

Using water chemistry to define ecological preferences within the moss genus *Scorpidium*, from Wales UK.

Jonathan Graham¹, Gareth Farr², Lars Hedenäs³, Aurelie Devez⁴ & Michael J Watts⁴.

¹ 2 Cross Road, Whittlesey, Cambridgeshire, PE7 1LX, UK

² British Geological Survey, Cardiff University, Park Place, Cardiff, CF10 3AT, UK

³ Swedish Museum of Natural History, Department of Botany Box 50007, SE -1040 05 Stockholm, Sweden

⁴ British Geological Survey, Nicker Hill, Keyworth, Nottingham, NG12 5GG, UK

Corresponding author email: jonathan.graham@ntlworld.com

1 **Abstract**

2 Three *Scorpidium* species; *S. scorpioides*, *S. cossonii* and *S. revolvens* are often associated with habitats
3 of high conservation value. This is the first attempt to define the chemical niches of these *Scorpidium*
4 species in Wales (UK) and allows us to compare to earlier European datasets. Water chemistry from
5 sixteen locations was analysed from direct squeezing of mosses from a total of 77 spots and the
6 principal water supply, e.g. springs and seepages. Spherical k-means clustering suggests there are two
7 distinct groups in the dataset; one characterised by *S. cossonii* and another by *S. revolvens*, associated
8 with differences in pH and Electric Conductivity (EC) of the habitat. The Wales habitats have higher pH
9 and EC than Scandinavian habitats of the species, which could potentially be a result of different
10 pollution histories or species compositions of the areas, the latter leading to different realised niches
11 along the mineral poor to rich gradient. It is hoped that a better understanding of the chemical niches
12 will support site managers and environmental regulators to make evidence-based decisions to protect
13 these species and their habitats.

14 **Keywords:** cluster analysis, petrifying springs, Habitats Directive

15 **Introduction**

16 Humans have long since influenced European nature both directly, through exploitation and
17 management of different resources (e.g., Antrop, 2004; Kaplan *et al.*, 2009; Price, 2000) and indirectly,
18 as shown by the ongoing climate warming (Met Office, UK's National Meteorological Service;
19 <https://www.metoffice.gov.uk/>; accessed 2019.01.05). To protect our environment, including its
20 biodiversity, we need methods and indicators that provide warnings when we come close to
21 irreparable damage. A relatively rare European habitat that could potentially be negatively influenced
22 by climate change-related effects, such as higher temperatures and increased evapotranspiration on
23 the one hand, and increased precipitation on the other, is the European Habitats & Species Directive
24 habitat 'H7220 Petrifying springs with tufa formation (Cratoneurion)'. Member states, including the

25 UK at the time of writing, are required to report on the conservation status of these habitats to the
26 European Commission.

27 Species of the genus *Scorpidium* (Calliergonaceae) are often, but not exclusively associated with
28 European Habitats & Species Directive habitat H7220, and Scandinavian studies (Kooijman & Hedenäs,
29 1991; Hedenäs & Kooijman, 1996; Tahvanainen, 2004) suggest that they are useful as indicators of its
30 status. *Scorpidium* includes three species, *S. scorpioides* (Hedw.) Limpr., *S. cossonii* (Schimp.) Hedenäs
31 and *S. revolvens* (Sw.ex Anonymo) Rubers (Hedenäs, 1989). The genus occurs globally from temperate
32 to arctic areas and in both the northern and southern hemispheres (Table 1). Water chemistry has
33 been used as a useful tool to distinguish habitat preferences within the genus in Scandinavia (Kooijman
34 & Hedenäs, 1991). This methodological approach has been applied in different ecological studies of
35 aquatic bryophytes (Kooijman et al., 1994; Ceschin et al., 2012) and its possible applicability to sites in
36 the UK with *Scorpidium* species is of interest to both environmental regulators and site managers for
37 whom an improved understanding of the habitat requirements, and risks, will better inform decisions
38 regarding management and conservation of these sites.

39 Field and laboratory observations (Kooijman & Hedenäs, 1991; Tahvanainen, 2004) show that *S.*
40 *revolvens* occurs in a much narrower range of pH and conductivity than *S. scorpioides* and *S. cossonii*
41 which are more often associated with calcium-rich spring-influenced habitats. Sites with *S. scorpioides*
42 and *S. cossonii* are characterised by relatively high pore water Ca concentrations (Mettrop *et al.*, 2018)
43 while *S. revolvens* is absent from calcium-rich sites (Hedenäs, 2003a). Acidification and eutrophication
44 can have a negative effect and has resulted in the decrease of *S. scorpioides* populations in the
45 Netherlands (Kooijman, A. 1992 and Kooijman & Westhoff, 1995). A recent study of Dutch sites
46 (Paulissen, *et. al.*, 2016) found that rich fens, i.e., mineral-rich fens that are often, but not always
47 species-rich and include certain indicator species, such as *S. cossonii* (Joosten *et al.*, 2017; Rydin *et al.*,
48 1999), are at risk of rapid transition to poor fens by increased deposition of reduced nitrogen.
49 Increased ammonium levels are shown to be directly toxic to *Scorpidium*, while *Sphagnum* was not

50 affected. In addition, nitrogen (in particular nitrate) can indirectly influence moss growth through
51 promoting vascular plants and Covuer et al. (2016) has highlighted the negative impact of
52 eutrophication (nitrite and particularly phosphate) on the H7220 habitat in Belgium. *Scorpidium*
53 *scorpioides* and *S. cossonii* sites in Sweden and the Netherlands are shown to be Ca-rich and Fe-poor
54 with low associated P-availability (Mettrop et al., 2018); this further highlights the potential
55 vulnerability of these sites to the process of eutrophication. Growth experiments comparing Canadian
56 and Dutch *S. scorpioides* (Vitt et. al., 1993) found that Canadian plants respond best when grown in
57 extreme-rich fen waters, while plants from the Netherlands respond best in waters from moderate-
58 rich fens and in nutrient enhanced conditions. These authors suggested that this could be a result of
59 ecotypic differentiation.

