The stable isotope composition of organic and inorganic fossils in lake sediment records: current understanding, challenges, and future directions.

van Hardenbroek M [a,*], Chakraborty A[b], Davies KL [c], Harding P [d], Heiri O [e], Henderson ACG [a], Holmes JA [d], Lasher GE [g], Leng MJ [h,i] , Panizzo VN [j], Roberts L [d], Schilder J [k], Trueman CN [l], Wooller MJ [m,n]

[a] School of Geography Politics and Sociology, Newcastle University, Newcastle-upon-Tyne, NE1 7RU, UK
[b] Birbal Sahni Institute of Palaeosciences, Lucknow, 226007, India
[c] School of Geography, Earth and Environmental Sciences, Plymouth University, Plymouth, PL4 8AA, UK
[d] Environmental Change Research Centre, Department of Geography, University College London, London, WC1E 6BT, UK
[e] Institute of Plant Sciences and Oescher Centre for Climate Change Research, University of Bern, CH-3013 Bern, Switzerland
[f] Geoecology, Department of Environmental Sciences, University of Basel, Klingelbergstrasse 27, CH-4056 Basel, Switzerland
[g] Department of Earth and Planetary Sciences, Northwestern University, Evanston, IL 60208, USA
[h] NERC Isotope Geosciences Facility, British Geological Survey, Nottingham, NG12 5GG, UK
[i] Centre for Environmental Geochemistry, School of Biosciences, Sutton Bonington Campus, University of Nottingham, Loughborough, LE12 5RD, UK
[j] Centre for Environmental Geochemistry, School of Geography, University of Nottingham, University Park, Nottingham, NG7 2RD, UK
[k] Department of Biological and Environmental Science, University of Jyväskylä, PO Box 35, 40014 Jyväskylä, Finland
[l] Ocean and Earth Science, University of Southampton Waterfront Campus, Southampton SO14 3ZH
[m] College of Fisheries and Ocean Sciences, University of Alaska Fairbanks, Fairbanks AK 99775 USA
[n] Alaska Stable Isotope Facility, Water and Environmental Research Center, University of Alaska Fairbanks, Fairbanks, AK 99775, USA

[*] corresponding author: maarten.vanhardenbroek@ncl.ac.uk

Abstract

This paper provides an overview of stable isotope analysis (H, C, N, O, Si) of the macro- and microscopic remains from aquatic organisms found in lake sediment records and their application in (palaeo)environmental science. Aquatic organisms, including diatoms, macrophytes, invertebrates, and fish, can produce sufficiently robust remains that preserve well as fossils and can be identified in lake sediment records. Stable isotope analyses of these remains can then provide valuable insights into habitat-specific

biogeochemistry, feeding ecology, but also on climatic and hydrological changes in and around lakes. Since these analyses focus on the remains of known and identified organisms, they can provide more specific and detailed information on past ecosystem, food web and environmental changes affecting different compartments of lake ecosystems than analyses on bulk sedimentary organic matter or carbonate samples. We review applications of these types of analyses in palaeoclimatology, palaeohydrology, and palaeoecology. Interpretation of the environmental 'signal' provided by taxon-specific stable isotope analysis requires a thorough understanding of the ecology and phenology of the organism groups involved. Growth, metabolism, diet, feeding strategy, migration, taphonomy and several other processes can lead to isotope fractionation or otherwise influence the stable isotope signatures of the remains from aquatic organisms. This paper includes a review of the (modern) calibration, culturing and modeling studies used to quantify the extent to which these factors influence stable isotope values and provides an outlook for future research and methodological developments for the different examined fossil groups.

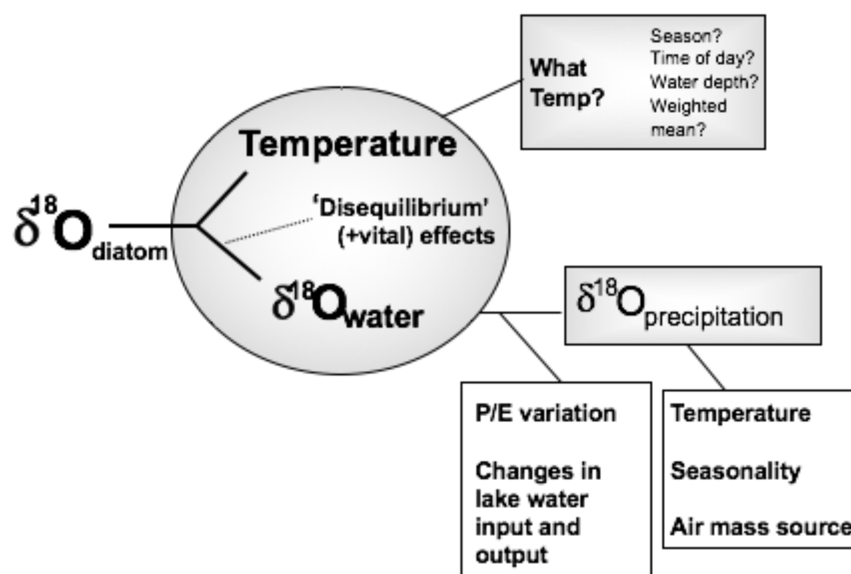
Keywords: Stable isotopes; Lake sediment; Organic remains; Inorganic remains; Diatoms; Invertebrates; Ostracods

1. Introduction

Stable isotope analysis provides a versatile tool for investigating lake sediment records based on the link between stable isotope ratios and a range of environmental and biological processes, including climate change, hydrology, biogeochemical cycling, and consumer-diet interactions in food webs (Leng and Henderson 2013). Stable carbon and nitrogen isotope analysis of bulk sedimentary organic matter (SOM) is often used in palaeoenvironmental records as SOM is easy to sample and relatively straightforward to measure (Meyers et al. 2001). Some sediments require chemical pre-treatment to remove carbonates, which in itself can affect the stable isotope composition of SOM (Brodie et al. 2011a, b). The information provided by SOM is always an integrated signal of catchment and in-lake processes, which can make it hard to interpret variations in its isotope composition. For example, an understanding of changes in $\delta^{13}C$ and $\delta^{15}N$ values of SOM

77 requires detailed information from the catchment as they are dependent on, amongst a
78 range of factors, the composition and amount of input from terrestrial vegetation,
79 anthropogenic nutrient input, lake volume, littoral-to-profundal ratio of the lake basin,
80 productivity, groundwater inputs and stratification (e.g., Meyers and Ishiwatari 1993).
81 Stable isotopes of endogenic (bulk) carbonates are also widely used in
82 palaeohydrology and palaeoclimatology. Changes in mineral $\delta^{18}O$ values are interpreted in
83 terms of changes in precipitation and temperature (Leng and Marshall 2004, Leng and
84 Barker 2006). The $\delta^{13}C$ values of endogenic carbonates are a complex sum of processes
85 taking place in the lake and its catchment, including temperature and productivity-
86 dependent fractionation, dissolved inorganic carbon (DIC) inflow, methanogenesis and
87 methane oxidation, CO_2 dissolution and outgassing and lake stratification (Siegenthaler
88 and Eicher 1986; Hollander and Smith 2001; Leng and Marshall 2004; Schwalb et al. 2013).
89 To complement stable isotope data derived from analyses of SOM or carbonates, it has
90 become more common to analyse the stable isotope composition of identifiable fossil
91 remains separated and manually picked from lake sediments (e.g. diatoms, invertebrates,
92 plant macrofossils), or to analyse stable isotopes of specific compounds chemically
93 isolated from the sediments (e.g., lipids, amino acids, pigments). These approaches offer
94 the great benefit of targeting specific organism groups or chemical biomarkers, which can
95 reflect particular habitats or locations in a lake, or provide valuable information about
96 their functional roles in an ecosystem. Targeting specific remains and compounds also
97 means that it is easier to understand and test how the stable isotope composition is
98 affected by biogeochemical and taphonomic processes over time.
99 In this review, we provide an overview of approaches based on stable isotopes
100 measured on taxon-specific samples. We include the isotope systems H, C, N, O, Si,
101 measured on diatoms, calcareous and chitinous invertebrate remains, fish remains, and
102 plant macrofossils. We discuss their palaeolimnological applications and provide an
103 overview of the current understanding of taphonomy and ecology that is required to
104 interpret sedimentary records.
105
106 **2. Diatoms**
107

108 Diatoms are unicellular, eukaryotic, micro-organisms, which are ubiquitous in nature. As
109 such, diatom silica is an interesting sediment component for isotope measurements in
110 lake (Leng and Barker 2006) and marine (Swann and Leng 2009) environments, where the
111 oxygen ($\delta^{18}O_{diatom}$), silicon ($\delta^{30}Si_{diatom}$), carbon ($\delta^{13}C_{diatom}$) and nitrogen ($\delta^{15}N_{diatom}$) isotope
112 compositions can all be used as proxies for environmental change. $\delta^{18}O_{diatom}$ tends to be
113 used as a measure of temperature/water composition variation (Fig. 1), $\delta^{30}Si_{diatom}$ as a
114 proxy for nutrient availability and utilisation (Fig. 2), and $\delta^{13}C_{diatom}$ and $\delta^{15}N_{diatom}$ for
115 nutrient cycling/source investigation (Leng and Henderson 2013).



116

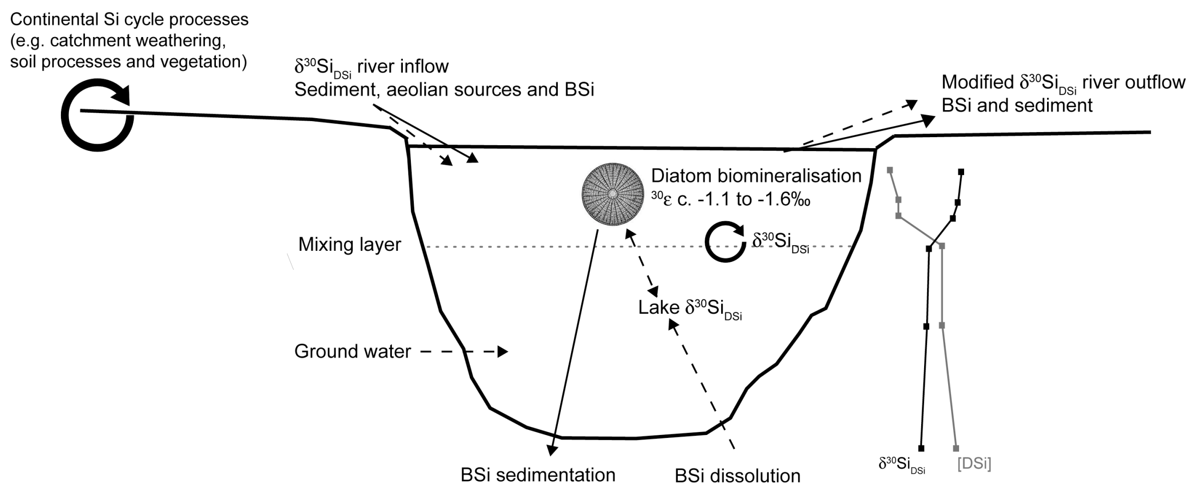
117 **Fig. 1**: *Controls on the stable oxygen isotope composition of biogenic silica*

118

119 Compared to $\delta^{18}O_{diatom}$ studies, the application of $\delta^{30}Si_{diatom}$, $\delta^{13}C_{diatom}$ and $\delta^{15}N_{diatom}$
120 techniques are in their relative infancy, with only a few studies being applied in lacustrine
121 systems (Alleman et al. 2005; Street-Perrott et al. 2008; Swann et al. 2010; Hurrell et al.
122 2011; Hernández et al. 2011, 2013; Opfergelt et al. 2011; Chen et al. 2012; Barker et al.
123 2013; Cockerton et al. 2015; Webb et al. 2016; Panizzo et al. 2016, 2018a, b). There are
124 several reviews of the use of isotopes in biogenic (including diatom) silica (including Leng
125 and Barker 2006; Swann and Leng 2009; Leng et al. 2009; Leng and Henderson 2013;
126 Sutton et al. 2018), which highlight, in detail, the many issues associated with such
127 analyses. These will be further elucidated upon here and include issues such as equilibrium
128 fractionation, sample purification, post mortem maturation of diatom silica, and

129 standardisation and inter-laboratory calibrations. Given that more recent advances have
130 been made in $\delta^{30}Si_{diatom}$ applications in lacustrine settings, in addition to the body of
131 literature on $\delta^{18}O_{diatom}$, we focus this review on these two proxies.

132 Diatom silica is a structurally complex mineral for $\delta^{18}O_{diatom}$ (see section 2.4 below)
133 measurement due to a hydrous component. O isotope extraction generally adopts
134 fluorination (offline) techniques, with measurement via gas-source isotope ratio mass
135 spectrometry (IRMS), a technique which can also be used to measure $\delta^{30}Si_{diatom}$ (Leng and
136 Sloane 2008). However, the use of multi-collector inductively-coupled-plasma mass
137 spectrometry (MC-ICP-MS) is becoming increasingly more dominant for $\delta^{30}Si_{diatom}$ analyses
138 due to the reduced sample size needed (~ 1 mg), while carbon and nitrogen (for $\delta^{13}C_{diatom}$
139 and $\delta^{15}N_{diatom}$) are measured on very small quantities of organic material hosted
140 (occluded) within the structure (Webb et al. 2016).
141



142
143

144 **Fig. 2**: *A schematic drawing of Si cycling in a lake system; solid arrows correspond to*
145 *particulate fluxes and dashed lines to dissolved phases or transformation processes. The*
146 *range in fractionation factors ($^{30}\varepsilon$) associated with the production of biogenic silica (in this*
147 *case, diatoms) from freshwater archives are also provided. Typical water column profiles*
148 *(for both lacustrine and oceanic settings) of DSi concentrations [DSi] and $\delta^{30}Si_{DSi}$ (‰)*
149 *signatures are drawn, highlighting the effects of biological uptake in surface waters (e.g.,*
150 *lower [DSi] and higher $\delta^{30}Si_{DSi}$). Reference to water column mixing is also made, which*
151 *depending on the limnological characteristics of individual sites, can compositionally*
152 *"reset" surface water $\delta^{30}Si_{DSi}$ (e.g., on a seasonal basis).*

153

154 Silicon has three naturally occurring stable isotopes ^{28}Si, ^{29}Si and ^{30}Si with a mean
155 abundance of 92.2%, 4.7% and 3.1% respectively. The isotope composition of any sample
156 (*x*), for ^{29}Si or ^{30}Si (*n*), is expressed in delta notation (δ), compared to the reference
157 standard NBS28, using the following equation:

158 $$\delta^{n}Si_{x}\ (‰) = ([(^{n}Si/^{28}Si)_{x} - (^{n}Si/^{28}Si)_{standard}]/[(^{n}Si/^{28}Si)_{standard}]) \times 1000$$

159 An overview of the key processes affecting $\delta^{30}Si$ in lakes are shown in Fig 2. These include
160 processes in a catchment (weathering, soil development, vegetation cover) and in a lake
161 (productivity, water supply, stratification, sedimentation, dissolution) and will be
162 discussed in more detail below.

163

164 *2.1 Interpreting stable oxygen and silicon isotopes of diatom opal*

165

166 $\delta^{18}O_{diatom}$ has been used as a proxy for climate and hydrological change in many studies
167 (Leng and Barker 2006) and the mineral-water fractionation has been estimated
168 previously from analyses of diatoms from freshwater (and marine) sediments, coupled
169 with estimates of the temperatures and isotope compositions of coexisting waters during
170 silica formation (Labeyrie 1974; Juillet-Leclerc and Labeyrie 1987; Matheney and Knauth
171 1989). There are very few calibration studies (e.g., Labeyrie and Juillet 1982; Wang and
172 Yeh 1985; Juillet-Leclerc and Labeyrie 1987; Shemesh et al. 1995), and published estimates
173 of the average temperature dependence for typical ocean temperatures range from –0.2
174 to –0.5 ‰ per ^{o}C (Juillet-Leclerc and Labeyrie 1987; Shemesh et al. 1992; Brandriss et al.
175 1998). However, in lake studies the oxygen isotope composition of diatom silica is often
176 more sensitive to changes in “non-temperature” aspects of climate, such as amount or
177 source of precipitation (Barker et al. 2001; Shemesh et al. 2001) and evaporation
178 (Hernández et al. 2008).

179 In a pioneering study, Schmidt et al. (1997) suggested that there is no regular
180 correlation between temperature and the oxygen isotope fractionation between modern
181 diatoms and the water in which they biomineralise. This led to the hypothesis that the
182 temperature-dependent oxygen isotope fractionation preserved in biogenic opaline
183 sediments may, in some environments, have been established during diagenesis (see

184 below) rather than acquired during growth, a subject open to recent investigation
185 (Menicucci et al. 2017; Tyler et al. 2017).

186 $\delta^{30}Si_{diatom}$ is a relatively underused technique in lacustrine systems, with few studies
187 published despite silicon cycling in lakes being a key component of the continental silicon
188 cycle. However, the field is growing, with contemporary studies examining the $\delta^{30}Si$
189 signature of lake surface waters and diatom opal as a means to validate the technique for
190 palaeolimnological applications (Opfergelt et al. 2011; Panizzo et al. 2016)**.** The method
191 has traditionally been more widely applied in oceanographic settings as a means to
192 reconstruct past biogeochemical cycling, with conventional interpretation of the method
193 as a diatom silica utilisation or water mass/circulation proxy, the latter particularly when
194 coupled with $\delta^{13}C_{diatom}$ and $\delta^{15}N_{diatom}$ or $\delta^{30}Si$ from other silicifying organisms (i.e. sponge
195 spicules and radiolarians) (Abelmann et al. 2015; Beucher et al. 2007; De La Rocha et al.
196 1998; Hendry and Brzezinski 2014; Hendry et al. 2016; Horn et al. 2011; Maier et al. 2013;
197 Panizzo et al. 2014).

198 Lake water dissolved silica (DSi) $\delta^{30}Si$ signatures ($\delta^{30}Si_{DSi}$) are essentially a product of
199 upstream catchment processes (e.g., weathering, erosion and soil processes, and
200 vegetation), regulated by regional climate systems, and within-lake biogeochemical
201 processes (e.g., diatom utilisation and dissolution; see section 2.2) (De la Rocha et al.
202 2000; Frings et al. 2016; Opfergelt and Delmelle 2012; Panizzo et al. 2018b). In instances of
203 more extreme climatic variability, particularly in tropical catchments, lake water budgets
204 can be considerably altered so that there are large opposing (seasonal) variations in
205 overall lake water DSi concentrations and $\delta^{30}Si_{DSi}$ signatures (with increasing compositions
206 of ~0.5 ‰ during dry season periods; Cockerton et al. 2013). This can be further
207 compounded at sites where groundwater inputs are considerable (Street-Perrott et al.
208 2008). Within-lake biomineralisation can significantly alter the outflowing concentration
209 and composition of DSi and $\delta^{30}Si_{DSi}$ (respectively). As such, lakes have been found to act as
210 a key buffer within the continental Si cycle (in addition to the soil-vegetation system)
211 (Frings et al. 2014).

212 Studies of $\delta^{30}Si_{DSi}$ and $\delta^{30}Si$ of diatom opal ($\delta^{30}Si_{diatom}$) therefore act as a tracer of
213 biogeochemical processes in nature. Early studies were conducted at lake sites with basalt
214 and volcanic geology, where Si weathering, riverine DSi fluxes and biomineralisation
215 potential are high (e.g., Alleman et al, 2005; Street-Perrott et al. 2008; Opfergelt et al.

