding further sampling sites, as it could reveal wind patterns on a local scale (*i.e.* considering katabatic winds).

### **Future directions and conclusions**

Contrary to the Antarctic diatom flora, which has been recently revised based on a more narrow morphology-based species concept (ex. Van de Vijver et al. 2010a, 2011, 2013, Kopalová et al. 2012, 2013, Taylor et al. 2014, among others), the Arctic diatom morphological taxonomy is insufficient at this time to make fine-scale comparisons between habitats. Moreover, Pinseel (2014) have identified a high number of new diatom taxa ready to be described. However, in this study, we found the genus-level to be of great use to compare cryoconite holes with adjacent freshwater habitats, and it became clear that our comparison set of localities in the immediate vicinity was not broad enough to fully assess 'the source' of the cryoconite diatom flora. It is possible that a more broad sampling effort to include non-aquatic habitats nearby, as well as more distant localities such as Iceland (Dagsson-Waldhauserova et al. 2015) are necessary to discern the cryoconite diatom flora 'source'. In any event, the diatom communities of cryoconite holes have the potential to inform researchers about microbial dispersal patterns through comparing the regional distribution of diatom taxa in between distinct polar habitats, and deserve further study in our investigation of the cryosphere.

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