60 All these studies show that water chemistry is a critical factor affecting *Scorpidium* species and has
61 important implications for the conservation of rich fens. Here we present the first Wales (taken to
62 represent UK) dataset of water chemistry characterising the different chemical niches of the three
63 *Scorpidium* species and compare this data set with data previously compiled in Sweden (Kooijman &
64 Hedenäs, 1991). We ask whether all three species occur under similar water chemistry conditions at
65 localities in Wales, and evaluate whether conditions differ between Wales and Sweden.

66 **Materials and methods**

67 *Scorpidium* sites were identified from wetlands within designated sites (e.g. Special Areas of
68 Conservation, National Nature Reserves), from the British Bryological Society (BBS) database and from
69 expert local knowledge. The location, site extent, altitude and geology, as well as other characteristics
70 and sampling years for the 16 sites included within this study are shown in Figure 1 with other key
71 study areas in Sweden, the Republic of Ireland, the Netherlands, Denmark and Finland illustrated on
72 the inset map. Bryophytes commonly associated with springs and flushes (such as *Scorpidium* species)
73 can often occur towards the margins of areas of flushed vegetation or as part of hummocky vegetation
74 that is partly raised above the flushing water (i.e., away from the main water supply to a site). To

75 reflect this, water samples were collected both directly from the main water supply and by squeezing
76 stands of the three *Scorpidium* species that represented the different habitats found by visual
77 inspection at each locality. Seventy seven samples were collected from 16 sites with a range of
78 between 1 and 9 samples per site reflecting both size and structural complexity of individual sites.
79 Water chemical sampling was undertaken in January 2015 and 2016.

80 First a sample was obtained from as close to the water source as possible. If the flow was too diffuse
81 to allow collection directly into a bottle, we used a 50 ml syringe to collect water. Field parameters
82 with temperature (°C), pH and electrical conductivity, ($\mu\text{s}/\text{cm}$) (EC) were measured using a 'YSI™
83 Professional' field meter. Samples for spring water were filtered on site using a 0.45 μm filter and
84 stored in two 30 mL Nalgene bottles. For sampling of individual stands of *Scorpidium* species, samples
85 were collected as described and the moss was squeezed using a stainless steel potato ricer into a
86 stainless steel jug, allowing sufficient depth of water from which to measure field parameters. Many
87 of these samples, from now onwards called 'moss water', were too turbid to filter in the field. In these
88 cases, water samples were collected in two 30 mL Nalgene bottles and returned to the lab to settle
89 and be filtered through a 0.45 μm single use membrane filter (Sartorius-Minisart®) into 30 mL Nalgene
90 bottles stored at 1-5 °C prior to conductivity and IC analysis. The sample for ICP-MS analysis was fixed
91 in 1% HNO₃ and stored at 1-5 °C (see below). pH and alkalinity measurements were undertaken on
92 unfiltered un-acidified samples. pH was measured in the field and also in the lab, potentiometrically
93 by immersing a combined multi-probe electrode (XC-161) into the solution. Titration was performed
94 using 0.005 M sulphuric acid prepared from VWR Prolabo Convolve Normadose 0.1N solution made up
95 to volume with freshly prepared MilliQ. Analysis of major anions, including Ca²⁺, Na⁺, Mg²⁺, K⁺, SO₄²⁻,
96 Cl⁻, HCO₃⁻, F⁻, NO₂⁻, Br⁻, NO₃⁻ and HPO₄²⁻, were undertaken on a Dionex ICS5000 Ion Chromatograph
97 system (S/N 10060887) controlled by the Chromeleon Software (version 7). Ionic balances were
98 performed for each sample. Fifty seven major and trace elements in the core waters were analysed
99 on an Agilent 7500cx series, quadrupole inductively coupled plasma mass spectrometer (ICP-MS) with
100 an ORS, in combination with a CETAC autosampler. The ICP-MS instrument was calibrated at the

101 beginning of every analytical run using at least three standards and a blank for each trace element and
102 three standards and a blank for major elements.

103 Relatively pure stands of *Scorpidium* species occurring within each site were identified and selected in
104 the field. Following squeezing to obtain a moss water sample the squeezed moss sample was collected
105 and dried. Once dried, the sample was carefully examined and the small percentage of other
106 bryophytes sometimes present estimated and recorded. For these occasional mixed stands,
107 associated species have been annotated against the relevant chemical analysis. All samples were
108 retained as a reference collection and all *S. cossonii* and *S. revolvens* specimens were checked
109 microscopically following Hedenäs (1989). Other species occurring at sites were also recorded with an
110 estimate of abundance using the DAFOR scale. Using ArcView, a Geographical Information System
111 (GIS), elevation to the nearest 5 maOD (above ordnance datum) and geological information (British
112 Geological Surveys' DigiMap' 1:50,000 scale bedrock and superficial mapping) was assigned to each
113 site.