216 2011; Chen et al. 2012; Cockerton et al. 2015). In lacustrine systems these techniques are
217 applied to investigate variations in water column mixing (e.g. DSi supply) and diatom
218 bloom duration (e.g. DSi utilisation), which are all regulated by intrinsic (e.g. water column
219 mixing, stratification, ice-cover duration) and extrinsic processes (e.g. climate and riverine
220 source water changes) (Fig. 2; Alleman et al. 2005; Street-Perrott et al. 2008; Opfergelt et
221 al. 2011; Panizzo et al. 2016, 2017, 2018a, b).
222
223 *2.2 Fractionation and vital effects of diatom opal*
224
225 As with other organisms, diatoms are assumed to be precipitated in isotope equilibrium as
226 predicted by thermodynamic fractionation. However, it has been widely shown in
227 carbonates that offsets from oxygen (and carbon) isotope equilibrium may arise in
228 response to variations in kinetic or metabolic processes within and between individual
229 taxa, e.g., changes in growth rates, nutrient availability or rates of calcification/silicification
230 (Duplessy et al. 1970; Wefer and Berger 1991; Spero and Lea 1993, 1996; Spero et al. 1997;
231 Bemis et al. 1998). For biogenic carbonates, such as ostracods (see section 3 below), the
232 impact of vital effects can be overcome by selecting species-specific samples for isotope
233 analysis. This is not feasible for diatoms due to their small size. A number of culture (Binz
234 1987; Brandriss et al. 1998; Schmidt et al. 2001), sediment trap (Moschen et al. 2005) and
235 down core studies (Sancetta et al. 1985; Juillet-Leclerc and Labeyrie 1987; Shemesh et al.
236 1995; Bailey et al. 2014) in lacustrine (and marine) systems do not show any oxygen
237 isotope vital effect exists in diatoms. While data in Brandriss et al. (1998) display a 0.6 ‰
238 difference between two laboratory cultured diatom taxa and Shemesh et al. (1995) found
239 a 0.2 ‰ offset between two different size fractions of diatoms, offsets of this magnitude
240 are within the range of reproducibility routinely achieved when analysing $\delta^{18}O_{diatom}$.
241 Early studies on the fractionation of diatom opal during valve dissolution suggested a
242 –0.55 ‰ enrichment of $\delta^{30}Si_{diatom}$ (Demarest et al. 2009) although this has since been
243 challenged, to suggest an absence of dissolution fractionation effects, based on analyses
244 of marine (Wetzel et al. 2014) and lacustrine sedimentary diatoms (Panizzo et al. 2016).
245 More complex, and still in dispute to a certain degree, are the isotope fractionations
246 associated with diatom biogenic silica production. When diatoms take up DSi (in the form
247 of silicic acid, $Si(OH)_4$) during biomineralisation they actively discriminate against the

248 heavier (^{30}Si, ^{29}Si) isotopes in favour of the lighter isotope (^{28}Si), leading to the enrichment
249 of the residual pool ($\delta^{30}Si_{DSi}$) (De La Rocha et al. 1997). Evidence from *in-vitro* and *in-situ*
250 studies from marine diatoms suggests that this per mille enrichment factor ($^{30}\varepsilon_{uptake}$) is
251 independent of temperature, pCO_2 and nutrient availability (De La Rocha et al. 1997;
252 Fripiat et al. 2011; Milligan et al. 2004; Varela et al. 2004). The general consensus for
253 $^{30}\varepsilon_{uptake}$ is −1.1 ± 0.4 ‰ (refer to Frings et al. 2016 for the latest compilation of data) with
254 good agreement with more recent *in-situ,* contemporary studies from freshwater diatoms
255 (published values ranging between −1.1 and −1.6 ‰; Alleman et al. 2005; Opfergelt et al.
256 2011; Panizzo et al. 2016; Sun et al. 2013) (Fig. 2). However, Sutton et al. (2013) have
257 challenged this consensus reporting species-dependent $^{30}\varepsilon_{uptake}$ ranging from −2.09 to
258 −0.54 ‰ (based on cultured polar and sub-polar marine diatom strains). While such
259 evidence has not (to date) been replicated, these studies highlight the ongoing challenges
260 in applying $\delta^{30}Si$ approaches to down core reconstructions and reinforce the value of
261 modern day, lake site-specific calibrations of the method (as per Panizzo et al. 2016).
262
263 *2.3 Sample purification*
264
265 Much effort is placed on diatom purification prior to isotope analysis (e.g., van Bennekom
266 and van der Gaast 1976; Shemesh et al. 1995; Schleser et al. 2001; Morley et al. 2004;
267 Rings et al. 2004; Lamb et al. 2005; Tyler et al. 2007; Brewer et al. 2008; Mackay et al.
268 2011) as high sample purity is required for $\delta^{18}O_{diatom}$ and $\delta^{30}Si_{diatom}$ analysis. This is because
269 oxygen and silicon are both common elements in many other components found in lake
270 sediments (including clay, silt, tephra and carbonates) and a high proportion of these can
271 introduce significant isotopic offsets. $\delta^{13}C_{diatom}$ and $\delta^{15}N_{diatom}$ are usually measured on
272 occluded (within the frustrule) organic matter and therefore external organic matter, a
273 contaminant, is removed using chemical oxidation (for further information consult
274 Robinson et al. (2004), Singer and Shemesh (1995) and references therein).
275 In most instances, standard chemical (oxidation) and physical separation approaches
276 (sieving, heavy density liquids) work well for samples with a high proportion of diatom
277 silica (>10 %). However, more complex and time consuming methods are required to clean
278 relatively diatom poor (<10 %) material, where sample sizes are small or where the
279 contaminant is similar in size and density to the diatom silica. These include SPLITT

280 (gravitational split-flow lateral-transport), micromanipulation, and chemical mass balance
281 modelling. SPLITT is an approach similar to heavy density separation (Giddings 1985),
282 whereby individual particles within a sample are separated under laminar flow of water on
283 the basis of their density, size and shape (Schleser et al. 2001; Rings et al. 2004; Leng and
284 Barker 2006). Micro-manipulation is an alternative approach: where a device is attached
285 to an inverted microscope, with a micro-injector system used to extract individual non-
286 diatom particles from a sample (Snelling et al. 2013). Although time-consuming this
287 technique is routinely used in other fields (e.g., cryptotephra, Lane et al. 2014), and may
288 be the only option, in some cases, to remove contaminants that are chemically and
289 physically similar to diatoms. In instances where all other methods are unsuccessful in
290 removing sample contaminants, mass balance chemical modelling can be applied. Here,
291 whole-rock geochemistry and electron-optical imaging provides a method for the
292 identification, quantification and subsequent removal of (e.g. via an offset correction
293 factor) the effects of different types of contamination (Lamb et al. 2005; Brewer et al.
294 2008; Mackay et al. 2011). This approach can also work with multiple contaminants so
295 long as they are well characterised (Wilson et al. 2014). Purity is routinely demonstrated in
296 publications either visually (e.g., scanning electron microscopy) or quantitatively, via
297 estimations of sample contamination <1 % (e.g., SiO_2:Al_2O_3 <1).
298
299 *2.4 The hydrous layer and maturation of oxygen isotopes in biogenic silica*
300
301 Biogenic silica has an amorphous structure containing Si-O-Si bonds, Si-OH bonds and
302 crystallization water (Knauth and Epstein 1982). These oxygen-bearing compounds (–OH
303 and H_2O) can exchange freely with their environmental lake water (Fig. 3), for example
304 with sedimentary pore water during the burial of diatoms (Mopper and Garlick 1971;
305 Kawabe 1978; Mikkelsen et al. 1978; Schmidt et al. 1997; Brandriss et al. 1998; Moschen
306 et al. 2006) or with laboratory water used in diatom cleaning preparation techniques
307 (Tyler et al. 2017). The hydrous layer must be removed prior to $\delta^{18}O$ measurements due to
308 its ready exchangeability (most notable in modern diatoms), which makes it a complex
309 mineral to analyse (Leng and Sloane 2008). Secondary processes, such as diagenesis, can
310 also alter $\delta^{18}O_{diatom}$ due to the presence of this hydrous layer.
311

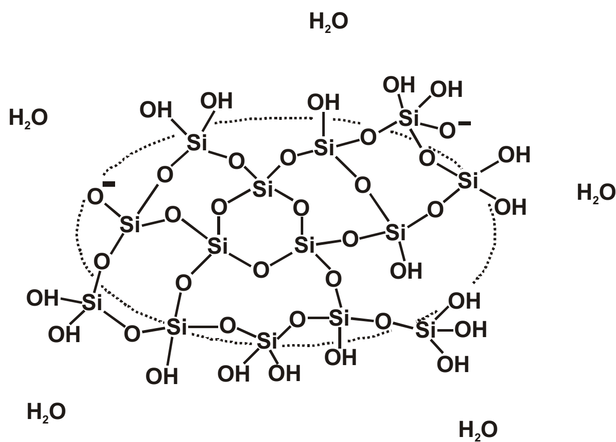


Fig. 3: *Schematic illustration of the nature of amorphous hydrated silica (from Leng and Marshall 2004)*

The influence of silica condensation on the isotope composition of sedimented opal, as a result of isotope exchange, has been described by Schmidt et al. (2001). Diatom silica ^{18}O enrichment is attributed to biogenic silica maturation (dehydroxylation i.e. reduction of Si–OH groups) following the removal of organic coatings (Moschen et al. 2006). Similarly, secondary processes are likely to affect sedimentary diatomaceous silica (especially the hydrous parts), although the vast proportion (c. 90%) of diatom valve oxygen is bound to silicon in SiO_4 tetrahedrons (forming the structurally bound oxygen) which should be more resistant to alteration (refer to reviews of Leng and Henderson, 2013; Swann and Leng, 2009). Furthermore, the absence of trends in $\delta^{18}O$ signatures through time would suggest progressive silica maturation does not occur and it is likely that there is a very slow progression of the maturation process after a fast initial phase of signal alteration. Therefore, some of the diatom $\delta^{18}O$ composition is acquired soon after the formation of biogenic silica, during early diagenesis in the water column and later during early sediment burial (Dodd and Sharp 2010). Interestingly, the conflating effects of temperature on $\delta^{18}O$ recorded by palaeo-diatom silica could be reduced or removed via maturation in deep lacustrine environments with nearly constant temperatures as re-equilibration of diatom silica occurs, thereby providing direct information on the $\delta^{18}O$ of the water (Dodd and Sharp 2010).

2.5 Methods and inter-laboratory calibrations

336

337 There are several techniques for the dehydration and release of O_2 from biogenic silica for
338 $\delta^{18}O$ analysis. However, only one silica standard is universally available (NBS28 quartz)
339 distributed by the IAEA, Vienna. Additional standards to calibrate the $\delta^{18}O$ values of
340 biogenic silica were introduced through an inter-laboratory comparison (Chapligin et al.
341 2011) by eight participating laboratories using their individual bespoke methods and IRMS.
342 The standard materials (diatoms, phytoliths and synthetically-produced hydrous silica)
343 were analysed in accordance to a prescribed protocol. Despite procedural differences at
344 each laboratory (controlled isotopic exchange, stepwise fluorination, inductive high-
345 temperature carbon reduction and inert gas flow dehydration; Chapligin et al. 2011; Leng
346 and Henderson 2013), all methods were in reasonable agreement, with a standard
347 deviation (SD) range for $\delta^{18}O$ between 0.3 ‰ and 0.9 ‰.

348 There are several methods published for the liberation of $\delta^{30}Si$ from biogenic silica,
349 these include both acid and alkaline dissolution/fusion, Si separation using cation
350 exchange, selective co-precipitation, and gas-source versus plasma-ionization (high and
351 low resolution) mass-spectrometric techniques (Reynolds et al. 2007). Three standards
352 were used for a $\delta^{30}Si$ inter-laboratory comparison exercise using a variety of chemical
353 preparation methods and mass spectrometric techniques. The standard reference
354 materials used were IRMM-018 (a SiO_2 standard), Big-Batch and Diatomite (natural
355 diatomites). All analyses were compared with the international Si standard NBS28
356 (RM8546) and were in reasonable agreement (within ±0.22 ‰ (1σ) for $\delta^{30}Si$) showing little
357 statistical difference between the mean values obtained by each laboratory, with the
358 notable exception of the IRMM-018. Overall, they concluded that all the methods have
359 similar precision and differences are limited to 0.2 ‰ in mean $\delta^{30}Si$ values for a given
360 sample between laboratories (or differences of 0.13 ‰ in mean $\delta^{29}Si$). On the basis of this
361 study, the reference standard Diatomite, is routinely reported to demonstrate analytical
362 precision (consensus value of +1.26 ‰ ± 0.2 ‰, 2 SD; Reynolds et al. 2007).

363 While there are no inter-calibration studies on $\delta^{30}Si_{DSi}$ in freshwaters, one was
364 recently undertaken on marine waters, tackling challenges associated with the purification
365 and instrumental precision of waters with low silicic acid concentrations (9 µmol L^{-1})
366 (Grasse et al. 2017). Overall consensus for high (113 µmol L^{-1}) and low concentration

367 seawaters was obtained between all laboratories (+1.25 ± 0.06 and +1.66 ± 0.13 ‰
368 respectively), although some small and significant differences between data were
369 obtained, which the authors attribute to the complex pre-concentration methods
370 (Triethylamine Molybdate; De la Rocha et al. 1996; MAGIC; Georg et al. 2006; or via Mg-
371 induced co-precipitation with purified ammonia; Zhang et al. 2014) and purification steps
372 involved, as well as instrument bias of the different laboratories (e.g., Neptune MC-ICP-
373 MS, Nu Plasma MC-ICP-MS and MAT 252 IRMS) (Grasse et al. 2017).

374 Diatom silica occluded organic matter is mainly derived of pleuralins, silaffins and
375 long chain polyamines. The $\delta^{13}C$ and $\delta^{15}N$ analysis of these compounds from diatoms is
376 thought to be a better representation of the carbon and nitrogen cycle rather than bulk
377 organic matter (Hecky et al. 1973; Kroger and Poulson 2008; Bridoux et al. 2010). There is
378 no universally accepted method for $\delta^{13}C$ and $\delta^{15}N$ analysis of occluded organic matter in
379 diatom silica (Leng and Swann 2010). These methods are more popular in
380 palaeoceanography with few published studies in (palaeo)limnology (Webb et al. 2016).
381 The advantages are: the C and N isotope composition within the diatom cell walls is not
382 affected by post depositional degradation and therefore potentially preserves an
383 unaltered signal of surface water conditions during diatom growth (Brenner et al. 1999;
384 Ficken et al. 2000) and avoids the generally heterogeneous nature of SOM (Hurrell et al.
385 2011).

386

387 *2.6 Future directions*

388

389 Understanding the isotope composition of diatom silica lags behind work on carbonates
390 by decades, probably because of the ongoing issues of contamination, the hydrous layer
391 and associated maturation of diatom silica. Within the community, research continues in
392 better understanding diatom species-specific fractionation effects for $\delta^{18}O_{diatom}$ and
393 $\delta^{30}Si_{diatom}$. In particular, lacustrine case studies are needed because until now data on
394 fractionation factors are predominantly derived from *in-vitro* and *in-situ* experiments of
395 marine diatom strains. The potential for expanding $\delta^{30}Si_{diatom}$ applications in lacustrine
396 systems has been highlighted here, however a solid understanding of contemporary
397 source water and biogenic opal endmembers (e.g. seasonal variability) is essential in order

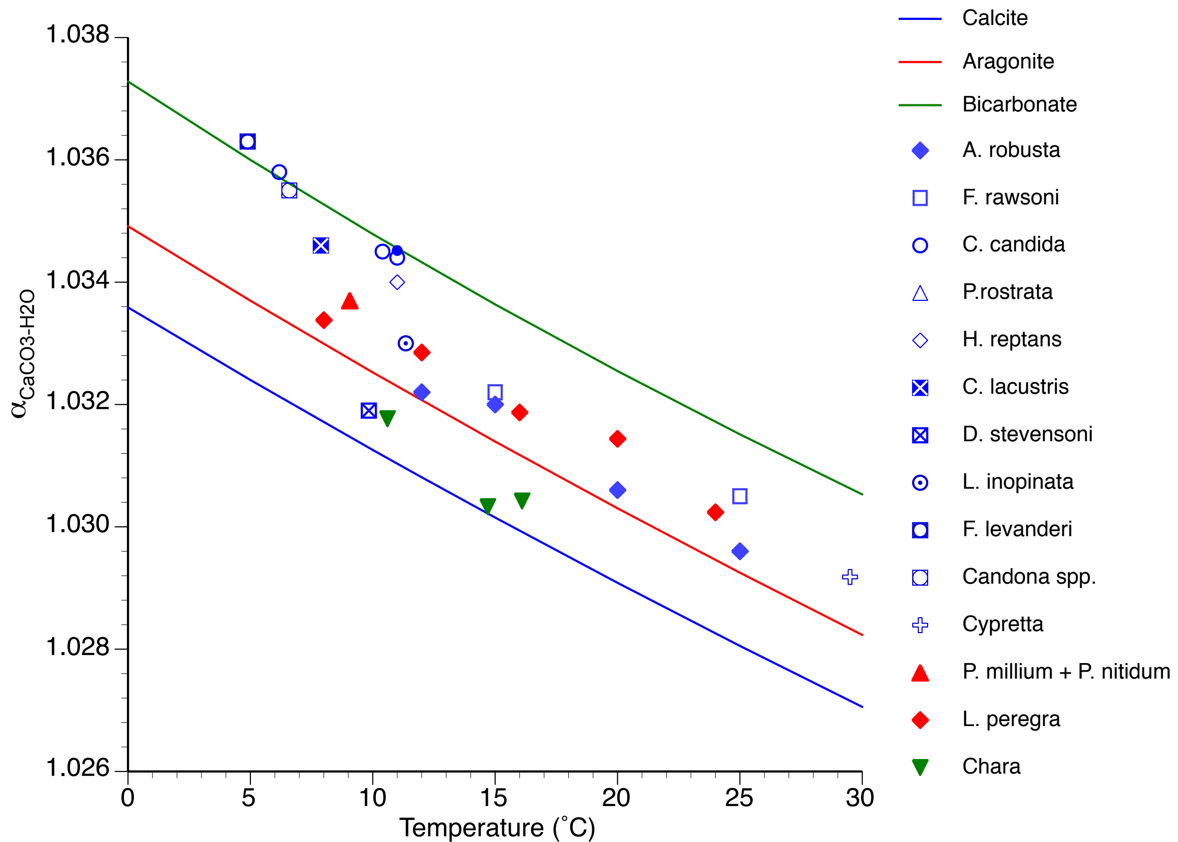
398 to best trace biogeochemical pathways (e.g. changes in DSi supply versus diatom
399 utilisation). In the natural environment, open-system models have been applied to lakes
400 to interpret diatom fractionation processes (Chen et al. 2012; Opfergelt et al. 2011;
401 Panizzo et al. 2016, 2017; 2018b) although closed-system approaches have also been
402 adopted to explain isotope evolution at certain sites (Cockerton et al. 2015). Modern-day
403 calibration studies are needed to best define the most appropriate system and would best
404 constrain any potential for a lake system to periodically shift to a closed-system approach
405 (e.g. during extended periods of ice cover or lake stratification). Furthermore, changes in
406 catchment chemical weathering rates on glacial-interglacial timescales will alter DSi fluxes
407 to lake basins and regulate their isotope composition (e.g., Cockerton et al. 2015; Frings et
408 al. 2016). The coupling of $\delta^{30}Si_{diatom}$ reconstructions with organic-bound $\delta^{15}N_{diatom}$ and
409 $\delta^{13}C_{diatom}$, in addition to stable isotopes of different silicifiers (e.g., sponge spicules), could
410 therefore be further explored as a means to independently constrain the supply of
411 nutrients and their utilisation over such timescales.
412
413
414 **3. Biogenic carbonates**
415
416 The shells of ostracods and aquatic molluscs have often been used in palaeolimnological
417 studies. Ostracods are small (generally microscopic) aquatic crustaceans that are common
418 in lakes. They secrete carapaces made of two low-Mg calcite valves, which are often
419 abundant and well preserved in lake sediments. Ostracods grow by moulting their shells,
420 up to 8 times following hatching from eggs until maturity. Shell formation occurs rapidly,
421 over the course of a few hours to days, and once the shell is formed there is no further
422 addition of calcite (Holmes 1992). Aquatic molluscs, in contrast, are macroscopic and grow
423 incrementally; they thus provide a more time-averaged record of their environment,
424 although carbonate formation may be markedly seasonal (Leng et al. 1999; Leng and
425 Lewis, 2016). Many mollusc species produce aragonitic shells although some may be
426 composed of calcite. Ostracod and mollusc shells are regularly used as sources of
427 carbonate for oxygen and carbon isotope analyses in palaeolimnological reconstructions
428 (Holmes 1996; Holmes and Chivas 2002; Leng and Lewis, 2016). Charophytes are complex
429 algae that are often abundant in shallow, alkaline, fresh to saline lakes (Schneider et al.