114 **Statistical methodology**

115 Four moss water chemistry parameters, EC, pH, Ca and HCO_3^- , were compared between Wales and
116 Sweden for each of the three species. We used a preliminary ANOVA to check that the residuals were
117 approximately normally distributed. This being the case, we compared our data sets for Wales and
118 Sweden using ANOVA in the program STATISTICA 13 (Dell Inc., 2015).

119 From the full water chemistry dataset for Wales, only major ions and trace elements with average
120 concentration > 0.2mg/L were considered relevant for statistical analysis. This included all the major
121 ions and the trace element Mn. In addition to standard calculations of standard deviation (SD),
122 standard error (SE), t-test (t), full statistical analysis of Welsh data was undertaken using a spherical k-
123 means clustering computer programme (Hill et. al., 2013).

124 **Results**

125 Following Kooijman & Hedenäs (1991), elevation and water chemistry data (moss water) of 124
126 samples from Sweden were compared against 77 samples from Wales. Water chemistry comprised
127 seven major ions (Ca, Mg, HCO_3^- , SO_4^{2-} , Cl^- , Na, K). The same relationship between the three *Scorpidium*
128 species and water chemistry (as described by Kooijman & Hedenäs, 1991) was seen with Welsh data,
129 where *S. revolvens* occurs in a much narrower range of pH and conductivity than the other 2 species.
130 Figure 2 shows this relationship between pH and EC for all Swedish and Welsh samples.

131 Swedish and Welsh data show ecological separation of *S. cossonii* and *S. revolvens* in terms of water
132 chemistry. *Scorpidium cossonii* is associated with water that has significantly higher concentrations of
133 HCO_3^- , pH, Ca, Mg and EC while conversely, *S. revolvens* is associated with significantly lower
134 concentrations of these. Although both Swedish and Welsh data shows clear ecological separation of
135 *S. cossonii* and *S. revolvens* in terms of water chemistry, Welsh *Scorpidium* sites occur at higher, pH
136 (all species) and EC (*S. revolvens*), levels than found at Swedish sites (Table 2).

137 **Statistical Results for Welsh sites**

138 Statistical analysis was undertaken on 77 water samples from 16 Welsh sites for pH, EC, major ions
139 and trace elements occurring at a mean concentration > 0.2 mg/L. Samples comprised both flush
140 water and moss water. Moss water samples comprise a pure bryophyte species stand or occasionally
141 up to as many as five separate bryophyte species. A summary of moss water quality data is provided
142 in Table 3. There are significant differences between the moss water chemistry of *Scorpidium cossonii*
143 and *Scorpidium revolvens* sites (SE and t value) with respect to HCO_3^- , pH, Ca, Mg and EC. These
144 conclusions further support the findings of Kooijman & Hedenäs (1991). We could not demonstrate
145 statistical difference between moss and flush water for *Scorpidium* species.

146 **Statistical species clustering**

147 Using a spherical k-means clustering analysis of combined site and water chemistry data, Welsh sites
148 were separated into two and three species clusters. The most distinct separation was found with a
149 two site/species cluster (Table 4), one group characterised by *Scorpidium cossonii* and a second group

150 characterised by *Scorpidium revolvens*. A summary of the differences in water chemistry between
151 these two groups is provided by Table 5.

152 Group 1 (*S. cossonii*) is associated with water with higher concentrations of Ca, Mg, frequent
153 *Palustriella falcata*, *Campylium stellatum*, *Bryum pseudotriquetrum*, occasional *Scorpidium*
154 *scorpioides* and are never associated with *Sphagna*. Group 2 (*S. revolvens*) is associated with water
155 with lower concentrations of Ca, Mg, more closely associated with *Sphagna* (most typically *S.*
156 *denticulatum*) and occasional *Sarmentypnum exannulatum*.

157 Discussion

158 The *Scorpidium* habitats in Wales fit within the habitat parameter ranges reported for the species
159 (Table 1) and, like in Scandinavia (Kooijman & Hedenäs, 1991), the habitats of *S. cossonii* and *S.*
160 *revolvens* differ from each other in their water chemistry. The latter is further reflected in that many
161 Welsh group 1 sites are associated with historic lime quarrying activities and can be large in extent
162 (0.06-0.08 ha) while Welsh Group 2 sites tend to be natural and typically small in extent (0.0004-0.008
163 ha). Interestingly, all three *Scorpidium* species occur at higher pH and sometimes EC (*S. revolvens*) in
164 Wales than in Sweden. Our information cannot fully explain this discrepancy, but several factors can
165 potentially be involved. The Swedish localities have a somewhat lower buffering capacity than the
166 Welsh ones (e.g., less Ca and HCO₃⁻, Table 2). Less well-buffered *Scorpidium* sites are more sensitive
167 to acidification (Kooijman, 2012) and potentially eutrophication (e.g., in relation to associated P-
168 availability as highlighted by Mettrop *et al.*, 2018) and possibly less well-buffered Welsh sites have
169 vanished due to higher atmospheric pollution levels in Wales than in Sweden. If the sites in Wales and
170 Scandinavia have different species compositions, potentially resulting from different species pools or
171 climates, this could also skew the realised niches of the *Scorpidium* species in different directions along
172 the mineral poor to rich gradient (cf., Shaw, 1985; Stieperaere *et al.*, 1997). Differences in geology or
173 genetic set-up of the species (see Hedenäs, 2009; Kophimai, 2013) between the two areas could also
174 affect the habitat preferences of the species. However, studies of ITS and *rp/16* suggest that *S. cossonii*