430 2015). Charophyte photosynthetic activity can promote the precipitation of calcium
431 carbonate as encrustations around the thallus and associated with the female
432 reproductive bodies known as gyrogonites, the two components being readily
433 distinguished under a light microscope. Dense beds of charophytes can have significant
434 impacts on the $\delta^{13}C$ of DIC within lakes as a result of bicarbonate utilization for
435 photosynthesis (Pentecost et al. 2006). Whilst the carbonate deposits associated with
436 charophytes are not strictly fossils in the same sense as ostracod or mollusc shells, the
437 encrustations often make up a large component of the carbonate sediments formed in
438 freshwater alkaline lakes (Soulié-Märsche et al. 2010), leading to the formation of so-
439 called *Chara*-marl, and so are considered briefly here. Ostracods shells and *Chara* remains
440 tend to be most abundant in lakes situated on carbonate rocks, or those that are in
441 hydrologically-closed or near-closed basins.
442 The oxygen isotope composition of lacustrine carbonate is controlled by water
443 temperature and water isotope composition, as for endogenic calcite (Leng and Marshall
444 2004), together with offsets from isotopic equilibrium, which are discussed further below.
445 The carbon isotope composition of lacustrine carbonate is determined primarily by the
446 carbon isotope composition of DIC: offsets from carbon isotope equilibrium appear to be
447 negligible although, for reasons outlined below, they are difficult to assess.
448 The interpretation of oxygen and carbon isotope signatures derived from lacustrine
449 carbonate depends on the climatic and hydrological characteristics of the lake and its
450 catchment, meaning that an understanding of the modern isotope systematics of the site
451 is beneficial. Species vary in their preferred habitat within a lake and also in their life cycle,
452 with some species calcifying in specific seasons (Decrouy et al. 2011). For molluscs,
453 whereas many taxa are gill-breathing, the fact that some are lung breathing may have an
454 impact on the isotope composition of their shells. Knowledge of an individual species'
455 physiology, ecology and life cycle is therefore also important when interpreting isotope
456 signatures (Shanahan et al. 2005).
457 There are a number of advantages to using shells as opposed to endogenic
458 carbonate for stable isotope analyses. First, the use of shells assures that the carbonate
459 was formed in water and avoids possible inclusion of detrital material with contrasting
460 isotope composition into the sample analysed. Second, the analysis of shells means that
461 constraints can be placed on the timing and location of carbonate formation within a lake.

462 Ostracods provide a specific temporal and spatial 'snapshot' of water conditions and
463 circumvent the problem of averaging. Moreover, the fact that some species have seasonal
464 preferences and inhabit defined zones (e.g., deep benthic versus littoral) within a lake
465 means that isotopic records derived from species with known life cycles and ecologies may
466 provide seasonal and habitat-specific information (von Grafenstein et al. 1999b). For
467 molluscs, whole-shell analyses provide a more time-averaged signal, since the shells grow
468 incrementally, although non-continuous growth throughout the year may mean that that
469 this signal is still seasonally biased (Leng and Lewis, 2016). Conversely, the isotope analysis
470 of individual growth increments, which is analytically feasible for some larger taxa, may
471 provide information about seasonal or inter-annual variability in environment (e.g., Leng
472 et al. 2009; Dettman et al. 1999), although this may not be possible when the short-term
473 changes in temperature or the isotope composition of water/DIC composition are small
474 (Shanahan et al. 2005). Thirdly, analysis of shells provides some certainty over the
475 mineralogy of the material being analysed. For ostracods, it is low-Mg calcite; for molluscs,
476 often aragonite. Because aragonite is thermodynamically unstable and recrystallization
477 leads to the 'resetting' of the isotopic signature (Leng and Marshall, 2004), it is important
478 to assess the degree of preservation of aragonitic mollusc shells, for example using X-ray
479 diffraction to confirm the presence of aragonite. These advantages also bring some
480 problems, however. The time- and space-specific character of an ostracod isotope
481 signature may not record the 'average' conditions within a lake that are generally required
482 in palaeolimnological reconstructions. Furthermore, because the calcification of ostracod
483 and mollusc shells is under strong biological mediation, isotopic fractionation may not
484 conform to expectations derived from investigations of inorganic carbonate. For
485 charophytes, different isotopic signatures may be recorded depending on where on the
486 plant the carbonate formed (Pentecost et al. 2006). In short, care is required in the
487 interpretation of such signatures.
488
489 *3.1 Oxygen and carbon isotope records from lakes*
490
491 Although the fundamental controls on isotope composition of carbonate are well
492 understood and quantifiable in some instances, the palaeoclimatic and
493 palaeoenvironmental interpretation of isotope records from lake sediments depends

494 strongly on the characteristics of individual lakes including their climatic setting, depth,
495 volume, hydrology, aquatic vegetation and catchment properties (Holmes 1996). Oxygen
496 isotope values derived from ostracod or mollusc shells, or marl, have been used in
497 reconstructions of air temperature and the oxygen isotope composition of rainfall (von
498 Grafenstein 2002; von Grafenstein et al. 1999a), effective moisture (Hodell et al. 1991;
499 Street-Perrott et al. 2000; Holmes et al. 2010; Hodell et al. 1995), meltwater influx
500 (Dettman et al. 1995) and changes in river routing within the lake's catchment (Schwalb et
501 al. 1994). Carbon isotopes are often more difficult to interpret but have been used to
502 reconstruct lake/catchment carbon cycling, productivity, and methanogenesis (Bridgwater
503 et al. 1999; Anadón et al. 2006; Li and Liu 2014; Schwalb et al. 2013), although many
504 studies report only the oxygen isotope results (e.g., Hodell et al. 1991; von Grafenstein et
505 al. 1999a; Holmes et al. 2010; Hodell et al. 1995).
506
507 *3.2 Disequilibrium*
508
509 Despite early suggestions that ostracod calcite is precipitated in isotopic equilibrium with
510 the host water (Durazzi 1977, albeit for marine taxa) it is now well established that
511 significant offsets exist, especially for oxygen. Similar offsets have been shown for some
512 mollusc taxa (e.g., White et al. 1999; Shanahan et al. 2005). Knowledge of such offsets is
513 needed if ostracod or mollusc shell isotope values are to be used in quantitative
514 environmental reconstruction (von Grafenstein et al. 1999a; Decrouy 2012; Devriendt et
515 al. 2017). Moreover, offsets have provided insights into calcification mechanisms (Keatings
516 et al. 2002; Shanahan et al. 2005; Decrouy 2012; Devriendt et al. 2017), although the
517 mechanism behind the observed offsets from oxygen and carbon isotope equilibrium
518 remains incompletely understood. Charophyte isotope records are potentially complicated
519 by differences in isotopic signatures between stem encrustations and calcified remains of
520 gyrogonites, which also vary depending on the strength of water flow (Andrews et al.
521 2004; Pentecost et al. 2006).
522
523 *3.2.1 Oxygen*
524

525 Evidence from field collections of ostracods under closely monitored conditions (e.g., von
526 Grafenstein et al. 1999b; Keatings et al. 2002a; Decrouy et al. 2011) as well as *in vitro*
527 cultures (Xia et al. 1997a; Chivas et al. 2002) have shown that their shells are precipitated
528 out of oxygen isotope equilibrium with the host water (Fig. 4). The offsets from equilibrium
529 are almost invariably positive, up to +3 ‰ or more. Several investigations have shown that
530 the magnitude of the offset varies taxonomically, with members of the same genus, or
531 even sub-family or family, sharing similar offset values (see table 10.1 in Decrouy 2012).
532 Adults and juveniles and males and females of the same species usually show offsets of
533 the same magnitude (Chivas et al. 2002; Decrouy 2012; von Grafenstein et al. 1999b). The
534 two culturing studies cited above suggest that offsets are greater at higher temperature.
535 Decrouy et al. (2011) have demonstrated that there are some differences in the magnitude
536 of the vital offset *within* taxa from different localities, suggesting that water chemistry, in
537 addition to taxonomy, may also play a role. Devriendt et al. (2017) have confirmed these
538 observations based on a comprehensive meta-analysis of studies undertaken over a very
539 large range of water types, and have argued that they can be explained by a carbonate ion
540 effect. At high pH, a greater proportion of the DIC is present as the CO_3^{2-} ion rather than as
541 HCO_3^-, which dominates in lower pH waters: the degree of fractionation between water
542 and the CO_3^{2-} ion is less than that with HCO_3^-, and since ostracod shells are formed from
543 DIC, they will have lower $\delta^{18}O$ values in higher pH waters (Devriendt et al. 2017). Positive
544 offsets from oxygen isotope equilibrium have been observed in some mollusc taxa (White
545 et al. 1999; Shanahan et al. 2005) although some species appear to precipitate their shells
546 in isotopic equilibrium (Leng et al. 1999).
547



548

549

550 **Fig. 4**. Fractionation factors between selected ostracod and mollusc species and water.
551 Fractionation between bicarbonate (Beck et al. 2005), synthetic calcite (Kim and O'Neil
552 1997) and synthetic aragonite (Kim et al. 2007) is also shown. Key to taxa and sources: *A.*
553 *robusta – Australocypris robusta* (Chivas et al. 2002): *F. rawsoni – Fabaeformiscandona*
554 *rawsoni* (Xia et al. 1997a): *C. candida – Candona candida* (Keatings, 1999; von Grafenstein
555 et al. 1999b): *P. rostrata – Pseudocandona rostrata: H reptans – Herpetocypris reptans*
556 (Keatings et al. 2002): *C. lacustris – Cytherissa lacustris: D. stevensoni – Darwinula*
557 *stevensoni: L. inopinata – Limnocythere inopinata: F. levanderia – Fabaeformiscandona*
558 *levanderi: Candona* spp. (von Grafenstein et al. 1999b): *C. brevisaepta - Cypretta*
559 *brevisaepta* (J. A. Holmes, unpublished): *P. millium + P. nitidum - Pisidium millium +*
560 *Pisidium nitidum* (Keatings 1999): *L. peregra - Lymnaea peregra* (White et al. 1999): *Chara*
561 – charophyte stem encrustations (Andrews et al. 2004).

562

563

564 The offsets from isotopic equilibrium must be corrected for if calculations of past water
565 temperature or past water isotope composition are to be derived from the oxygen isotope

566 values of ostracod shells, otherwise significant errors may arise (von Grafenstein 2002;
567 Devriendt et al. 2017).
568
569 *3.2.2 Carbon*
570
571 Assessing offsets from carbon isotope equilibrium in shells is more difficult because the
572 $\delta^{13}C$ composition of DIC, the source of carbon for calcification, varies significantly at the
573 micro scale meaning that there may be a weak relationship between the $\delta^{13}C$ at the site of
574 calcification and that within the main lake, which is the value that is typically measured.
575 This is much more so than for oxygen isotopes, which tend to be relatively more
576 homogenous within a lake. For example, the DIC within sediment pores may be ^{13}C-
577 depleted as a result of mineralisation of SOM. In methane-producing lakes, however, the
578 formation of ^{13}C-enriched co-genetic CO_2 may have the opposite effect (Durand et al.
579 1984), which may lead to very high $\delta^{13}C$ values in the shells of infaunal ostracods
580 (Bridgwater et al. 1999). Interestingly, the opposite (very low ostracod $\delta^{13}C$ values) can be
581 observed when methane is oxidized close to the sediment-water interface and ^{13}C-
582 depleted carbon is added to pore water DIC that is then available for incorporation into
583 ostracod shells (Schwalb et al. 2013). Close to the leaves of submerged macrophytes that
584 utilise HCO_3^-, DIC may also be ^{13}C-enriched as a result of preferential uptake of ^{12}C by
585 plants for photosynthesis (Kelts and Talbot 1990). Hence, organisms co-existing in different
586 micro-environments within a lake, or calcifying at different seasons, may have markedly
587 contrasting $\delta^{13}C$ values (Bridgwater et al. 1999). For molluscs, contrasts can be seen
588 between gill-breathing and lung-breathing species, which may relate to differences in
589 physiology or life history (Shanahan et al. 2005). Variations in $\delta^{13}C$ values between
590 different taxa are therefore often regarded as habitat effects rather than offsets from
591 equilibrium *sensu stricto* (Heaton et al. 1995).

592

593 *3.3 Sample preparation and shell cleaning*
594
595 Shells destined for isotope analysis need to be well preserved (cf. section 3.4, below) and
596 free from detrital contamination. Various methods have been used to remove

597 contamination from ostracod shells, especially organic material, which may interfere with
598 isotope determinations. However, the potential of these techniques to modify the primary
599 isotope composition of the shell as well as to remove any contaminant has not been fully
600 assessed. Five techniques have been used to remove contaminants, namely simple manual
601 cleaning with a fine paint brush and (usually) methanol, roasting *in vacuo*, heating in an
602 oxygen plasma and chemical oxidation with sodium hypochlorite (clorox) or hydrogen
603 peroxide (Keatings et al. 2006). For each technique, a variety of conditions (treatment
604 times, temperatures, reagents strengths) has been used. Keatings et al. (2006) compared
605 treated and untreated (manually cleaned) valves of the same carapace of late Pleistocene
606 lacustrine ostracods and showed that in most instances, the mean impact of the cleaning
607 techniques over manual cleaning was small, although treatment other than simple
608 mechanical cleaning typically increased within-sample variability. Roberts et al. (2018)
609 confirmed that treatment can cause changes in both oxygen and carbon isotope values,
610 but concluded that hydrogen peroxide treatment would be preferable if treatment were
611 required, since this reagent does not appear to have a significant impact on the isotope
612 composition. Despite the relatively small changes imparted by each of the treatment
613 methods, the increase in variability that may result suggests that anything other than
614 simple mechanical cleaning should only be undertaken if absolutely necessary (i.e. to
615 remove detrital contamination that could not otherwise be eliminated). The potential
616 impact on variability needs to be borne in mind if single-shell samples are analysed
617 (section 3.5) and, moreover, implications of cleaning for trace elements must be taken into
618 account in cases where 'tandem' isotope and trace-element determinations are
619 undertaken on the same shells, as in Chivas et al. (1993) and Xia et al. (1997b). Similar
620 methods have been employed for the removal of organic material from mollusc shells
621 destined for isotope analyses: modifications to techniques involving heating and or
622 grinding are required in order to prevent inversion of aragonite to calcite (White et al.
623 1999; Dettman et al. 1999). Marl samples, including those dominated by charophyte
624 carbonate, are commonly treated by heating or chemical oxidation to remove organic
625 matter (e.g., Apolinarska and Hammarlund 2009).
626
627 *3.4 Signal preservation and diagenetic alteration*
628

An advantage of analysing shell calcite over endogenic calcite is that diagenetic alteration can usually be detected. For ostracods, early diagenetic alteration can usually be detected by the presence of etching, opaque rather than transparent or translucent appearance, and obscured or altered surface ornament (Keatings et al. 2002b). Ideally, shells showing signs of such alteration should be excluded from analyses. However, in some cases pristine material may not be preserved in sediment sequences, meaning that ostracod specimens that have undergone some degree of alteration need to be analysed. Limited attention has been paid to the impacts of early diagenesis on isotope signatures. Keatings et al. (2002b) found no clear evidence of an impact of preservation on the oxygen isotope signature of late Pleistocene lacustrine ostracods from a Jamaican hardwater lake: evidence for the absence of an impact on carbon isotopes was less conclusive. The potential for alteration increases for older material. Bennett et al. (2011) showed that isotope values, especially for oxygen, of Carboniferous non-marine ostracods from Scotland primarily reflected the degree of diagenetic alteration rather than a palaeoenvironmental signature and warned of the potential for cryptic diagenesis to alter the shells and their isotope values. However, Bajpai et al. (2013) derived plausible palaeoenvironmental signatures from both the oxygen and carbon isotope values from Late Cretaceous non-marine shells from peninsular India, some pristine and others showing signs of alteration. For aragonitic mollusc shells, recrystallization to calcite can effectively ‘reset’ the isotopic signature: mineralogical assessment of shells destined for isotope analyses is therefore often undertaken (e.g., Leng et al. 1999).

3.5 Measurement of multiple versus single-shell samples

Adult ostracod shells weigh anything from a few micrograms for a single shell of small or weakly calcified taxa, such as species belonging to the genera *Limnocythere, Darwinula* and *Cypria,* to several hundred micrograms for large and well-calcified taxa belonging to genera such as *Sclerocypris*, although many taxa have shells in the tens of micrograms range (J. A. Holmes, unpublished). Juveniles are not only smaller than adults of the same species but also less well calcified, meaning that they weigh substantially less. Duel-inlet mass spectrometers are therefore able to measure isotope ratios for single shells of adults of many ostracod species (Chivas et al. 1993). Most stratigraphic studies of ostracods have

661 used single, multiple-shell, monospecific samples, where the number of shells analysed is
662 determined not by the material requirements of the instrument, but by the need to obtain
663 a sample that is representative of 'average' conditions within the lake and over the time
664 interval represented by the increment of sediment from which the shells are recovered.
665 However, there is increasing interest in undertaking analyses of multiple single shells at
666 individual stratigraphic intervals in a lake-sediment sequence. Such an approach has the
667 advantage of providing information about short-term variability within the lake as well as
668 allowing any analytical outliers to be identified, although it does of course increase
669 analytical costs. Pilot studies of large numbers of shells from a few intervals can be used to
670 determine optimum sample size (Escobar et al. 2010). Such an approach has been used to
671 assess short-term changes in effective moisture in NW India (Dixit et al. 2015) and in
672 Mayan lakes from Central Mexico (Escobar et al. 2010), and variations in meltwater input
673 in Lake Huron (Dettman et al. 1995). Despite the fact that a single mollusc shell typically
674 integrates a longer time interval than a single ostracod shell, and would typically provide
675 sufficient material for an isotope determination, similar issues have arisen with the isotope
676 analysis of single mollusc shells, especially for species that have short life spans
677 (Apolinarska et al. 2015).
678
679 *3.6 Other isotopes and future developments*
680
681 Further work is needed to compare oxygen-isotope signatures in different minerals or
682 biomolecules. Theoretically, comparison of signatures in calcite or biogenic silica, in which
683 oxygen-isotope fractionation is temperature dependent (section 3.2.1), with those in
684 materials such as cellulose or chitin, which show temperature-independent fractionation
685 (section 4.2.2), should lead to quantitative reconstructions of water temperature.
686 However, existing studies show that this does not always work in practice. For example,
687 Rozanski et al. (2010) attributed unrealistically-high estimates of temperature from
688 coupled calcite and cellulose oxygen isotope analyses for an eastern European lake to
689 kinetic effects during rapid carbonate formation. Comparison of δ^{18}O values of diatom
690 silica and endogenic calcite in Lake Pinarbasi, Turkey, show contrasting trends in spite of
691 their mutual dependence on the water δ^{18}O and lake-water temperature. The most likely
692 explanation for this divergence is difference in seasonality of biological productivity

693 mediated by the strongly continental climate of the Anatolian plateau. The endogenic
694 calcite $\delta^{18}O$ is thought to be temporally limited to a few summer months and the diatom
695 silica $\delta^{18}O$ provides seasonally-specific water isotope composition in the spring and
696 autumn and, at least in the record in question, captures periods of heavy snow through
697 the spring thaw (Leng et al. 2001). Further investigations are needed to indicate the
698 circumstances in which lake water temperature can be reconstructed from $\delta^{18}O$ offsets
699 between calcareous/siliceous and organic remains.
700 Ostracod shells have been used in analyses of other isotopes in a limited number of
701 studies, including $^{87}Sr/^{86}Sr$ (Janz and Vennemann 2005; Holmes et al. 2007) and U-series
702 (for dating) (Bischoff et al. 1998). The chitinous shell linings, which remain after dissolving
703 the calcium carbonate with a 5 % HCl solution, can be used for measuring $\delta^{13}C$ and $\delta^{15}N$ to
704 provide insights in the position of ostracods in the aquatic food web (Fig. 6). Future
705 developments require additional calibration work to improve understanding of the
706 mechanisms behind vital offsets and their quantification for additional species, especially
707 for oxygen isotopes. Future work may also lead to the use of 'novel' isotope systems like
708 Ca (Oehlerich et al. 2015) and clumped isotope analysis (Mering 2015) on biogenic
709 carbonates.
710
711 **4. Organic remains of aquatic plants and animals**
712
713 Many organisms living in lakes produce organic structures that are remarkably robust to
714 degradation once buried in lake sediments. These remains are composed (partly) of
715 flexible polymers that are difficult to break down including chitin, keratin, lignin, collagen,
716 and cellulose. These polymers are bonded or otherwise associated with proteins, calcium,
717 carbonate, or other compounds to provide strength (Leschine 1995; Nation 2002;
718 Korniłłowicz-Kowalska and Bohacz 2011; Jex et al. 2014). We will focus on identifiable
719 organic remains of some of these organisms, including invertebrates, aquatic macrophytes
720 and fish. We will review how taxon-specific analysis of stable isotopes on these remains
721 are used in palaeolimnological studies to understand changes in carbon cycling, food web
722 structure, eutrophication, hydrology and climate.
723

724 *4.1 Chitinous invertebrate remains*

725

726 Invertebrates are ubiquitous in lakes and their exoskeleton fragments and resting eggs are
727 preserved in lake sediments for tens of thousands of years (e.g., Engels et al. 2010).
728 Remains of a wide range of invertebrates can be identified using microscopy and the
729 composition of fossil invertebrate assemblages can be indicative of ecological and
730 environmental conditions (Frey 1964). The exoskeleton fragments from various insect
731 orders (e.g., Coleoptera, Diptera, Ephemeroptera, Trichoptera), crustaceans (e.g.,
732 Cladocera, Ostracoda), and mites (Oribatida) are commonly found in lake sediments, as
733 are the resting stages of Cladocera and moss animals (Bryozoa).

734 Robust organic remains of many invertebrate groups consist predominantly of
735 chitin cross-linked with protein (Stankiewicz et al. 1996; 1998). In suitable conditions they
736 can remain relatively unchanged over thousands of years (Miller 1991; Verbruggen et al.
737 2010a). Isotope analyses of these remains can therefore be used to reconstruct past
738 changes in the isotope composition of the formerly living organisms (Wooller et al. 2004;
739 Verbruggen et al. 2011). The chemical composition of these remains means that they
740 contain relatively large amounts of hydrogen, carbon, oxygen, some nitrogen, and to a
741 lesser extent sulphur. Isolating the small remains from sediments and preparing them for
742 stable isotope analysis can be time consuming, but efficient protocols are available (Wang
743 et al. 2008), which can be adapted depending on the chemical pre-treatment steps
744 required (e.g., Heiri et al. 2012). Using a 200-micrometre mesh (instead of the commonly
745 used 90 to 100-micrometre mesh) can decrease processing time by 30 to 58 % because of
746 the disproportionally large gain in sample mass from larger fragments (van Hardenbroek
747 et al. 2010a). Using large mesh sizes may cause the loss of smaller fragments, which could
748 lead to systematic bias against particular taxa and body parts – and subsequently against
749 certain habitats, or feeding habits.

750 The amount of sample required for stable isotope analysis depends on the
751 chemical element being analysed, the relative abundance of the element in the
752 invertebrate remains as well as on the analytical equipment used. The carbon content of
753 chitinous invertebrate remains is relatively high, followed by oxygen, nitrogen, and
754 hydrogen (Table 1). The elemental composition (% by weight) of invertebrate remains is
755 generally 40-50 %C, 25-30 %O, 7-10 %N, and 5-6 %H. Interestingly, the nitrogen content of

756 Bryozoa remains (12.7 ± 1.7 %N) is consistently higher than that of other invertebrates. A
757 lower carbon and nitrogen content has been observed in the carapaces of Cladocera (18.3
758 ± 2.7 %C and 2.8 ± 0.7 %N, respectively). At present it is unclear what causes the
759 differences between taxa and different types of remains; further analysis of the chemical
760 composition of the remains is required.
761
762 **Table 1**: *Elemental composition (% by weight) with 1 standard deviation (SD) of*
763 *invertebrate remains in surface and down core sediments (n = number of data points).*
764 *Data from van Hardenbroek et al. (2010b, 2012, 2013b, 2014, 2018, van Hardenbroek,*
765 *Heiri and Wooller unpublished data, Perga unpublished data).*

	%C	SD	n	%N	SD	n
Chironomidae head capsules	46.4	5.4	247	8.6	1.3	247
Cladocera ephippia	44.4	5.4	380	8.8	1.8	379
Cladocera carapaces	18.3	2.7	28	2.8	0.7	28
Bryozoa statoblasts	45.7	3.9	168	12.7	1.7	167
Ephemeroptera mandibles	39.0	10.0	19	7.5	3.0	19
Ostracoda shell lining	41.7	7.6	11	10.4	2.1	11
Trichoptera frontoclypeus/mandible)	42.5	9.5	20	8.1	1.7	20
Sialis (frontoclypeus/mandible)	47.0	3.7	13	10.0	1.2	13
Chaoborus (mandible)	45.3	7.4	22	8.8	1.4	22
Coleoptera (elytron)	43.1	2.8	14	8.2	1.0	14

	%O	SD	n	%H	SD	n
Chironomidae head capsules	29.6	1.8	27	5.6	0.3	27
Cladocera ephippia	28.6	2.6	77	5.5	0.5	77
Bryozoa statoblasts	25.5	3.9	48	5.2	1.1	48

766
767
768 For $\delta^{13}C$ and $\delta^{15}N$ analyses an Elemental Analyser IRMS (EA-IRMS) setup is
769 commonly used, whereas $\delta^{18}O$ and δD analysis often rely on a Thermal Conversion /
770 Elemental Analyzer (TC/EA)-IRMS setup. With this equipment, analyses are typically based
771 on tens to hundreds of invertebrate remains. A reduction of the sample size is possible
772 using Laser Ablation nano Combustion Gas Chromatography (LA/nC/GC)-IRMS (Schilder et
773 al. 2018) or Spooling-Wire Microcombustion (SWiM)-IRMS (Zhao et al. 2017), allowing $\delta^{13}C$
774 analysis of individual invertebrate remains. The Supplementary Table provides an
775 overview of mean weight of individual remains, to allow better estimates of the minimum
776 number of remains required for stable isotope measurements. The mean weight of

777 remains in down core samples is seemingly smaller than that of remains in surface
778 sediments (Supplementary Table). Material from surface and down core samples
779 originates from different sites and includes different species, making it difficult to make
780 conclusive statements about differences between remains in surface and down core
781 samples in the Supplementary Table without more work on the taphonomy of
782 invertebrate remains.
783
784 *4.2 Dietary and environmental isotopes reflected by chitinous remains*
785
786 Analyses of the isotopic offsets between aquatic invertebrates, their food and their
787 chitinous remains focussed on different organism groups such as marine crustaceans,
788 freshwater crustaceans and aquatic insects. Much of this work started in the 1980s mainly
789 with marine decapods (Schimmelmann and DeNiro 1986a, b; Schimmelmann 2011).
790 However, results based on marine crustaceans may not be representative for chitinous
791 microfossils of many freshwater invertebrate taxa. This is because calcium carbonate
792 forms an important component of the exoskeleton of many crustaceans (Greenaway
793 1985; Willis 1999), but calcium may be less relevant in some planktonic freshwater
794 crustaceans (Jeziorski and Yan, 2006) and the cuticles of other freshwater invertebrates
795 groups (e.g., aquatic insects, Willis 1999) do not contain calcium carbonate. Many
796 experimental studies assessing the relationships between isotope sources and isotopic
797 contents of invertebrates have selectively isolated and analysed chitin or chitin-derived
798 compounds (Schimmelmann and DeNiro 1986a, b; Schimmelmann 2011) and this needs to
799 be considered when using this information for interpreting stable isotope studies based on
800 whole chitinous remains (including proteins, lipids, etc.) of aquatic invertebrates.
801
802 *4.2.1 Carbon and nitrogen isotopes*
803
804 Carbon and nitrogen in chitin of heterotrophic organisms originates from their diet
805 (DeNiro and Epstein 1978, 1981; Schimmelmann 2011). Metabolic activities and life stage
806 may also influence the isotope composition for some elements and organism groups
807 (Schimmelmann 2011). Chitinous remains of aquatic invertebrates in lake sediments do
808 not only consist of chitin but include other organic components such as proteins or lipids

809 (e.g. Verbruggen et al. 2010a). The exact composition of fossilising exoskeleton parts may
810 differ between organism groups but also within the same organism, between different
811 types of structures (e.g., Jeziorski et al. 2008). Ideally, the relationship between the
812 isotope composition of diet, bulk tissue and fossilizing structures is therefore established
813 for each organism group and isotope pair of interest before developing down core isotope
814 records. Experiments and environmental measurements constraining these relationships
815 for C and N are presently available for planktonic cladocerans, chironomid larvae and
816 bryozoans. Laboratory experiments with *Chironomus* (Chironomidae) larvae showed that
817 there are only very minor offsets (reported here as Δ values) between the $\delta^{13}C$ values of
818 the food and chironomid biomass ($\Delta^{13}C$ of ca. –1.5 to +1.5 ‰ in most cases; Goedkoop et
819 al. 2006; Wang et al. 2009; Heiri et al. 2012; Frossard et al. 2013). In contrast, observed
820 offsets between foods and body tissue for $\delta^{15}N$ were more pronounced ($\Delta^{15}N$ –1.5 to +3.4
821 ‰; Goedkoop et al. 2006; Wang et al. 2009; Heiri et al. 2012). For fourth instar larvae,
822 head capsule $\delta^{13}C$ and $\delta^{15}N$ values were very similar to but on average ca. 1 ‰ lower than
823 the isotope composition of the remaining larval tissue (Heiri et al. 2012; Frossard et al.
824 2013).

825 The $\delta^{13}C$ values of the exoskeleton of the cladocerans *Daphnia* and *Bosmina* were
826 shown to be very similar to the values for the bodies: offsets are –0.8 ± 0.2 ‰ and 1.4 ±
827 0.7 ‰ for *Daphnia* and *Bosmina*, respectively (Perga 2010). Lower $\delta^{15}N$ values for *Daphnia*
828 exoskeletons were observed compared with the whole bodies (by as much as 7.9 ± 0.5 ‰;
829 Perga (2010). This offset was very consistent along a gradient of *Daphnia* $\delta^{15}N$ values,
830 however, and a strong relationship between $\delta^{15}N$ values of *Daphnia* exoskeletons and the
831 entire organisms was found. The fossilizing sheaths of resting eggs (ephippia) of *Daphnia*
832 *pulicaria* are very similar in their isotope composition to the *Daphnia* they originated from,
833 with mean offsets of +0.2 and –1.5 ‰ for $\delta^{13}C$ and $\delta^{15}N$, respectively (Schilder et al.
834 2015a). Experimental results indicate that the $\delta^{13}C$ values of *Daphnia* are also close to the
835 isotope composition of the available food, with offsets between food and *Daphnia* of 0.5 ±
836 0.3 ‰ ($\Delta^{13}C$) and +3.4 ± 0.3 ‰ ($\Delta^{15}N$) and between food and ephippia of 0.7 ± 0.2 ‰
837 ($\Delta^{13}C$) and 1.8 ± 0.4 ‰ ($\Delta^{15}N$) (Schilder et al. 2015a).

838 Experimental results indicate that $\delta^{13}C$ values of chitinous resting stages
839 (statoblasts) of the bryozoans *Plumatella* and *Lophopus* are similar as those of both their

840 food and soft tissue parts of these organisms: statoblasts have between 0 to 1.7 ‰ lower
841 $\delta^{13}C$ values than the living colonies (van Hardenbroek et al. 2016). In a survey of lakes in
842 Northwest and Central Europe, median $\delta^{13}C$ values of bryozoan colonies of *Cristatella*
843 *mucedo*, *Pectinatella magnifica* and *Plumatella* were observed to be closely related with
844 the isotope composition of the statoblasts for most sites (offsets generally between –3 to
845 +4.5 ‰; van Hardenbroek et al. 2016). The seemingly large variability of offsets recorded
846 in the above study suggests that, in field situations, food sources can change between the
847 formation of fossilizing structures and the formation of new soft tissue. Chitinous
848 fossilising invertebrate structures are formed within a period of 1-2 weeks (Candy and
849 Kilby 1962; Nation 2002) and their stable isotope values will largely represent food sources
850 in this period and shortly before. In the limited studies comparing living invertebrates and
851 their fossilising structures, the $\delta^{13}C$ values of fossilising structures are within one standard
852 deviation of the mean $\delta^{13}C$ values of living chironomids and cladocerans (Morlock et al.
853 2017; Schilder et al. 2017). This strongly suggests that $\delta^{13}C$ values of samples of fossilising
854 structures are representative for mean $\delta^{13}C$ values of the invertebrates at longer (annual
855 to decadal) time scales. The large number of sedimentary remains required for regular EA-
856 IRMS measurements will not normally allow detection of short-term (weekly to seasonal)
857 variability in stable isotope values found in living invertebrates (but see Zhao et al. 2017
858 and Schilder et al. 2018). Future studies should quantify the impact of tissue turnover time
859 and the sources of material incorporated during the formation of chitinous structures on
860 the stable isotope composition of these remains. This would be similar to stable isotope
861 studies on turnover time of different tissues in other aquatic organisms such as fish (e.g.,
862 Pinnegar and Polunin 1999; Hanisch et al. 2010).

863 When quantifying offsets between soft tissues and fossilising structures, controlled
864 laboratory experiments give more consistent results than field studies. Culturing
865 experiments have demonstrated that variability in these offsets is generally low: 0.2–0.9
866 ‰ for $\delta^{13}C$ and 0.3–0.5 ‰ for $\delta^{15}N$ (Perga 2010; Heiri et al. 2012; Frossard et al. 2013;
867 Schilder et al. 2015a). Information from such controlled experiments is crucial as it
868 demonstrates that variability > 0.9 ‰ ($\delta^{13}C$) and > 0.5 ‰ ($\delta^{15}N$) is most likely related to
869 environmental processes rather than natural variability in offsets between soft tissues and
870 fossilising structures.

871

872 *4.2.2 Oxygen and hydrogen isotopes*

873

874 Less information, compared with C and N, is available that documents the relationship
875 between the stable O and H isotope composition of aquatic invertebrates, their fossilizing
876 remains, and their diet and source water. Available evidence from lakes indicates that
877 most of the O in aquatic invertebrates originates from lake water (56 to 84 %; Wang et al.
878 2009; Nielson and Bowen 2010; Soto et al. 2013; Schilder 2015a) with smaller
879 contributions from dietary O. The exact proportions vary between the available
880 experiments and therefore may also vary between different organism groups, tissue types
881 and chemical compounds. Schilder et al. (2015a) demonstrated that changing the oxygen
882 isotope composition of the water influenced δ^{18}O values of *Daphnia* ephippia, which were
883 on average 0.8 ± 0.4 ‰ lower than for the entire *Daphnia*. Limited evidence from this
884 culturing experiment also suggested no significant temperature-dependent fractionation
885 between δ^{18}O values of water and δ^{18}O values of *Daphnia* tissues (Schilder et al. 2015a). A
886 close relationship between δ^{18}O of aquatic invertebrate remains and lake water has also
887 been confirmed by environmental surveys demonstrating a high correlation between δ^{18}O
888 values of chitinous invertebrate remains in lake sediments and lake water δ^{18}O (Fig. 5 and
889 Wooller et al. 2004; Verbruggen et al. 2011; Lombino 2014; Mayr et al. 2015; Chang et al.
890 2016, 2017; Lasher et al. 2017).

891 The greatest proportion of H in freshwater invertebrate biomass and chitinous
892 remains apparently originates from the diet of these organisms (Solomon et al. 2009;
893 Wang et al. 2009; Soto et al. 2013; Belle et al. 2015b). Based on controlled experiments,
894 estimates of the amount of H in freshwater invertebrate tissue originating from water
895 range from 20 to 47 % (Solomon et al. 2009; Wang et al. 2009; Soto et al. 2013). Belle et al.
896 (2015b) reported offsets between chironomid larval bodies and fossilizing head capsules of
897 –24 ± 7 ‰. In a field study of bryozoan colonies in 23 lakes, median δD values of the
898 fossilizing statoblasts of the colonies were relatively close to the median δD values of their
899 soft tissues for *Plumatella* (observed offsets –34 to +16 ‰) but distinctly more negative (–
900 75 to –16 ‰) for *Cristatella* (van Hardenbroek et al. 2016). This finding might be related to
901 differences in food type, mobility and seasonality of resting stage production between the

902 examined bryozoan groups. The H-isotope composition of many food types available in
903 lakes (e.g., algae and organisms feeding on these) will be related to the isotope
904 composition of the lake water. Therefore, δD values of aquatic invertebrate remains will in
905 many cases be related to lake water δD values, even if food sources play a major role in
906 determining aquatic invertebrate δD values (see, e.g., van Hardenbroek et al. 2016).