175 samples from the British Isles do not differ genetically from populations in Scandinavia (Hedenäs,
176 2009). Obviously, more detailed studies are required to find out which explanation or combination of
177 explanations are most likely. The cluster analysis of associated bryophytes shows that *S. cossonii*,
178 together with *S. scorpioides*, and *S. revolvens* belong to two rather different fen moss communities.
179 The respective species associations lend statistical support to earlier observations on the ecology of
180 the *Scorpidium* species in Britain (as summarised by Blockeel 2000). The corresponding water
181 chemistry data suggests that this factor is likely decisive in explaining which species occur in the
182 respective associations. However, the present analysis is limited to Welsh sites and to verify the
183 general validity of the two moss associations identified, the results should be compared with the
184 species growing with *Scorpidium* at sites across a wider geographical area.

185 This study could not demonstrate a statistical difference between moss and flush water for the
186 *Scorpidium* species. In hindsight, we believe that both more frequent and detailed water sampling are
187 required to fully test if temporal and microtopographic site variation explain this lack of difference. An
188 investigation of the water chemistry of *Scorpidium*-rich fens in Finland (Tahvanainen, 2005) found
189 variation in pH only at a microtopographic level (between capillary-held and surface water), and that
190 this variation had a diurnal pattern probably relating to production and consumption of CO₂.

191 **Conclusions**

- 192 • This study confirms the findings of Kooijman & Hedenäs (1991) that two species of *Scorpidium*
193 (*S. cossonii* and *S. revolvens*) occupy different ecological niches in terms of water chemistry
194 and that their methodology is repeatable.
- 195 • The ecology of Welsh *Scorpidium* sites is better understood following statistical cluster
196 analysis of associated bryophytes species.
- 197 • This is the first detailed water chemistry dataset for Wales (and the UK) *Scorpidium* sites and
198 will support environment managers in condition monitoring and conservation of *Scorpidium*
199 sites.

200 **Acknowledgments**

201 The authors would like to thank the following: Sam Bosanquet, Peter S Jones, Jonathan Saville and
202 Julie Creer from Natural Resources Wales. David Schofield, British Geological Survey for funding the
203 chemical analysis and Dr Mark Hill for help with data analysis using spherical k-means. Gareth Farr,
204 Aurelie Devez and Michael Watts publish with the permission of the executive director, British
205 Geological Survey, NERC. Annemieke Kooijman allowed us to use Swedish data collected jointly with
206 Lars Hedenäs. Comments by reviewers and A. Vanderpoorten significantly improved the paper.

207 **Declaration of interest statement**

208 No potential conflict of interest was reported by the authors.

209 **Funding details**

210 This work is in part the result of an initial commercial project for Natural Resources Wales
211 supplemented with British Geological Survey (Welsh office) science funding for additional chemical
212 analysis. The authors contributed their own time for additional survey, sampling and preparation of
213 this manuscript.

214 **References**

215 **Antrop, M.** 2004. Landscape change and the urbanization process in Europe. *Landscape and Urban*
216 *Planning* 67, 9-26.

217

218 **Blockeel, T. L.** 2000. The identification of *Drepanocladus revolvens* and *D. cossonii*, and their
219 distribution in Britain and Ireland. *Bulletin of the British Bryological Society*, **75**, 32-40.

220

221 **Ceschin S., Aleffi M., Bisceglie S., Savo V., Zuccarello V.,** 2012. Aquatic bryophytes as ecological
222 indicators of water quality in the Tiber basin, Italy. *Ecological Indicators* 14(1), 74-81.

223

224 **Covuer et al.,** 2016. Factors affecting the presence and the diversity of bryophytes in the petrifying
225 sources habitat (7220) in Wallonia and the Brussels Capital Region, Belgium.

226

227 **Dell Inc.** 2015. Dell Statistica (data analysis software system), version 13. software.dell.com.

228

229 **Farr, G., Graham, J & C, Stratford, C.** 2014 *Survey, characterisation and condition assessment of*
230 *Palustriella dominated springs 'H7220 petrifying springs with tufa formation (Cratoneurion) in*