907 The observed variability in offsets between soft tissues and fossilising structures can
908 be as large as 50‰ for δD in field studies due to natural variability. Controlled laboratory
909 experiments are required to evaluate what part of this variability is due to changes in the
910 environment (food supply, temperature, isotope composition of lake water) and what part
911 is the inherent natural variability in the offset between soft tissues and fossilising remains.
912 Limited data from controlled studies suggests that natural variability in this offset may be
913 as small as ca. 0.4‰ for $\delta^{18}O$ (Schilder et al. 2015a) and ca. 7% for δD (Belle et al. 2015b).
914 This implies that variations > 0.4 ‰ for $\delta^{18}O$ and > 7 ‰ for δD will in many instances be
915 related to environmental processes and could be interpreted in palaeoenvironmental
916 records.

917

918 *4.3 Effects of chemical treatment and taphonomy*

919

920 Chitinous invertebrate remains for isotope analysis are isolated from sediments by a
921 combination of mechanical and chemical treatments (van Hardenbroek et al. 2010a).
922 Sediments are usually first chemically treated by exposing the samples in a 5 to 10 % KOH
923 solution to facilitate sieving and eliminate easily degradable organic material. For C and O
924 isotopic analysis it is essential that carbonates are eliminated prior to isotope analysis by
925 exposing the samples to acids (e.g., low concentration HCl solution or fumigation
926 Verbruggen et al. 2010a; Heiri et al. 2012; Belle et al. 2014) or a buffered NH_4Cl solution
927 (Verbruggen et al. 2010b). Commonly used chemical pre-treatment methods apparently
928 do not strongly affect the $\delta^{13}C$ values of chitinous and fossilizing structures (head capsules)
929 of chironomid larvae. In contrast, the oxygen isotope composition of chironomid cuticles
930 can be strongly affected by strong alkali or acid treatments (Verbruggen et al. 2010a;
931 Lombino 2014) and such treatments can lead to selective removal of protein or chitin from
932 the cuticles. Furthermore, acid solutions can promote oxygen exchange between the

933 solution and the cuticles (Verbruggen et al. 2010b). Less information is available on the
934 effects of chemical pre-treatments on the fossilizing structures of other chitinous
935 invertebrates and on the N- and H-isotope composition of remains. Treatment with 10 %
936 KOH solution has no apparent effect on the C, O and N isotope composition of the
937 ephippia sheaths of *Daphnia pulicaria* (Schilder et al. 2015a).

938 Taphonomic changes in chitinous arthropod cuticles have mainly been studied for
939 marine crustaceans, whereas much less information is available for lacustrine
940 invertebrates. Major losses of chitin and protein have been observed during the first
941 weeks of biodegradation of shrimp cuticle and other arthropod remains (Stankiewicz et al.
942 1998; Schimmelmann et al. 1986). The C, N, O and H isotope composition was found to be
943 preserved during partial degradation of chitin leading Schimmelmann et al. (1986) to
944 conclude that chitin in ancient archaeological deposits can still be expected to faithfully
945 carry the original isotopic signature. However, as mentioned above, experimental results
946 based on decapod cuticles may not be representative for chitinous cuticles of other
947 aquatic invertebrate groups. Perga (2011) studied the effects of degradation of cladoceran
948 exoskeletons in in lake water and anoxic sediments. In degradation experiments with
949 cladoceran exoskeletons changes in $\delta^{13}C$ were very small (<1 ‰) and happened within the
950 first three months (Perga 2011). No major changes in $\delta^{15}N$ were observed if exoskeletons
951 were incubated in sediments, but effects were larger in oxic or anoxic lake water (+2 to +5
952 ‰). The chemical structure of chironomid head capsules isolated from 15,000-year old
953 sediments was shown to be very similar to modern head capsules, suggesting that no
954 extensive chitin or protein decomposition occurred in these structures after initial
955 decomposition (Verbruggen et al. 2010a). Down core analyses demonstrated that
956 chironomid head capsules from sediments 11,000-15,000 years old were still
957 characterized by the centennial- to millennial-scale changes in $\delta^{18}O$ values that
958 characterized precipitation and lake water compositions in Europe, during the end of the
959 last ice age (Verbruggen et al. 2010b; Lombino 2014).

960 In summary, available evidence indicates that pre-treatment and initial decomposition
961 affects the isotope composition of different elements differently. Only very small sample
962 pre-treatment effects are reported for $\delta^{13}C$ values of invertebrate remains. More
963 significant effects have been described for $\delta^{18}O$ values, whereas little information is

964 available for $\delta^{15}N$ and δD values. Similarly, some evidence indicates that effects of
965 degradation may be more relevant for $\delta^{15}N$ than $\delta^{13}C$ values of invertebrate remains,
966 whereas studies describing these effects for $\delta^{18}O$ or δD values are lacking. Nevertheless, it
967 appears that chitinous remains preserve well and (as far as evidence is available) retain
968 their isotopic signature in lake sediments once initial stages of decomposition have been
969 completed, which is also confirmed by the interpretable shifts in isotope composition
970 reported for invertebrate remains in late Quaternary sediment records (see sections 4.4
971 and 4.5 below).
972
973 4.4 *Stable carbon and nitrogen isotope records based on chitinous invertebrate remains*
974
975 *4.4.1 Impacts of land use, eutrophication, and food sources*
976
977 Section 4.2.1 discussed how $\delta^{13}C$ and $\delta^{15}N$ values of invertebrates and their remains
978 closely reflect dietary composition, usually with a systematic offset. This makes it possible
979 to provide information about the diet of invertebrates using the carbon and nitrogen
980 isotope composition of their remains. Diets usually consist of a mixture of sources, each
981 with its own isotope composition, and more nutritious or more easily digestible sources
982 may therefore be assimilated preferentially (Kamjunke et al. 1999; Goedkoop et al. 2006).
983 The food sources available to invertebrates also strongly depend on invertebrate feeding
984 ecology and will differ for detritivores, grazers, filterer-feeders, or predators (Merritt et al.
985 2008; Thorp and Rogers 2015), so it is crucial to understand the parent organism's modern
986 habitat and ecology when interpreting stable isotope values (more detailed examples in
987 van Hardenbroek et al. (2012, 2014)). Overall, $\delta^{13}C$ and $\delta^{15}N$ values of invertebrate
988 remains and bulk sediment tend to follow the same trends in many lakes (Struck et al.
989 1998; Wooller et al. 2008; Griffiths et al. 2010; van Hardenbroek et al. 2014; Kattel et al.
990 2015), but significantly different $\delta^{13}C$ profiles have also been reported (Perga et al. 2010;
991 Schilder et al. 2017). Recent palaeolimnological studies that have measured taxon-specific
992 $\delta^{13}C$ values of chironomid head capsules indicate that benthic invertebrates are sensitive
993 to changes in available carbon sources (van Hardenbroek et al. 2014; Belle et al. 2017a,b;
994 Schilder et al. 2017), including methane-derived carbon (see section 4.4.2).

995 Planktonic cladocerans are typically filter-feeding on particulate organic matter
996 (POM) in the water column, predominantly consisting of algae and bacteria (Kamjunke et
997 al. 1999) and their stable isotope composition follows that of their food sources during the
998 annual cycle (Perga and Gerdeaux 2006; Morlock et al. 2017). As primary consumers,
999 Cladocera are very sensitive to changes in primary productivity and the stable isotope
1000 composition of their remains can be used to trace the impact of anthropogenic nutrient
1001 enrichment of lakes and recovery from it (Perga et al. 2010; Frossard et al. 2014; van
1002 Hardenbroek et al. 2014). In large lakes, there can be a strong relationship between
1003 Cladocera $\delta^{13}C$ values and the CO_2 concentration in the lake water (Smyntek et al. 2012).
1004 This is due to the strong relationship between algal carbon fractionation and dissolved CO_2
1005 availability (e.g., Hollander and McKenzie 1991; Laws et al. 1995). Based on this
1006 mechanism, Perga et al. (2016) have shown that in three large, temperate lakes the $\delta^{13}C$
1007 values of Cladocera remains can be used as an indicator of past summer surface water CO_2
1008 concentrations. In smaller lakes, however, Cladocera are more likely to also incorporate
1009 allochthonous or methanogenic (see section 4.4.2) carbon, which can compromise the
1010 relationship between Cladocera $\delta^{13}C$ values and CO_2 concentrations.
1011 Bryozoa are another invertebrate group that produces resting stages (statoblasts),
1012 which have been used for stable isotope analyses (van Hardenbroek et al. 2016). Bryozoa
1013 are mostly sessile organisms, filter-feeding on suspended POM, which consists to a
1014 considerable extent of algae and bacteria (Kaminski 1984). Bryozoan $\delta^{13}C$ values generally
1015 follow the trend in $\delta^{13}C$ values of POM or, in down core studies, SOM (Turney 1999; van
1016 Hardenbroek et al. 2014, 2018; Morlock et al. 2017; Rinta et al. 2016).
1017 The impact of urban and agricultural pollution to lakes can result in the increase of
1018 $\delta^{15}N$ values of SOM of aquatic origin (Cabana and Rasmussen 1996; Laevitt et al. 2006). A
1019 number of studies have found that $\delta^{15}N$ of Cladocera remains and SOM are related (Struck
1020 et al. 1998; Wooller et al. 2008), but the addition of nutrients can disturb this relationship.
1021 In Lake Annecy, Perga et al. (2010) observed increasing $\delta^{15}N$ values of SOM and *Bosmina*
1022 carapaces during the 1960s and 1970s eutrophication. During re-oligotrophication $\delta^{15}N$
1023 values of SOM decreased again, but $\delta^{15}N$ values of *Bosmina* remained high, possibly as a
1024 result of a shift in their diet from algae to flagellates: changing the trophic position of
1025 *Bosmina*. Griffiths et al. (2010) investigated the effect of marine-derived nutrients from

1026 sea-bird colonies on arctic lakes and showed that this input led to an increase in SOM $\delta^{15}N$
1027 values. Chironomid remains showed the same trend, albeit with greater variability,
1028 whereas *Daphnia* remains did not show any change at all, indicating that both groups of
1029 organisms can assimilate different sources of nitrogen, at least in the periods when the
1030 fossilizing structures are formed.

1031 Aquatic food webs can be highly complex, with large spatial and temporal variations in
1032 the quantity of carbon and nitrogen sources as well as the isotope composition of these
1033 sources. Attempts to interpret sedimentary stable isotope records of invertebrate remains
1034 to estimate the relative contribution of different dietary components (algae,
1035 allochthonous OM, bacterial biomass) requires reliable estimates of the stable isotope
1036 composition of these dietary components. This is challenging in modern food web studies,
1037 and even more so in palaeoenvironmental applications. Emerging compound-specific
1038 stable isotope analysis of microbial and algal compounds (e.g., Castañeda and Schouten
1039 2011; Middelburg 2014; Taipale et al. 2015), combined with stable isotope analysis of
1040 specific organic remains may hold the key to a substantial increase in understanding
1041 trophic interactions over long timescales.

1042

1043 *4.4.2 Methane-fuelled food webs*

1044

1045 Notably low $\delta^{13}C$ values of invertebrates compared with $\delta^{13}C$ values of algae and SOM
1046 occur where ^{13}C-depleted methane and methane oxidizing bacteria (MOB) provide carbon
1047 sources for lacustrine food webs (e.g., Gebruk 1993; Bunn and Boon 1993; Jones et al.
1048 2008; Devlin et al. 2015; Grey 2016). Filter-feeding and deposit-feeding invertebrates can
1049 partially feed on MOB biomass (or on ciliates feeding on MOB) and their $\delta^{13}C$ values
1050 become lower with increasing contributions of methane-derived carbon.

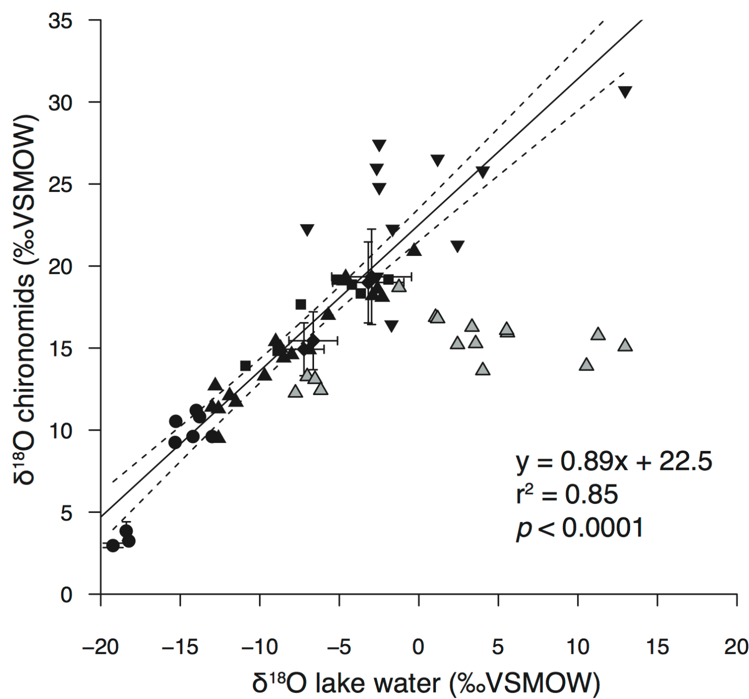
1051 The $\delta^{13}C$ values of some invertebrate groups and their remains in surface
1052 sediments are systematically related to methane concentrations or fluxes in lakes, at least
1053 in some regions and lake types. For example, $\delta^{13}C$ values of Chironomini larval head
1054 capsules and of *Daphnia* ephippia were negatively correlated to diffusive methane fluxes
1055 in 17 lakes studied in Sweden and arctic Siberia (van Hardenbroek et al. 2013b). Also, $\delta^{13}C$
1056 values of *Daphnia* ephippia in 15 European lakes were found to correlate to lake methane

1057 concentrations (Schilder et al. 2015b) and $\delta^{13}C$ values of Chironomini larvae in 6 lakes in
1058 arctic Alaska were correlated to methane oxidation rates (Hershey et al. 2015). These
1059 findings agree with the observation that, in small European lakes, the abundance of ^{13}C-
1060 depleted fatty acids in surface sediments, originating at least partially from MOB, also
1061 increases with increasing CH_4 concentrations (Stötter et al. 2018). This suggests that MOB
1062 concentrations in these lakes also increase with CH_4 concentrations. Sediment core studies
1063 indicated higher uptake of methane-derived carbon in certain invertebrate groups (mainly
1064 belonging to the chironomids and cladocerans) with increased primary productivity in
1065 lakes. This was either driven by anthropogenic nutrient addition (Frossard et al. 2015;
1066 Belle et al. 2016a, b; Rinta et al. 2016; Schilder et al. 2017), or triggered by warmer/wetter
1067 climatic conditions (Wooller et al. 2012; van Hardenbroek et al. 2013b). Quantifying the
1068 uptake of methane-derived carbon using isotope mixing models is possible, using the $\delta^{13}C$
1069 values of the two end member food sources, MOB and algae. POM/SOM has been used as
1070 approximation for algae (Wooller et al. 2012; Belle et al. 2014, 2015b, 2016b; Schilder et
1071 al. 2017), although this approach is associated with high levels of uncertainty due to
1072 variations in $\delta^{13}C$ values of methane and algal material between lakes.

1073 Measurements of δD values of invertebrates can also potentially be used to assess
1074 the contribution of MOB to their diet, since methanogens also strongly discriminate
1075 against the heavier D, and the resulting low δD values are transferred to certain taxa of
1076 chironomid larvae (Deines et al. 2009; Belle et al. 2015b). In addition, in lakes with
1077 significant contributions of methane to the lacustrine food web, ^{14}C analyses of
1078 invertebrate remains may in some situations be used to constrain the amount of old
1079 carbon entering the food web: e.g., via methane from decomposing Pleistocene deposits
1080 (Wooller al. 2012; Elvert et al. 2016). However, in hard water lakes this approach is further
1081 complicated by other old carbon sources such as carbonates derived from local bedrock.

1082 The stable isotope composition of invertebrates can vary seasonally in relation to
1083 MOB availability, notably during autumn turnover when methane stored in the
1084 hypolimnion is mixed (e.g., Grey et al. 2004; Kankaala et al. 2010; Yasuno et al. 2012). The
1085 $\delta^{13}C$ values of invertebrate remains, however, are more likely to represent average values
1086 of these seasonal variations (Morlock et al. 2017; Schilder et al. 2017, 2018; van
1087 Hardenbroek et al. 2018). Likewise, spatial variation of methane production in lakes can

1088 lead to spatial variability of $\delta^{13}C$ values of invertebrates (Deines and Grey 2006; Agasild et
1089 al. 2013), which can also be observed in the remains (van Hardenbroek et al. 2012; Belle et
1090 al. 2015a). Interesting examples of spatial variability of methane-derived carbon are
1091 provided by Frossard et al. (2014, 2015), who observed an expansion (to shallower depths)
1092 of the anoxic hypolimnion with ongoing eutrophication. As a result, chironomid head
1093 capsule $\delta^{13}C$ values became progressively lower, a process that was first observed in the
1094 deepest cores, followed a few decades later in shallower cores.
1095
1096 *4.5 Stable oxygen and hydrogen isotope records based on chitinous invertebrate remains*
1097
1098 The $\delta^{18}O$ values of organic remains can provide information on the $\delta^{18}O$ values of lake
1099 water in which the organic remains were produced. Wooller et al. (2004) were the first to
1100 measure $\delta^{18}O$ values of chironomid head capsules in lake sediment records and
1101 demonstrated that, within analytical error, chironomid $\delta^{18}O$ values agree with the
1102 expected $\delta^{18}O$ values of modern regional precipitation. The relationship between $\delta^{18}O$
1103 values of lake water and chironomid remains has since been further validated and
1104 explored via laboratory experiments (section 4.2.2) and field surveys. The field
1105 relationship has been reproduced and further explored over larger transects and multiple
1106 regions (Fig. 5), including Europe (Verbruggen et al. 2011; Lombino 2014), South America
1107 (Mayr et al. 2015), Greenland (Lasher et al. 2017), and Australia (Chang et al. 2016, 2017).
1108 In lakes with short residence times (≤ 1 year), i.e. open basins, the relationship between
1109 chironomid head capsules and lake water $\delta^{18}O$ values is strong (Fig. 5). Here, chironomid
1110 $\delta^{18}O$ values are enriched relative to lake water by ~ 22.5 ‰. This relationship appears to
1111 fail in closed basin lakes with long residence times. Sampled lake water from such lakes
1112 may not reflect $\delta^{18}O$ values of waters during the growing season due to evaporation
1113 effects. In lakes where lake water – chironomid enrichment factors differ greatly from the
1114 mean relationship demonstrated in open basins, an assessment of lake hydrology is
1115 necessary to clarify what chironomid $\delta^{18}O$ values are recording.
1116
1117



1118

1119

1120 **Fig. 5**: *Relationship between δ^{18}O values of (lake) water and δ^{18}O values of chironomid*
1121 *head capsules from surface sediment samples. Plotted data points are from Verbruggen et*
1122 *al. 2011 (closed triangles), Lombino 2014 (closed diamonds), Mayr et al. 2015 (closed*
1123 *squares), Chang et al. 2016 (shaded triangles), Chang et al. 2017 (closed inverted*
1124 *triangles), and Lasher et al. 2017 (closed circles). Error bars indicate 1 standard deviation*
1125 *variability in replicate data from individual sites, where available. The linear regression*
1126 *excludes data points from Chang et al. 2016, as these contain closed systems with high*
1127 *evaporation, where the δ^{18}O of sampled lake water may not be indicative of δ^{18}O value of*
1128 *water during chironomid growth.*

1129

1130 Although chironomid larvae are often abundant in lake sediments, recalcitrant
1131 organic structures of other aquatic organisms have also been used to measure δ^{18}O
1132 values, including cladoceran ephippia (Verbruggen et al. 2011; Schilder et al. 2015a) and
1133 elytra of aquatic beetle genera *Helophorus* and *Hydroporus* (van Hardenbroek et al.
1134 2013a).