- 231 *Wales*. NERC, 211pp. (Natural Resources Wales Evidence Report No. 136, WL/NEC03832/13_14/T6,
232 OR/14/043) (Unpublished).
- 233 **Graham, J & Farr, G.** 2014 Petrifying springs in Wales. *Field Bryology* (112). 19-29.
- 234 **Hedenäs, L.** 1989. The genera *Scorpidium* and *Hamatocaulis*, gen. nov. in northern Europe.
235 *Lindbergia*, **15**, 8-36.
- 236
- 237 **Hedenäs L.** 1999. Altitudinal distribution in relation to latitude; with examples among wetland
238 mosses in the Amblystegiaceae. *Bryobrothera*, **5**, 99-115.
- 239
- 240 **Hedenäs, L.** 2003a. The European species of the *Calliergon-Scorpidium-Drepanocladus* complex,
241 including some related or similar species. *Meylania* (Newsletter of the Swiss association of Bryology
242 and Lichenology), **28**, 1-116.
- 243
- 244 **Hedenäs, L.** 2003b. Amblystegiaceae (Musci). *Flora Neotropica Monograph*, **89**, 1-107.
- 245
- 246 **Hedenäs, L.** 2009. Relationships among arctic and non-arctic haplotypes of the moss species
247 *Scorpidium cossonii* and *Scorpidium scorpioides* (Calliergonaceae). *Plant Systematics & Evolution*,
248 **277**, 217-231.
- 249
- 250 **Hedenäs L.** 2014. *Calliergonaceae*. In: *Flora of North America* Editorial Committee, ed. *Flora of North*
251 *America, north of Mexico*. Volume 28. Bryophyta, Part 2. New York & Oxford: Oxford University
252 Press, pp. 384-403.
- 253
- 254 **Hedenäs, L. & Kooijman, A.** 1996. Förändringar i rikkärret SV om Mellansjön (Mellanslättsjön) i
255 Västergötland. *Svensk Botanisk Tidskrift* 90: 113-121.
- 256
- 257 **Joosten, H., Couwenberg, J., Moen, A. & Tanneberger, F.** 2017. Mire and peatland terms and
258 definitions in Europe. In: Joosten, H. Tanneberger, F. & Moen, A. (eds). *Mires and peatlands of*
259 *Europe*. Stuttgart: E. Schweizerbart'sche Verlagsbuchhandlung (Nägele u. Obermiller), pp. 65-96.
- 260
- 261 **Hill, M.O., Harrower, C.A. & Preston, C.D.** 2013. Spherical k-means clustering is good for interpreting
262 multivariate species occurrence data. *Methods in Ecology and Evolution*, **4**, 542-551.
- 263
- 264 **Kaplan, J.O., Krumhardt, K.M. & Zimmermann, N.** 2009. The prehistoric and preindustrial
265 deforestation of Europe. *Quaternary Science Reviews* 28, 3016-34.
- 266
- 267 **Kooijman, A.** 2012. 'Poor rich fen mosses': atmospheric N-deposition and P-eutrophication in base-
rich fens. *Lindbergia* 35, 42-52.
- 268
- 269 **Mettrop, I.S., Neijmeijer, T., Cusell, C., Lamers, P.M., Hedenäs, L. & Kooijman, A.** 2018. Calcium and
iron as key drivers of brown moss composition through differential effects on phosphorus
270 availability, *Journal of Bryology*.
- 271
- 272 **Kooijman, A.** 1992. The decrease of fen rich bryophytes in the Netherlands. *Biological Conservation*,
59, 139-143.
- 273
- 274 **Kooijman, A. & Hedenäs, L.** 1991. Differentiation in habitat requirements within the genus
275 *Scorpidium*, especially between *S. revolvens* and *S. cossonii*. *Journal of Bryology*, **16**, 619-627.
- 276

277 **Kooijman A.M., Beltman B. & Westhoff V.** 1994 Extinction and reintroduction of the
278 bryophyte *Scorpidium scorpioides* in a rich-fen spring site in the Netherlands. *Biological*
279 *Conservation*, **69**, 87-96
280

281 **Kooijman, A.M & Westhoff, V.** 1995. Variation in habitat factors and species composition of
282 *Scorpidium scorpioides* communities in NW-Europe. *Vegetatio*, **117**, 133-150
283

284 **Kooijman, A.M.** 2012. 'Poor rich fen mosses': atmospheric N-deposition and P-eutrophication in
285 base-rich fens. *Lindbergia*, **35**, 42-52.
286

287 **Kophimai Y.** 2013. *Population genetics of the fen specialist moss Scorpidium cossonii in northeastern*
288 *Switzerland* (PhD thesis). Bern: Universität Bern.
289

290 **Paulissen, M.P.C.P., Bobbink, R., Robat, S.A. & Verhoeven, J.T.A.** 2016. Effects of Reduced and
291 Oxidised Nitrogen on Rich-Fen Mosses: a 4-Year Field Experiment. *Water Air Soil Pollut*, **227**, 18 (p. 1-
292 14).

293 **Price T.D. (ed.).** 2000. *Europe's first farmers*. Cambridge: Cambridge University Press.

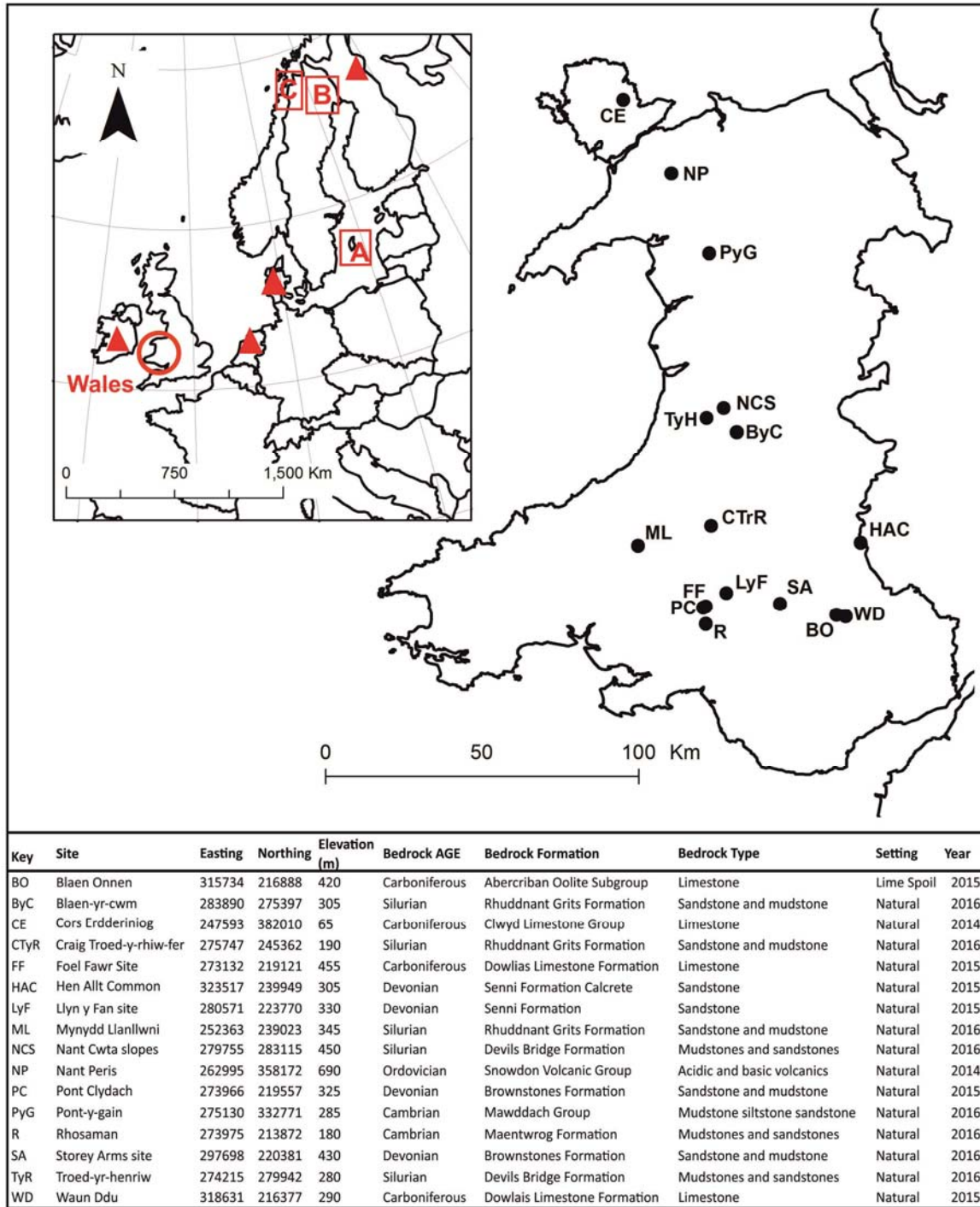
294 **Rydin, H., Sjörs, H. & Löfroth, M.** 1999. Mires. *Acta Phytogeographica Suecica* 84, 91-110. **Shaw, J.**
295 1985. The relevance to ecology of species concepts in bryophytes. *Bryologist* 88, 199-206.

296 **Stieperaere, H., Heylen, O. & Podoor, N.** 1997. Differences in species composition of the bryophyte
297 layer of some Belgian and Dutch pinewoods with and without the invading hepatic *Lophocolea*
298 *semiteres* (Lehm.) Mitt. *Journal of Bryology* 19, 425-434.

299 **Tahvanainen, T.** 2004. The growth of *Scorpidium revolvens* in relation to calcium and magnesium.
300 *Lindbergia*, **29**, 123-128.

301 **Tahvanainen, T.** 2005. *Diversity of water chemistry and vegetation of mires in the Kainuu region,*
302 *middle boreal Finland*. PhD dissertations in biology No. 33, University of Joensuu.
303

304 **Vitt, D.H, Wirdum, G.V, Halsey, L & Zoltai, S.** 1993. The Effects of Water Chemistry on the Growth of
305 *Scorpidium scorpioides* in Canada and The Netherlands. *Bryologist*, **96**, 106-111
306
307
308
309
310
311
312
313
314
315
316
317
318
319
320
321
322
323