1135 Based on these demonstrated positive relationships between the δ^{18}O values of
1136 organic invertebrate remains and those of lake water (Fig. 5), a growing number of
1137 palaeoecological and palaeoclimatic studies have used δ^{18}O analyses of organic

1138 invertebrate remains as a palaeoclimatic proxy. Records have largely been developed from
1139 sites from higher latitudes including Greenland (Wooller et al. 2004; Lasher et al. 2017),
1140 Iceland (Wooller et al. 2008), Alaska (Wooller et al. 2012; Graham et al. 2016) and
1141 Svalbard (Arppe et al. 2017; Luoto et al. 2018), but also from mid-latitudes (Verbruggen et
1142 al. 2010b; Lombino 2014). These $\delta^{18}O$ records do not only reflect temperature changes,
1143 but a combination of drivers including (1) seasonality and source region of precipitation,
1144 (2) the balance between inputs (precipitation, snow melt, inflow) and evaporation, and (3)
1145 composition of invertebrate assemblages, their habitat and ecology. As a result, the $\delta^{18}O$
1146 records may in some situations be more variable than independent temperature
1147 reconstructions (e.g., Wooller et al. 2004, 2008, 2012). In some records, however,
1148 chironomid $\delta^{18}O$ values closely follow minor climate oscillations (Verbruggen et al. 2011b;
1149 Lombino 2014; Arppe et al. 2017). Chironomid $\delta^{18}O$ has also been used to show the
1150 reduction in freshwater availability attributed to the local extinction of woolly mammoth
1151 on St. Paul Island (Graham et al. 2016).

1152 In contrast to the $\delta^{18}O$ values from chironomids and other aquatic invertebrates, the
1153 δD values of these organisms have been found to be primarily controlled by the δD values
1154 of diet rather than source water (section 4.2.2). Given this finding, δD values from
1155 chironomids have found to be a valuable dietary biomarker, and used to trace the use of
1156 terrestrial and methanogenic carbon by aquatic organisms (Deines et al. 2009; Karlsson et
1157 al. 2012; Belle et al. 2015b; Mariash et al. in press, and see section 4.4.2).

1158

1159 4.6 *Other organic remains*

1160

1161 *4.6.1 Aquatic plant macrofossils*

1162

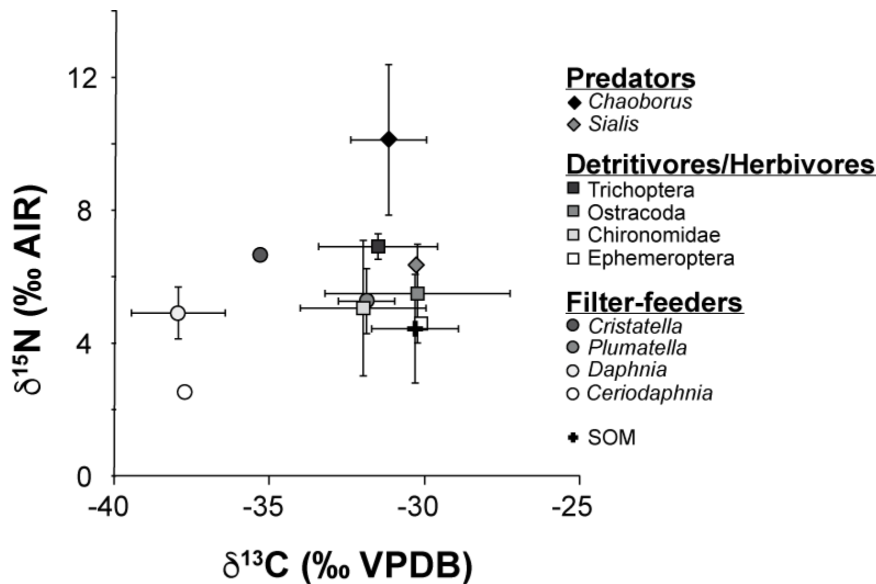
1163 Records of $\delta^{13}C$ and $\delta^{18}O$ of aquatic cellulose in lake sediments are widely available (e.g.,
1164 Wolfe et al. 2007; Mayr et al. 2015; Street-Perrot et al. 2018) but are not discussed here. In
1165 contrast, continuous, high-resolution sediment records of aquatic plant remains have only
1166 in rare cases been analysed for their stable isotope composition. For example, Turney
1167 (1999) found little variation in the $\delta^{13}C$ values of terrestrial plant remains compared with
1168 $\delta^{13}C$ of SOM in two kettle-hole lakes in the UK, while $\delta^{13}C$ values of aquatic plant remains

1169 (*Potamogeton*) was more variable, possibly in response to changes in productivity. A
1170 systematic study of the difference between δ^{13}C values of *Potamogeton* and δ^{13}C values of
1171 DIC in lakes on the Tibetan Plateau and Yakutia (Herzschuh et al. 2010a), demonstrated
1172 that this difference is dependent on the growth rate, which can be interpreted in terms of
1173 productivity and applied to sediment records. Another study linked variations in δ^{15}N
1174 values of aquatic macrophyte remains with N-limitation and productivity (Herzschuh et al.
1175 2010b). When the organic matter produced within lakes by aquatic macrophytes and algae
1176 is an important component of SOM, the stable isotope composition of SOM and algae (e.g.
1177 *Botryococcus*) are strongly coupled, even on millennial time scales (Heyng et al. 2012). In a
1178 study of 40 lakes on the Tibetan Plateau δ^{13}C values of aquatic macrophytes correlated
1179 positively with δ^{13}C values of SOM, although much stronger correlations existed between
1180 δ^{13}C values of *Potamogeton* and δ^{13}C values of mid-chain n-alkanes, which are largely
1181 produced by the macrophytes (Aichner et al. 2010). This study also demonstrated the
1182 effect of terrestrial organic matter on δ^{13}C of SOM and the importance of measuring δ^{13}C
1183 values of different carbon sources to understand palaeolimnological records of SOM δ^{13}C
1184 values (e.g., Gu et al. 2006; Das et al. 2008; Drew et al. 2008). The use of spooling-wire
1185 microcombustion IRMS makes is possible to analyse very small samples of carbonised
1186 terrestrial plant material (Urban et al. 2010, 2013). This approach could also be applied to
1187 aquatic plant remains to disentangle the relative importance of terrestrial and aquatic
1188 carbon sources in SOM.
1189 Stable oxygen isotopes of aquatic plant macrofossils have been studied in a similar
1190 way as aquatic invertebrate remains (section 4.5), indicating a strong relationship between
1191 δ^{18}O values of lake water and aquatic moss fragments (Zhu et al. 2014; Lasher et al. 2017).
1192 Furthermore, stable oxygen isotope values of aquatic moss macrofossils and those of
1193 purified cellulose are strongly correlated in some lakes, with a reported mean δ^{18}O offset
1194 from cellulose of ~2.7 ‰ for aquatic vascular plants and ~1.3 ‰ for aquatic mosses (Zhu et
1195 al. 2014).
1196
1197 *4.6.2 Fish remains*
1198
1199 Fish have crucial top-down impact on the structure of lake ecosystems, generally taking up

1200 the highest trophic positions in lacustrine food webs. Fish remains (e.g., scales, teeth,
1201 bones and otoliths) can be preserved in lake sediments (e.g., Wooller et al. 2015) and the
1202 assemblages in surface sediments broadly reflect the species in living fish communities
1203 (Davidson et al. 2003). Fish remains contain organic and calcified moieties. The organic
1204 fraction, mostly collagen, is derived from diet, whereas the calcified fraction can come
1205 from both diet and DIC in lake water (Hoie et al. 2003). A good relationship between the
1206 δ^{13}C values of scales and muscle tissue was found, with systematic offsets of ca. 2.5 to 4 ‰
1207 and –1.5 to 0 ‰ for δ^{13}C and δ^{15}N, respectively (Perga and Gerdeaux 2003; Kelly et al.
1208 2006). Decalcifying scales can improve this relationship (Perga and Gerdeaux 2003), but
1209 has a minimal effect on δ^{13}C and δ^{15}N values (Ventura and Jeppesen 2010). Stable isotope
1210 analysis of fish remains from lake sediments (Patterson et al. 1993; Wooller et al. 2015),
1211 archived fish scale collections (Gerdeaux and Perga 2006), and fish remains from
1212 archaeological settings (Häberle et al. 2016) hold great potential for investigating changes
1213 in food chain length and trophic interactions in response to, for example, eutrophication,
1214 non-native species introductions, fisheries and aquaculture.
1215
1216 *4.7 Future directions*
1217
1218 An increasing number of studies use stable isotope analyses on organic remains in
1219 sediment records from arctic, boreal, and temperate regions. In contrast, subtropical and
1220 tropical sediment records are almost completely absent, although the same pattern
1221 applies to modern limnological studies (e.g., Iglesias et al. 2017; Sanseverino et al. 2012).
1222 Other aspects that deserve further study are understanding drivers of spatial variability
1223 within lake basins, for example by comparing shallow and deep-water cores (Frossard et
1224 al. 2015), and by measuring the stable isotopes on components of the modern lake
1225 (eco)system and catchment to better understand taphonomic processes and constrain
1226 palaeolimnological interpretations (Morlock et al. 2017; Arppe et al. 2017). Related to this
1227 is the continued need for controlled growth and feeding experiments, especially those that
1228 investigate temperature-dependent fractionation (Schilder et al. 2015a).
1229 A novel direction is the use of dual δ^{13}C and δ^{15}N values from organic remains of
1230 organisms, at different trophic positions throughout a sediment record, which allows the

1231 study of changes in food web structure over time. Initial work on invertebrate remains
1232 from surface sediments indicates that ranges of ~10 ‰ for both $\delta^{13}C$ and $\delta^{15}N$ values can
1233 be observed (Fig. 6), similar to the ranges found in soft tissues of these organisms (e.g.,
1234 Jones and Grey 2011). Related to this is the potential for using $\delta^{34}S$ values (e.g., Grey and
1235 Deines 2005) to understand trophic relationships as preserved in fossil assemblages.

1236



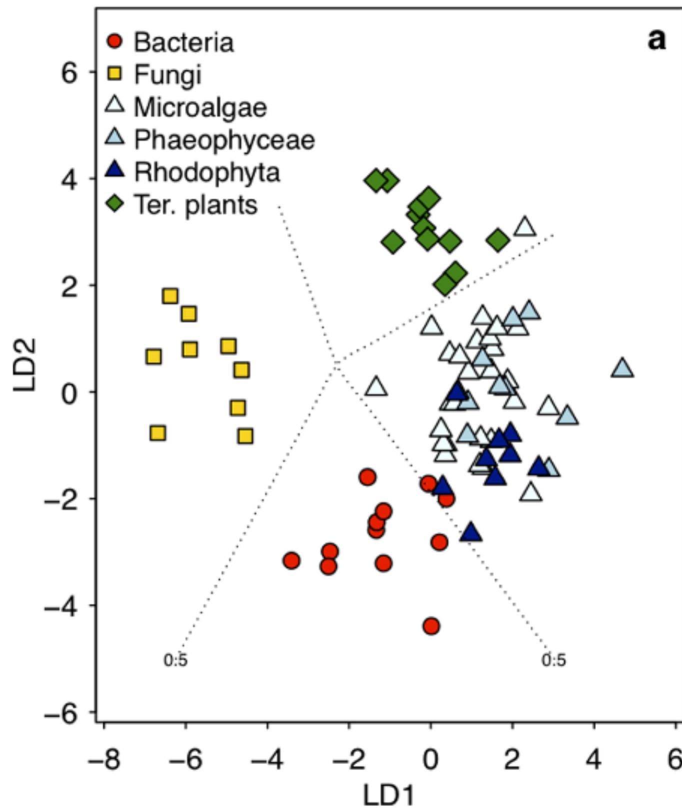
1237
1238

1239 **Fig. 6**: *$\delta^{13}C$ and $\delta^{15}N$ values of invertebrate remains and SOM from surface sediments in*
1240 *Lake De Waay, The Netherlands (van Hardenbroek, Heiri, Schilder, Wooller unpublished*
1241 *data).*

1242

1243 A further approach to understanding food web structure is by analysing stable
1244 carbon isotopes of essential amino acids in tissues (Larsen et al. 2009). These amino acids
1245 have specific stable isotope values, or isotope "fingerprints", depending on the primary
1246 producer type that synthesised the essential amino acids: aquatic photosynthetic (derived
1247 from dissolved organic carbon), terrestrial photosynthetic (derived from atmospheric
1248 carbon dioxide) and microbial (Fig. 7). These fingerprints are independent of the isotope
1249 composition of the carbon source that was originally used to synthesise the essential
1250 amino acids and they are retained in consumer tissues (Larsen et al. 2009). These
1251 fingerprints can also be used in mixing model approaches to determine the proportional
1252 contribution of different primary production sources (e.g. terrestrial vs. aquatic vs.

1253 microbial) to consumers (Larsen et al. 2009; Larsen et al. 2013). This approach has been
1254 tested in some marine ecosystems (Larsen et al. 2013; Arthur et al. 2014) and initial
1255 palaeolimnological work is promising (Carstens et al. 2013).
1256
1257



1258
1259

1260 **Fig. 7:** *Linear discriminant (LD) function analysis of the stable carbon isotope composition*
1261 *of essential amino acids demonstrates the "fingerprinting" of a range of primary*
1262 *production sources (from Larsen* et al. *2013).*

1263
1264

1265 **5. Stable isotope modelling in palaeolimnology**

1266

1267 Stable isotopes, for all their attractive qualities as environmental, ecological and
1268 behavioural tracers, are at times rather blunt instruments: tissue isotope compositions are
1269 influenced by a wide range of potential drivers, so that interpreting the cause(s)
1270 contributing to any measured variation in isotope compositions can be challenging.

Modern ecology and hydrology have greatly profited from explicit modelling of variations in stable isotope composition due to known stressors, impacts, and processes, which can help to separate realistic from unrealistic scenarios even in complicated situations with overlapping variabilities due to seasonal changes, different drivers, and site- or taxon-specific isotopic offsets. This type of modelling is mostly lacking when interpretations are made in palaeolimnological studies, and some examples are outlined here that show how approaches developed in modern modelling studies could be applied to palaeorecords.

This section will focus on the potential and pitfalls of modelling carbon and nitrogen isotopes in modern and palaeo food webs and how this can contribute to the interpretation of stable isotope records based on fossil remains in lake sediments. Recent advances in the use of stable isotopes of oxygen and hydrogen in (palaeo)hydrological modelling are only briefly highlighted, as the topic was reviewed recently (Jones et al. 2016a).

5.1 Carbon and Nitrogen

In a review of the use of stable isotopes in ecology, Boecklen et al. (2011) identify 46 separate sources of variation in stable isotope compositions (predominantly $\delta^{13}C$ and $\delta^{15}N$ values) of organisms. In their review, the isotope composition of an organism is viewed as an emergent property of that organism, influenced by the factors that are frequently the subject of palaeontological investigation such as diet, trophic level, and environmental variability (e.g., habitat and seasonality), but also by a large range of additional physiological, ecological and biochemical factors that are often difficult to assess in palaeoecological and palaeoenvironmental studies such as temporal and spatial variations in stable isotope compositions of nutrients and food sources and physiological influences on isotopic fractionation. Interpretations of stable isotope data are therefore vulnerable to making overly simplified assumptions, and it is helpful to consider how sources of uncertainty and variance can impact the validity of isotope-based ecological or environmental interpretation before identifying possible solutions to deal with uncertainty.

In the simplest ideal case, the isotope composition of carbon and nitrogen in consumers reflects the isotope composition of their prey. Where a consumer derives

1303 resources from two or more isotopically distinct prey sources, the relative importance of
1304 those sources can be determined from a mass-balance based isotope mixing model (for
1305 example, in the case of carbon derived from phytoplankton and methane, see section
1306 4.4.2). Measuring stable isotopes on 'pure' algal or bacterial biomass is challenging in
1307 modern food webs and culturing experiments (Templeton et al. 2006; Vuorio et al. 2006).
1308 This hinders defining the isotope values of sources at the base of lake food webs and leads
1309 to assumptions and uncertainties in mass-balance models. Compound-specific stable
1310 isotope analysis of biomarkers typical for particular algal (n-alkanes, pigments, fatty acids,
1311 amino acids) and bacterial groups (fatty acids, hopanols, amino acids), are now emerging
1312 as palaeolimnological tools (Huang et al. 1999; Aichner et al. 2010; Castañeda and
1313 Schouten 2011; Larsen et al. 2013; Middelburg 2014; Taipale et al. 2015; Elvert et al. 2016)
1314 and will potentially address this issue.

1315 Mass-balance type models also assume the isotopic offset between diet and tissue
1316 is known (see, e.g., McCutchan et al. 2003 and sections 4.2). More advanced Bayesian
1317 mixing models such as SIAR (Parnell et al. 2013), mixSIR (Semmens et al. 2009) and FRUITS
1318 (Fernandes et al. 2014) have been developed to incorporate uncertainties in diet-tissue
1319 isotopic fractionation and to consider isotopic distributions of potential food sources.
1320 However, mixing models are still simplifications of food webs with relatively limited
1321 consideration of temporal or spatial variation in either isotope compositions or trophic
1322 interactions. Model outputs should be interpreted with care to avoid oversimplification, as
1323 highlighted by authors of isotopic mixing models and isotope metrics themselves (Layman
1324 et al. 2012; Phillips et al. 2014), but can nevertheless provide estimates of the likelihood of
1325 different scenarios, although it remains difficult to include temporal and spatial variability.

1326 Seasonality has a marked effect on biogeochemical cycles, associated nutrient
1327 isotope compositions, food availability and food web structure (e.g., Woodland et al.
1328 2012; Junker and Cross 2014; Visconti et al. 2014). Most isotope-based ecological analyses
1329 either explicitly compare the isotope composition of the animal in question to a reference
1330 baseline animal, assuming a constant isotopic baseline or infer that temporal change in
1331 consumer isotope values reflects changes in the baseline isotope compositions. In all these
1332 cases, short-term (e.g., seasonal) variability in isotope baselines can complicate
1333 interpretations of consumer isotope data. This will be particularly pertinent for consumers
1334 with high growth or metabolic rates such as chironomids, cladocerans, and

1335 ephemeropterans, where dynamic changes in baseline isotope values will be rapidly
1336 incorporated into consumer tissues, although this is mediated to some extent because
1337 many fossil specimens are needed for one stable isotope measurement, and therefore
1338 represent a larger period of time. Woodland et al. (2012) built a time dynamic isotope
1339 mixing model by coupling combined time-dependent functions of temporal baseline $\delta^{13}C$
1340 variation and functions predicting consumer isotope composition as a function of growth.
1341 Relaxing assumptions of isotopic equilibrium, both in terms of consumer growth and
1342 isotopic baselines, resulted in different and more realistic reconstructions of the
1343 contribution of benthic resources to diets. Studies that quantify and model the isotope
1344 variability in aquatic organisms are essential for understanding stable isotope records
1345 from lake sediment cores, although few modern surveys are designed with
1346 palaeoenvironmental applications in mind (e.g., von Grafenstein et al. 1999b; Dixit et al.
1347 2015; Morlock et al. 2017; Schilder et al. 2017; section 4.5).

1348 When different locations and palaeolimnological studies are considered,
1349 potentially confounding aspects of spatial/geographical variability, diagenesis and time
1350 averaging are also added. Very quickly the number of potential variables that could be
1351 drawn on to explain an observed temporal change either in means or distributions of
1352 organism stable isotope compositions increases, leading to a range of possible scenarios
1353 that could explain a measured isotopic response. One approach to quantitatively choosing
1354 between alternative mechanisms responsible for measured isotopic variability is through
1355 mechanistic modelling. For example, Magozzi et al. (2017) estimated spatial variations in
1356 inter-annual ranges of $\delta^{13}C$ values for phytoplankton, and show that these vary
1357 systematically with latitude, at less than 1.5 ‰ in equatorial regions to >9 ‰ in highly
1358 seasonal arctic systems. Estimates of spatial and temporal variability in isotopic baselines
1359 can form the basis for more complex simulations of isotopic variability in food webs under
1360 differing scenarios, and with differing sampling protocols.

1361 As a simple conceptual example, a time series of $\delta^{13}C$ and $\delta^{15}N$ values from benthic
1362 insect remains from a sediment record within a small temperate lake can be assumed.
1363 Changes in isotope composition and distributions through time are observed and the
1364 question is what contributed to the observed change. A change in external conditions is
1365 suspected (e.g., a change in the supply of terrestrial organic matter into the lake), but
1366 uncertainty remains whether any other potential driving mechanisms (e.g., Boecklen et al.

1367 2011) or systematic changes in sampling associated with sedimentation could have
1368 contributed to the observed pattern. To set up a basic conceptual model of the isotopic
1369 systematics in the lake it is necessary to establish variables associated with isotope
1370 composition of nutrient inputs to the lake, pelagic production and benthic production.
1371 Each of these variables will have defined seasonal, inter-annual and longer-term
1372 variability, which can be varied in the model.

1373

1374 **Table 2:** *Example of variables needed to derive a basic simulation of time series of lake*
1375 *sediment isotopic records.*

Nutrient input	Primary production	Benthic production	Taphonomy
The number of isotopically distinct sources of carbon and nitrogen entering the lake. Proportional contribution of each nutrient source to primary production. The isotope composition of each source. **Temporal variation in the above**	Isotopic fractionation associated with primary production (with uncertainty) – can be linked to temperature and/or cell growth rate. Proportion of primary production formed in each season. **Temporal variation in the above**	Proportional contributions of detrital and pelagic organic matter to sediment. Contribution of methane to sediment organic matter. **Temporal variation in the above**	Time of year represented in the sample. Number of years assimilated into a single sample. Variability in sample time averaging. Differential loss of isotopically distinct fractions of production.

1376

1377 Simple conceptual model systems building on variables suggested in Table 2 allow
1378 investigators to explore sensitivity of the measured output (the expected means or
1379 isotopic distributions of a population of invertebrates) to a range of variables, including
1380 seasonality, benthic vs. pelagic production, temperature-related differences in isotopic
1381 fractionation in phytoplankton, proportional methane contributions and terrestrial detrital
1382 matter input. In addition, the sensitivity of the system to naturally uneven sampling can be
1383 explored. Using this type of modelled time series in tandem with palaeoenvironmental
1384 records from lake sediments make it possible to test the likelihood of alternative scenarios

1385 and interpretations. This is not unlike studies that test models of fossil assemblage data
1386 from sediment record to understand sensitivity or response of lake ecosystems to critical
1387 transitions (e.g., Seddon et al. 2014; Doncaster et al. 2016).
1388
1389 *5.2 Oxygen and hydrogen*
1390
1391 An increasing number of palaeoenvironmental studies uses the systematic relationship
1392 between climatic variables and δD and δ^{18}O values measured from environmental water
1393 (Dansgaard 1964; Gat 1996) to interpret proxies of palaeo lake water recorded in
1394 sediment archives (Darling et al. 2006, Jones et al. 2016a; Swann et al. 2018). Mass-
1395 balance models based on the linear resistance model of Craig and Gordon (1965) for the
1396 isotope composition of evaporating water have been used to explain large proportions of
1397 the variability in lake water isotopes, although the many steps between the isotope
1398 composition of precipitation and the isotopic signal preserved in proxy records require a
1399 thorough understanding of the 'proxy systems' and how they filter and adapt this
1400 variability (Feng et al. 2016; Gibson et al. 2016; Jones et al. 2016b). Jones et al. (2016a)
1401 suggest that such proxy system models (PSMs) could potentially be used to predict
1402 'forward' modelled proxy time series. These predictions could then be tested against
1403 output from General Circulation Models (GCMs) that have stable water isotope physics
1404 included, although downscaling GCM output to make it compatible with local palaeo
1405 records is challenging (e.g., Sturm et al. 2010). The inclusion of stable water isotopes in
1406 GCMs now also allows quantitative validation of GCMs using water isotope data from
1407 palaeoenvironmental records (especially when based on large, amalgamated data sets
1408 that go beyond the local scale, as described by Horton et al. 2016). The water isotope
1409 component of GCMs also creates opportunities for hypothesis-driven exploration of
1410 palaeo climate data based on GCM output (e.g., Holmes et al. 2016). Further effort is
1411 needed from the modelling, monitoring, and palaeo communities to quantifying the
1412 physical processes in the atmosphere that drive spatial and temporal heterogeneity in
1413 water isotopes. It might be challenging, however, to find 'well-behaved' lake systems due
1414 to large variations and complexity of local hydrology (both spatially and temporally) that
1415 affect the stable isotope composition of lake systems compared to marine systems.
1416

6. Concluding remarks

This review shows the extraordinary potential for using stable isotope systems of H, C, N, O, and Si from the macro- and microscopic remains of a wide range of organisms commonly preserved in lake sediments. The stable isotope composition of these siliceous, carbonate and organic components have been related to a range of ecological and environmental variables and processes including evaporation, climate, nutrient cycling, productivity, and methane cycling. Stable isotope measurements on fossil remains have been used to reconstruct past changes in these variables and processes. Furthermore, new approaches for reconstructing past environmental and ecosystem change (e.g., analysing changes in primary productivity or the structure of palaeo food webs) are continuously being developed, expanding the range of applications for isotope analyses based on macro- and microscopic remains in lake sediments. Our review has provided an overview of the level of understanding of the driving variables for environmental proxies based on isotope measurements. Complicating factors such as the effects of seasonality, transport, sample preparation, offsets between different tissues, and of confounding environmental variables, have also been highlighted. These factors remain a major challenge for emerging but also established isotope based approaches analysing biotic remains in lake sediments. An important aim of the review is therefore to also highlight the importance of calibration studies, be it controlled experiments or in field surveys. Such calibrations are essential to better understand the relationships between stable isotope composition and ecological/environmental variables, and are crucial to interpret variability measured in down core applications in terms of an ecological/environmental ‘signal’ or stochastic ‘noise’. Modelling can be a valuable tool to increase our ability to distinguish likely scenarios from unlikely ones. We have also touched on recent methodological advances that have led to expanding the use of Si isotope measurements on diatoms, or reducing the sample size needed for analysis of C isotopes on organic samples, making it clear that technical developments are necessary to continue to increase the stable isotope toolkit available to palaeolimnological studies.

Author contributions

MvH initiated the workshop on stable isotopes in fossils and organic compounds in lake sediments, coordinated the review and manuscript, and drafted sections 1, 4.5, 4.6, 4.7 and 5.2; ML and VP drafted section 2; JAH drafted section 3; OH drafted section 4.2 and 4.3; MJW drafted section 4.4; CT drafted section 5.1. All co-authors participated in workshop discussion groups, provided input for the manuscript, and suggested improvements.

Acknowledgements

The authors would like to thank the University of Southampton (Department of Geography and Environment and the School of Ocean and Earth Science, National Oceanography Centre Southampton) for hosting the workshop and AOC Labs, Beta Analytic, and Isoprime for their financial support. Quinn Asena, Nadine Dräger, Anne-Lise Jourdan, Maren Pauly, and Kadri Sohar are gratefully acknowledged for their contribution to discussions at the workshop. We thank Marie-Elodie Perga and an anonymous reviewer for their constructive feedback on an earlier version of the manuscript.

References

Abelmann, A., Gersonde, R., Knorr, G., Zhang, X., Chapligin, B., Maier, E., Esper, O., Friedrichsen, H., Lohmann, G., Meyer, H., Tiedemann, R., 2015. The seasonal sea-ice zone in the glacial Southern Ocean as a carbon sink. Nature Communications 6.

Agasild, H., Zingel, P., Tuvikene, L., Tuvikene, A., Timm, H., Feldmann, T., Salujõe, J., Toming, K., Jones, R.I., Nõges, T., 2013. Biogenic methane contributes to the food web of a large, shallow lake. Freshwat. Biol. 59, 272-285.

Aichner, B., Herzschuh, U., Wilkes, H., 2010. Influence of aquatic macrophytes on the stable carbon isotopic signatures of sedimentary organic matter in lakes on the Tibetan Plateau. Org. Geochem. 41, 706–718.

Alleman, L.Y., Cardina, D., Cocquyt, C., Plisnier, P-D., Descy, J-P., Kimirei, I., Sinyinza, D., André, L. 2005. Silicon Isotopic Fractionation in Lake Tanganyika and Its Main Tributaries. Journal of Great Lakes Research 31, 509–519.

Anadón, P., Moscariello, A., Rodriguez-Lazaro, J., Filippi, M.L., 2006. Holocene environmental changes of Lake Geneva (Lac Leman) from stable isotopes ($\delta^{13}C$, $\delta^{18}O$) and trace element records of ostracod and gastropod carbonates. J. Paleolimnol. 36, 117–117.

Andrews J.E., Coletta, P., Pentecost A., Riding R., Dennis S., Dennis P.F., Spiro, B., 2004. Equilibrium and disequilibrium stable isotope effects in modern charophyte calcites: implications for palaeoenvironmental studies. Palaeogeogr. Palaeoclimatol. Palaeoecol. 204, 101–14.

1488 Apolinarska K., Hammarlund D., 2009 Multi-component stable isotope records from Late
1489 Weichselian and early Holocene lake sediments at Imiołki, Poland: palaeoclimatic and
1490 methodological implications. J. Quaternary Sci. 24, 948–959
1491 Apolinarska, K., Peechaty, M., Kossler, A., 2015. Within-sample variability of $\delta^{13}C$ and $\delta^{18}O$
1492 values of freshwater gastropod shells and the optimum number of shells to measure
1493 per sediment layer in the Paddenluch palaeolacustrine sequence, Germany. J.
1494 Paleolimnol. 54, 305–323.
1495 Arppe, L., Kurki, E., Wooller, M.J., Luoto, T.P., Zajączkowski, M., Ojala, A.E.K., 2017. A 5500-
1496 year oxygen isotope record of high arctic environmental change from southern
1497 Spitsbergen. Holocene. doi: 10.1177/0959683617715698
1498 Arthur, K.E., Kelez, S., Larsen, T., Choy, C.A., Popp, B.N., 2014. Tracing the biosynthetic
1499 source of essential amino acids in marine turtles using delta C–13 fingerprints. Ecology,
1500 95(5): 1285–1293.
1501 Bailey, H., Henderson, A.C.G., Sloane, H.J., Snelling, A., Leng, M.J., Kaufman, D.S. 2014. The
1502 effect of species on lacustrine $\delta^{18}O_{diatom}$ and its implication for palaeoenvironmental
1503 reconstructions. J. Quatern. Sci. 29, 393–400.
1504 Bajpai, S., Holmes, J. A., Bennett, C., Mandal, N., Khosla, A., 2013. Palaeoenvironment of
1505 Peninsular India during the late Cretaceous Deccan volcanic episode from trace-
1506 element and stable-isotope geochemistry of Intertrappean ostracod shells. Global.
1507 Planet. Change 107, 82–90.
1508 Barker, P.A., Street-Perrott, F.A., Leng, M.J., Greenwood, P.B., Swain, D.L., Perrott, R.A.,
1509 Telford, J., Ficken, K.J., 2001. A 14 ka oxygen isotope record from diatom silica in two
1510 alpine tarns on Mt Kenya. Science 292, 2307–2310.
1511 Barker, P.A., Hurrell, E.R., Leng, M.J., Plessen, B., Wolff, C., Conley, D.J., Keppens, E., Milne,
1512 I., Cumming, B.F., Laird, K.R., Kendrick, C.P., Wynn, P.M., Verschuren, D., 2013. Carbon
1513 cycling within an East African lake revealed by the carbon isotope composition of
1514 diatom silica: a 25-ka record from Lake Challa, Mt. Kilimanjaro. Quatern. Sci. Rev. 66,
1515 55–63.
1516 Beck, W.C., Grossman, E.L., Morse, J.W., 2005. Experimental studies of oxygen isotope
1517 fractionation in the carbonic acid system at 15 degrees, 25 degrees, and 40 degrees C.
1518 Geochim. Cosmochim. Acta 69, 3493–3503.
1519 Belle, S., Parent, C., Frossard, V., Verneaux, V., Millet, L., Chronopoulou, P.-M., Sabatier, P.,
1520 Magny, M., 2014. Temporal changes in the contribution of methane-oxidizing bacteria
1521 to the biomass of chironomid larvae determined using stable carbon isotopes and
1522 ancient DNA. J. Paleolimnol. 52, 215–228.
1523 Belle, S., Millet, L., Gillet, F., Verneaux, V., Magny, M., 2015a. Assemblages and paleo-diet
1524 variability of subfossil Chironomidae (Diptera) from a deep lake (Lake Grand Maclu,
1525 France). Hydrobiologia 755, 145–160.
1526 Belle, S., Verneaux, V., Millet, L., Parent, C., Magny, M., 2015b. A case study of the past CH_4
1527 cycle in lakes by the combined use of dual isotopes (carbon and hydrogen) and ancient
1528 DNA of methane-oxidizing bacteria: rearing experiment and application to Lake
1529 Remoray (eastern France). Aquat. Ecol. 49, 279–291.
1530 Belle, S., Millet, L., Lami, A., Verneaux, V., Musazzi, S., Hossann, C., Magny, M., 2016a.
1531 Increase in benthic trophic reliance on methane in 14 French lakes during the
1532 Anthropocene. Freshwat. Biol. 61, 1105–1118.
1533 Belle, S., Millet, L., Verneaux, V., Lami, A., David, E., Murgia, L., Parent, C., Musazzi, S.,
1534 Gauthier, E., Bichet, V., 2016b. 20th century human pressures drive reductions in

1535 deepwater oxygen leading to losses of benthic methane-based food webs. Quaternary
1536 Sci. Rev. 137, 209–220.
1537 Belle, S., Verneaux, V., Mariet, A.L., Millet, L., 2017a. Impact of eutrophication on the
1538 carbon stable-isotopic baseline of benthic invertebrates in two deep soft-water lakes.
1539 Freshwat. Biol. 62, 1105–1115.
1540 Belle, S., Luoto, T.P., Kivilä, H.E., Nevalainen, L., 2017b. Chironomid paleo diet as an
1541 indicator of past carbon cycle in boreal lakes: Lake Kylmänlampi (Kainuu province;
1542 Eastern Finland) as a case study. Hydrobiologia 785, 149–158.
1543 Bemis, B.E., Spero, H., Bijma, J., Lea, D.W. 1998. Reevaluation of the oxygen isotopic
1544 composition of planktonic foraminifera: experimental results and revised
1545 paleotemperature equations. Paleoceanography 13: 150–160.
1546 Bennett, C.E., Williams, M., Leng, M.J., Siveter, D.J., Davies, S.J., Sloane, H.J., Wilkinson,
1547 I.P., 2011. Diagenesis of fossil ostracods: implications for stable isotope based
1548 palaeoenvironmental reconstruction. Palaeogeog. Palaeoclimatol. Palaeoecol. 305,
1549 150–161.
1550 Beucher, C.P., Brzezinski, M.A., Crosta, X., 2007. Silicic acid dynamics in the glacial sub-
1551 Antarctic: Implications for the silicic acid leakage hypothesis. Global Biogeochem. Cycles
1552 21.
1553 Binz, P., 1987. Oxygen-isotope analysis on recent and fossil diatoms from Lake Walen and
1554 Lake Zurich (Switzerland) and its application on paleoclimatic studies, PhD Thesis, Swiss
1555 Federal Institute of Technology, Zurich, pp. 165.
1556 Bischoff, J.L., Bullen, T.D., Canavan R.W. IV, Forester, R.M.,1998. A test of uranium series
1557 dating of ostracode shells from the Last Interglaciation at Owens Lake, California, Core
1558 OL-92. In: Bischoff, J.L. (Ed.), The Last Interglaciation at Owens Lake, California: Core OL-
1559 92. U.S. Geological Survey, Open-File Report 98-132, pp.180–186.
1560 Boecklen, W.J., Yarnes, C.T., Cook, B.A., James, A.C., 2011. On the use of stable isotopes in
1561 trophic ecology. Ann. Rev. Ecol. Evol. System. 42, 411–440.
1562 Brandriss, M.E., O'Neil, J. R., Edlund, M.B., Stoermer, E.F., 1998. Oxygen isotope
1563 fractionation between diatomaceous silica and water. Geochim. Cosmochim. Acta 62:
1564 1119–1125.
1565 Brenner, M., Whitmore, T.J., Curtis, J.H., Hodell, D.A., Schelske, C.L. 1999. Stable isotope
1566 (delta C-13 and delta N-15) signatures of sedimented organic matter as indicators of
1567 historic lake trophic state. J. Paleolimnol. 22, 205–221.
1568 Brewer, T.S., Leng, M.J., Mackay, A.W., Lamb, A.L., Tyler, J.J., Marsh, N.G., 2008 Unravelling
1569 contamination signals in biogenic silica oxygen isotope composition: the role of major
1570 and trace element geochemistry. J. Quat. Sci. 23: 321–330.
1571 Bridgwater, N.D., Heaton, T.H.E., O'Hara S.L., 1999. A late Holocene palaeolimnological
1572 record from central Mexico, based on faunal and stable-isotope analysis of ostracod
1573 shells. J. Paleolimnol. 22, 383–97.
1574 Bridoux, M.C., Ingalls, A.E. 2010. Structural identification of long chain polyamines
1575 associated with diatom biosilica in a Southern Ocean sediment core. Geochim.
1576 Cosmochim. Acta 74, 4044.
1577 Brodie, C.R., Leng, M.J., Casford, J.S.L., Kendrick, C.P., Lloyd, J.M., Yongqiang, Z., Bird, M.I.,
1578 2011a. Evidence for bias in C and N concentrations and $\delta^{13}C$ composition of terrestrial
1579 and aquatic organic materials due to pre-analysis acid preparation methods. Chem.
1580 Geol. 282, 67–83.