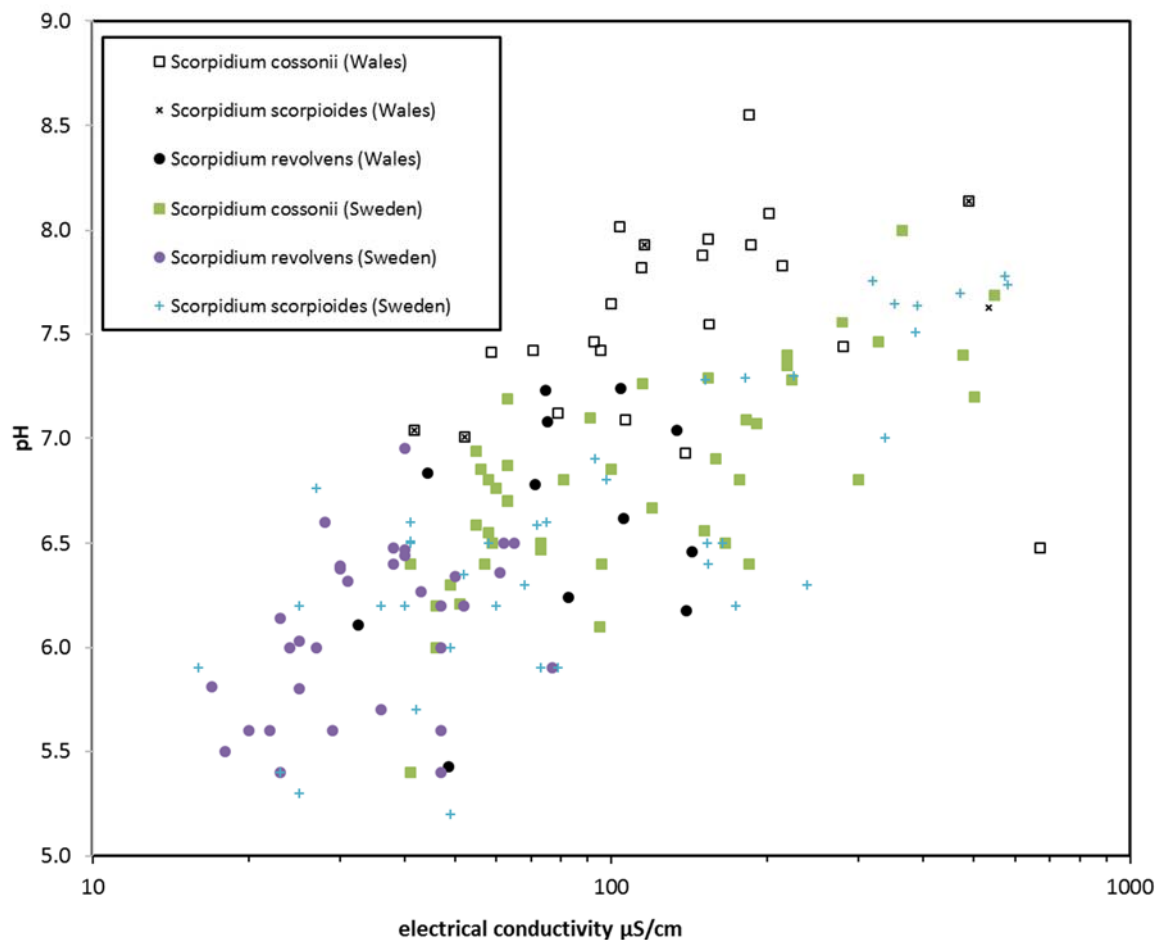
324
325

326 **Figure 1.** Sample locations: Welsh sites, elevation, geology, setting and sampling year. Inset: previous
327 studies in Sweden represented by squares labelled A, B and C (Koojiman & Hedenäs, 1991) and in
328 Republic of Ireland, Netherlands and Finland by triangles (Koojiman & Westhoff, 1995). Contains
329 Ordnance Survey Data © Crown Copyright and database rights 2018.

330

331

332



333

334 **Figure 2 – Relationship between pH and EC between Welsh and Swedish analysis. Swedish data**
 335 **collected between 1990-1997 (from Kooijman & Hedenäs 1991; plus unpublished data from**
 336 **different parts of Sweden by LH and A. Kooijman), Welsh data from 2014-2015.**

337

338

339

340

341

342

343

344

345

346

347

348

349

350

351

352

353

354

355

356

357

358 Table 1
359

Table 1. Habitat preferences and geographical ranges for *Scorpidium* species based on Blockeel (2000) and Hedenäs (1999, 2003a, 2003b, 2014). IR (Ionic Ratio), an indicator of water type = $2 [Ca^{2+}]/2 [Ca^{2+}] + [Cl]$ (van Wirdum 1991).

Species	Altitude range (m aOD)	Habitat	pH	EC ($\mu\text{S}/\text{cm}$)	Ca ²⁺ (mg/L)	IR	Geographical range
<i>S. scorpioides</i>	Global: 0–5100 Sweden 293 (mean) Wales 398 (mean)	Calcium-rich fens (pools & lake shores)	5.2–8.5	14–582	1.2–141.0	0.34–1.00	Widespread (often common) in temperate to arctic areas of northern hemispheres (and in the Andes and a few places in Australia)
<i>S. cossonii</i>	Global: 0–4500 Sweden 264 (mean) Wales 362 (mean)	Calcium-rich fens (springs, periodically water-filled depressions & lake shores)	5.0–8.1	18–681	2.3–130	0.42–1.0	Widespread (often common) in temperate to sub-polar areas of both hemispheres (and in the Andes)
<i>S. revolvens</i>	Global: 0–3100 Sweden 305 (mean) Wales 358 (mean)	More oligotrophic fens (often spring-influenced) but absent from calcium-rich sites	5.1–7.1	16–166	0.7–27.7	0.17–1.0	Widespread (often common) in temperate to sub-polar areas of both hemispheres (and in Papua New Guinea)

360
361
362
363 Table 2

Table 2. Comparison of EC, pH, Ca²⁺, and HCO₃⁻ in moss water of the three *Scorpidium* species in Wales and Sweden, respectively. Statistically significant differences between the areas are indicated by **: 0.01 > p > 0.001; ***: p < 0.001.

Region	Species	EC, $\mu\text{S cm}^{-1}$		pH		Ca ²⁺ , mg L ⁻¹		HCO ₃ ⁻ , mg L ⁻¹	
		N	Mean (S.E.)	N	Mean (S.E.)	N	Mean (S.E.)	N	Mean (S.E.)
Wales	<i>S. cossonii</i>	23	168 (30)	23	7.6 (0.1)***	23	34.7 (5.0)	23	97 (14)
Sweden	<i>S. cossonii</i>	44	150 (19)	44	6.8 (0.1)	53	26.3 (3.3)	53	67 (12)
Wales	<i>S. revolvens</i>	12	88 (11)***	12	6.6 (0.2)**	13	7.2 (2.0)	13	18 (6)
Sweden	<i>S. revolvens</i>	32	38 (3)	32	6.1 (0.1)	32	6.2 (0.6)	32	10 (2)
Wales	<i>S. scorpioides</i>	6	223 (92)	6	7.5 (0.2)**	6	34.3 (14.5)	6	101 (45)
Sweden	<i>S. scorpioides</i>	39	155 (25)	39	6.6 (0.1)	39	31.4 (6.5)	39	84 (17)

364
365
366
367 Table 3

	<i>Scorpidium cossonii</i> (n=23)		<i>Scorpidium revolvens</i> (n=12)		<i>Scorpidium cossonii</i> - <i>Scorpidium revolvens</i>	<i>Scorpidium cossonii</i> - <i>Scorpidium revolvens</i>
	Av	SE	Av	SE	SE	t
EC, $\mu\text{S}/\text{cm}$	167.50	30.17	88.02	10.87	14.54	5.78
HCO ₃ ⁻ , mg/l	96.78	13.34	12.64	5.79	0.19	5.19
Ca, mg/l	34.69	5.02	7.05	2.19	5.48	5.04
Cl, mg/l	12.45	1.43	11.18	1.76	0.34	2.5
pH	7.57	0.10	6.60	0.16	32.06	2.48
Na, mg/l	6.82	0.74	6.75	1.09	1.15	1.55
SO ₄ ²⁻ , mg/l	3.89	0.74	2.10	0.89	2.26	0.56
SiO ₂ , mg/l	3.53	0.41	3.55	0.33	0.39	0.54
Mg, mg/l	2.41	0.29	1.56	0.18	1.32	0.05
K, mg/l	2.20	0.27	2.49	0.86	0.25	-0.04
Si, mg/l	1.65	0.19	1.66	0.15	0.52	-0.04
NO ₃ ⁻ , mg/l	0.55	0.38	0.34	0.09	0.29	-0.17
Mn, mg/l	0.33	0.14	0.38	0.26	0.9	-0.33

368
369
370
371
372
373

Site Name	Total number of samples	<i>Bryum pseudotriquetrum</i>	<i>Calliergonella cuspidata</i>	<i>Campyllum stellatum</i>	<i>Cratoneuron filicinum</i>	<i>Ctenidium molluscum</i>	<i>Fissidens adianthoides</i>	<i>Palustriella commutata</i>	<i>Palustriella falcata</i>	<i>Philonotis calcaria</i>	<i>Philonotis fontana</i>	<i>Scorpidium cossonii</i>	<i>Scorpidium scorpioides</i>	<i>Sphagnum papillosum</i>	<i>Scorpidium revolvens</i>	<i>Aneura pinguis</i>	<i>Breutelia chrysocoma</i>	<i>Hylocomium splendens</i>	<i>Sarmentypnum exannulatum</i>	<i>Sphagnum denticulatum</i>	<i>Sphagnum fallax</i>	<i>Sphagnum subnitens</i>	<i>Sphagnum teres</i>	<i>Straminergon stramineum</i>
Cors Erdderiniog Site 7e	4			1								1	1			2								
Foel Fawr site 1	7					1	1		1				1											
Nant Cwta slopes (Eisteddfa Gurig)	5	1					1					1			1	2								
Storey Arms site 1	3						1					1	1											
Storey Arms site 2	3			1								1	1											
Blaen Onnen	23	1	1	1	1			1	1	1	1	1				2								
Foel Fawr site 2	10	1		1					1	1		1	1											
Llyn y Fan site 1	12	1		1				1	1			1				2								
Llyn y Fan site 2	7							1	1		1	1				2								
Pont Clydach site 1	8	1		1											2	2				2	2			
Pont Clydach site 3	10	1		1					1			1	1											
Waun Ddu	17	1	1	1	1		1		1			1								2	2			
Blaen-yr-cwm	2														2			2						
Craig Troed-y-rhiw-fer	4														2	2	2						2	
Mynydd Llanllwni site 1	3														2	2			2					
Mynydd Llanllwni site 2	4			1											2				2	2				
Mynydd Llanllwni site 4	6			1											2	2			2	2				
Nant Peris site 1	4											1			2	2								2
Nant Peris site 3	2														2	2								
Pont-y-gain	3														2	2							2	
Troed-yr-henriw	4			1											2	2								

		pH	EC	Ca	Mg	Na	K	HCO ₃ ⁻	Cl ⁻	SO ₄ ²⁻	NO ₃ ⁻	Si	SiO ₂	Mn
			µS/cm	mg/l	mg/l	mg/l	mg/l	mg/l	mg/l	mg/l	mg/l	mg/l	mg/l	mg/l
Group 1 characterised by <i>Scorpidium cossonii</i>	Min	5.3	26	0.7	0.3	2.5	0.2	5	3.8	1	0.2	0.2	0.4	0
	Max	11.8	787	98	9.7	113.7	7	261	201.4	17.6	9.2	5	10.6	11.7
	Mean	7.5	197	35.2	2.8	11.2	2.2	101	19.6	4.4	0.8	1.8	3.9	0.5
Group 2 characterised by <i>Scorpidium revolvens</i>	Min	5.1	33	0.3	0.4	3.8	0.2	5	5.4	1	0.2	0.4	0.8	0
	Max	7.2	143	26	2.7	16.1	10.7	68	23.2	10	1.3	4.4	9.5	0.4
	Mean	6.3	79	5	1.4	6.4	2.3	10	10.2	3.6	0.7	1.7	3.5	0.1