1581 Brodie, C.R., Heaton, T.H.E., Leng, M.J., Kendrick, C.P., Casford, J.S.L., Lloyd, J.M., 2011b.
1582 Evidence for bias in measured $\delta^{15}N$ values of terrestrial and aquatic organic materials
1583 due to pre-analysis acid treatment methods. Rapid Commun. Mass Sp. 25, 1089–1099.
1584 Bunn, S.E., Boon, P.I., 1993. What sources of organic carbon drive food webs in billabongs?
1585 A study based on stable isotope analysis. Oecologia 96, 85–94.
1586 Cabana, G., Rasmussen, J.B., 1996. Comparison of aquatic food chains using nitrogen
1587 isotopes. Proceedings of the National Academy of Sciences 93, 10844–10847.
1588 Candy, D.J., Kilby, B.A., 1962. Studies on Chitin Synthesis in the Desert Locust. J. Exp. Biol.
1589 39, 129–140.
1590 Carstens, D., Lehmann, M.F., Hofstetter, T.B., Schubert, C.J., 2013. Amino acid nitrogen
1591 isotopic composition patterns in lacustrine sedimenting matter. Geochim. Cosmochim.
1592 Acta 121, 328–338.
1593 Castañeda, I.S., Schouten, S., 2011. A review of molecular organic proxies for examining
1594 modern and ancient lacustrine environments. Quaternary Sci. Rev. 30, 2851–2891.
1595 Chang, J.C., Shulmeister, J., Woodward, C., Michalski, G., 2016. Can stable oxygen and
1596 hydrogen isotopes from Australian subfossil Chironomus head capsules be used as
1597 proxies for past temperature change? J. Paleolimnol. 56, 331–348.
1598 Chang, J.C., Shulmeister, J., Gröcke, D.R., Woodward, C.A., in press. Toward more accurate
1599 temperature reconstructions based on oxygen isotopes of subfossil chironomid head-
1600 capsules in Australia. Limnol. Oceanogr. 63, 295-307.
1601 Chapligin, B., Leng, M.J., Webb, E., Alexandre, A., Dodd, J.P., Ijiri, A., Lucke, A., Shemesh, A.,
1602 Abelmann, A., Herzschuh, U., Longstaffe, F.J., Meyer, H., Moschen, R., Okazaki, Y., Rees,
1603 N.H., Sharp, Z.D., Sloane, H.J., Sonzogni, C., Swann, G.E.A., Sylvestre, F., Tyler, J.J., Yam,
1604 R. 2011. Inter-laboratory comparison of oxygen isotope compositions from biogenic
1605 silica. Geochim. Cosmochim. Acta 75, 7242–7256.
1606 Chen, J.A., Li, J., Tian, S.H., Kalugin, I., Darin, A., Xu, S., 2012. Silicon isotope composition of
1607 diatoms as a paleoenvironmental proxy in Lake Huguangyan, South China. Journal of
1608 Asian Earth Sciences 45, 268–274.
1609 Chivas, A.R., De Deckker, P., Cali, J.A., Chapman, A., Kiss, E., Shelley, J.M.G., 1993. Coupled
1610 stable-isotope and trace-element measurements of lacustrine carbonates as
1611 paleoclimatic indicators. In: Swart, P.K., Lohmann, K.C., McKenzie, J., Savin S. (Eds.),
1612 Climate Change in Continental Isotopic Records. Geophysical Monograph Series 131, pp.
1613 227–247. American Geophysical Union, Washington DC.
1614 Chivas, A.R., De Deckker, P., Wang, S.X., Cali, J.A., 2002. Oxygen-isotope systematics of the
1615 nektic ostracod *Australocypris robusta*. In: Holmes, J.A., Chivas, A.R. (Eds.), The
1616 Ostracoda: Applications in Quaternary Research. Geophysical Monograph Series 131,
1617 301–313. American Geophysical Union, Washington DC.
1618 Cockerton, H.E., Street-Perrott, F.A., Leng, M.J., Barker, P.A., Horstwood, M.S.A., Pashley,
1619 V., 2013. Stable-isotope (H, O, and Si) evidence for seasonal variations in hydrology and
1620 Si cycling from modern waters in the Nile Basin: implications for interpreting the
1621 Quaternary record. Quaternary Science Reviews 66, 4–21.
1622 Cockerton, H.E., Street-Perrott, F.A., Barker, P.A., Leng, M.J., Sloane, H.J., Ficken, K.J., 2015.
1623 Orbital forcing of glacial/interglacial variations in chemical weathering and silicon
1624 cycling within the upper White Nile basin, East Africa: Stable-isotope and biomarker
1625 evidence from Lakes Victoria and Edward. Quaternary Science Reviews 130, 57–71.

1626 Craig, H., Gordon, L.I., 1965. Deuterium and oxygen-18 in the ocean and marine
1627 atmosphere. In: Tongiorgi, E. (Ed.), Stable Isotopes in Oceanographic Studies and
1628 Paleotemperatures, Spoleto, Italy, pp. 9–130.
1629 Dansgaard, W., 1964. Stable isotopes in precipitation. Tellus 16, 436–468.
1630 Darling, G.W., Bath, A.H., Gibson, J.J., Rozanski, K., 2006. Isotopes in water. In: Leng, M.J.
1631 (Ed.), Isotopes in Palaeoenvironmental Research. Springer, The Netherlands, pp. 1–66.
1632 Das, S.K., Routh, J., Roychoudhury, A.N., Klump, J.V., 2008. Elemental (C, N, H and P) and
1633 stable isotope (δ^{15}N and δ^{13}C) signatures in sediments from Zeekoevlei, South Africa: a
1634 record of human intervention in the lake. J. Paleolimnol. 39, 349–360.
1635 Davidson, T.A., Sayer, C.D., Perrow, M.R., Tomlinson, M.L., 2003. Representation of fish
1636 communities by scale sub-fossils in shallow lakes: implications for inferring percid–
1637 cyprinid shifts. J. Paleolimnol. 30, 441-449.
1638 De la Rocha, C.L., Brzezinski, M.A., DeNiro, M.J., 1996. Purification, recovery, and laser-
1639 driven fluorination of silicon from dissolved and particulate silica for the measurement
1640 of natural stable isotope abundances. Anal. Chem. 68, 3746–3750.
1641 De La Rocha, C.L., Brzezinski, M.A., DeNiro, M.J., 1997. Fractionation of silicon isotopes by
1642 marine diatoms during biogenic silica formation. Geochim. Cosmochim. Acta 61, 5051–
1643 5056.
1644 De La Rocha, C.L., Brzezinski, M.A., DeNiro, M.J., Shemesh, A., 1998. Silicon-isotope
1645 composition of diatoms as an indicator of past oceanic change. Nature 395, 680–683.
1646 De la Rocha, C.L., Brzezinski, M.A., DeNiro, M.J., 2000. A first look at the distribution of the
1647 stable isotopes of silicon in natural waters. Geochim. Cosmochim. Acta 64, 2467–2477.
1648 Decrouy, L., 2012. Biological and environmental controls on isotopes in ostracod shells. In:
1649 Horne, D.J., Holmes, J.A., Rodriguez-Lazaro, J., Viehberg, F., (Eds.), Ostracoda as Proxies
1650 for Quaternary Climate Change. Developments in Quaternary Science 17, 165–181.
1651 Decrouy, L., Vennemann, T. W., Ariztegui, D., 2011. Controls on ostracod valve chemistry:
1652 part 2. Carbon and oxygen isotope compositions. Geochim. Cosmochim. Acta 75, 7380–
1653 7399.
1654 Grey, J., Deines, P., 2005. Differential assimilation of methanotrophic and
1655 chemoautotrophic bacteria by lake chironomid larvae Aquat. Microb. Ecol. 40, 61–66.
1656 Deines, P., Grey, J., 2006. Site-specific methane production and subsequent midge
1657 mediation within Esthwaite Water, UK. Arch. Hydrobiol. 167, 317–334.
1658 Deines, P., Wooller, M.J., Grey, J., 2009. Unravelling complexities in benthic food webs
1659 using a dual stable isotope (hydrogen and carbon) approach. Freshwat. Biol. 54, 2243–
1660 2251.
1661 Demarest, M.S., Brzezinski, M.A., Beucher, C.P., 2009. Fractionation of silicon isotopes
1662 during biogenic silica dissolution. Geochim. Cosmochim. Acta 73, 5572–5583.
1663 DeNiro, M.J., Epstein, S., 1978. Influence of diet on the distribution of carbon isotopes in
1664 animals. Geochimica and Cosmochimica Acta 42, 495–506.
1665 DeNiro, M.J., Epstein, S., 1981. Influence of diet on the distribution of nitrogen isotopes in
1666 animals. Geochimica and Cosmochimica Acta 45, 341–351.
1667 Dettman, D.L., Smith, A.J., Rea, D.K., Lohmann, K.C., Moore Jr., T.C., 1995. Glacial melt-
1668 water in Lake Huron during early post-glacial times as inferred from single-valve
1669 analysis of oxygen isotopes in ostracodes. Quat. Res. 43, 297–310.
1670 Dettman, D.L., Reische, A.K., Lohmann, K.C., 1999. Controls on the stable isotope
1671 composition of seasonal growth bands in aragonitic fresh-water bivalves (unionidae).
1672 Geochim. Cosmochim. Acta 63, 1049–1057.

1673 Devlin, S.P., Saarenheimo, J., Syväranta, J., Jones, R.I., 2015. Top consumer abundance
1674 influences lake methane efflux. 6, 8787.
1675 Devriendt, L.S., McGregor, H.V., Chivas, A.R., 2017. Ostracod calcite records the $^{18}O/^{16}O$
1676 ratio of the bicarbonate and carbonate ions in water. Geochim. Cosmochim. Acta 214,
1677 30–50.
1678 Dixit, Y., Hodell, D.A., Sinha, R., Petrie, C.A., 2015. Oxygen isotope analysis of multiple,
1679 single ostracod valves as a proxy for combined variability in seasonal temperature and
1680 lake water oxygen isotopes. J. Paleolimnol. 53, 35–45.
1681 Dodd JP, Sharp ZD (2010) A laser fluorination method for oxygen isotope analysis of
1682 biogenic silica and a new oxygen isotope calibration of modern diatoms in freshwater
1683 environments. Geochim. Cosmochim. Acta 74, 1381–1390.
1684 Doncaster, C.P., Alonso Chávez, V., Viguier, C., Wang, R., Zhang, E., Dong, X., Dearing, J.A.,
1685 Langdon, P.G., Dyke, J.G., 2016. Early warning of critical transitions in biodiversity from
1686 compositional disorder. Ecology 97, 3079–3090.
1687 Drew, S., Flett, I., Wilson, J., Heijnis, H., Skilbeck, C., 2008. The trophic history of Myall
1688 Lakes, New South Wales, Australia: interpretations using $\delta^{13}C$ and $\delta^{15}N$ of the
1689 sedimentary record. Hydrobiologia 608, 35–47.
1690 Duplessy, J.C., Lalou, C., Vinot, A.C., 1970. Differential isotopic fractionation in benthic
1691 foraminifera and paleotemperatures revised. Science 213, 1247–1250.
1692 Durand, A., fontes, J. Ch., Gasse, F., Icole, M., Lang, J., 1984. Le nord-ouest du lac Tchad au
1693 Quatemaire: ewtude de paléoenvironnements alluviaux, éoliens, palustres et lacustres.
1694 Palaeoecol. Africa 16, 215–243.
1695 Durazzi, J.T., 1977. Stable isotopes in the ostracod shell: a preliminary study. Geochim.
1696 Cosmochim. Acta 41, 1168–1170.
1697 Elvert, M., Pohlman, J.W., Becker, K.W., Gaglioti, B., Hinrichs, K.-U., Wooller, M.J., 2016.
1698 Methane turnover and environmental change from Holocene lipid biomarker records in
1699 a thermokarst lake in Arctic Alaska. Holocene 26, 1766-1777.
1700 Engels, S., Helmens, K.F., VÄLiranta, M., Brooks, S.J., Birks, H.J.B., 2010. Early Weichselian
1701 (MIS 5d and 5c) temperatures and environmental changes in northern Fennoscandia as
1702 recorded by chironomids and macroremains at Sokli, northeast Finland. Boreas 39,
1703 689–704.
1704 Escobar, J., Curtis, J.H., Brenner, M., Hodell, D.A., Holmes, J.A., 2010. Isotope
1705 measurements of single ostracod valves and gastropod shells for climate
1706 reconstruction: evaluation of within-sample variability and determination of optimum
1707 sample size. J. Paleolimnol. 43, 921–938.
1708 Feng, X., Lauder, A.M., Posmentier, E.S., Kopec, B.G., Virginia, R.A., 2016. Evaporation and
1709 transport of water isotopologues from Greenland lakes: the lake size effect. Quat. Sci.
1710 Rev. 131, 302e315.
1711 Fernandes, R., Millard, A.R., Brabec, M., Nadeau, M.J., Grootes, P., 2014. Food
1712 reconstruction using isotopic transferred signals (FRUITS): a Bayesian model for diet
1713 reconstruction *PLoS ONE* 9: e87436.
1714 Ficken, K.J., Li, B., Swain, D.L., Eglinton, G. 2000. An n-alkane proxy for the sedimentary
1715 inputs of submerged/floating freshwater aquatic macrophytes. Org. Geochem. 31, 745–
1716 749.
1717 Frey, D.G., 1964. Remains of animals in Quaternary lake and bog sediments and their
1718 interpretation. Advances in Limnology 2, 1–114.

1719 Frings PJ, Clymans W, Jeppesen E, Lauridsen TL, Struyf E, Conley DJ. Lack of steady-state in
1720 the global biogeochemical Si cycle: Emerging evidence from lake Si sequestration.
1721 Biogeochemistry. 2014;117(2–3):255–77.
1722 Frings, P.J., Clymans, W., Fontorbe, G., De La Rocha, C.L., Conley, D.J., 2016. The
1723 continental Si cycle and its impact on the ocean Si isotope budget. Chem. Geol. 425, 12–
1724 36.
1725 Fripiat, F., Cavagna, A.J., Dehairs, F., Speich, S., Andre, L., Cardinal, D., 2011. Silicon pool
1726 dynamics and biogenic silica export in the Southern Ocean inferred from Si-isotopes.
1727 Ocean Sci. 7, 533–547.
1728 Frossard, V., Belle, S., Verneaux, V., Millet, L., Magny, M., 2013. A study of the δ13C offset
1729 between chironomid larvae and their exuvial head capsules: implications for
1730 palaeoecology. J. Paleolimnol. 50, 379–386.
1731 Frossard, V., Verneaux, V., Millet, L., Jenny, J.-P., Arnaud, F., Magny, M., Perga, M.-E., 2014.
1732 Reconstructing long-term changes (150 years) in the carbon cycle of a clear-water lake
1733 based on the stable carbon isotope composition (δ13C) of chironomid and cladoceran
1734 subfossil remains. Freshwat. Biol. 59, 789–802.
1735 Frossard, V., Verneaux, V., Millet, L., Magny, M., Perga, M.-E., 2015. Changes in carbon
1736 sources fueling benthic secondary production over depth and time: coupling
1737 Chironomidae stable carbon isotopes to larval abundance. Oecologia 178, 603–614.
1738 Gat, J.R., 1996. Oxygen and hydrogen isotopes in the hydrologic cycle. Annual Review of
1739 Earth and Planetary Sciences 24, 225–262.
1740 Gebruk AV, Kuznetsov AI, Namsaraev BB, YM, M., 1993. The role of bacterial organic
1741 matter in the nutrition of deep-water benthic fauna in Frolikha Bay (Lake Baikal) under
1742 conditions of increased thermal flow (in Russian). Izvestiya Akademii Nauk: Seriya
1743 Biologicheskaya 6, 903–908.
1744 Georg, R.B., Reynolds, B.C., Frank, M., Halliday, A.N., 2006. New sample preparation
1745 techniques for the determination of Si isotopic compositions using MC-ICPMS. Chem.
1746 Geol. 235, 95–104.
1747 Gerdeaux, D., Perga, M.-E., 2006. Changes in Whitefish Scales δ^{13}C during Eutrophication
1748 and Reoligotrophication of Subalpine Lakes. Limnol. Oceanogr. 51, 772–780.
1749 Gibson, J.J., Birks, S.J., Yi, Y., 2016. Stable isotope mass balance of lakes: a contemporary
1750 perspective. Quaternary Sci. Rev. 131, 316–328.
1751 Giddings, J.C., 1985. A system based on split-flow lateral transport thin (SPLITT) separation
1752 cells for rapid and continuous particle fractionation. Separation Science Technology 20,
1753 749–768.
1754 Goedkoop, W., Akerblom, N., Demandt, M.H., 2006. Trophic fractionation of carbon and
1755 nitrogen stable isotopes in Chironomus riparius reared on food of aquatic and
1756 terrestrial origin. Freshwat. Biol. 51, 878–886.
1757 Graham, R.W., Belmecheri, S., Choy, K., Culleton, B.J., Davies, L.J., Froese, D., Heintzman,
1758 P.D., Hritz, C., Kapp, J.D., Newsom, L.A., Rawcliffe, R., Saulnier-Talbot, É., Shapiro, B.,
1759 Wang, Y., Williams, J.W., Wooller, M.J., 2016. Timing and causes of mid-Holocene
1760 mammoth extinction on St. Paul Island, Alaska. Proceedings of the National Academy of
1761 Sciences 113, 9310–9314.
1762 Grasse, P., Brzezinski, M.A., Cardinal, D., de Souza, G.F., Andersson, P., Closset, I., Cao, Z.,
1763 Dai, M., Ehlert, C., Estrade, N., François, R., Frank, M., Jiang, G., Jones, J.L., Kooijman, E.,
1764 Liu, Q., Lu, D., Pahnke, K., Ponzevera, E., Schmitt, M., Sun, X., Sutton, G.N., Thil, F., Weis,

1765 D., 2017. GEOTRACES inter-calibration of the stable silicon isotope composition of
1766 dissolved silicic acid in seawater. JAAS 32, 562 – 578.
1767 Greenaway, P., 1985. Calcium balance and moulting in the Crustacea. Biological Reviews
1768 60, 425–454.
1769 Grey, J., 2016. The Incredible Lightness of Being Methane-Fuelled: Stable Isotopes Reveal
1770 Alternative Energy Pathways in Aquatic Ecosystems and Beyond. Frontiers in Ecology
1771 and Evolution 4, doi: 10.3389/fevo.2016.00008.
1772 Grey, J., Deines, P., 2005. Differential assimilation of methanotrophic and
1773 chemoautotrophic bacteria by lake chironomid larvae Aquat. Microb. Ecol. 40, 61–66.
1774 Grey, J., Kelly, A., Ward, S., Sommerwerk, N., Jones, R.I., 2004. Seasonal changes in the
1775 stable isotope values of lake-dwelling chironomid larvae in relation to feeding and life
1776 cycle variability. Freshwat. Biol. 49, 681–689.
1777 Griffiths, K., Michelutti, N., Blais, J., Kimpe, L., Smol, J., 2010. Comparing nitrogen isotopic
1778 signals between bulk sediments and invertebrate remains in High Arctic seabird-
1779 influenced ponds. J. Paleolimnol. 44, 405–412.
1780 Gu, B., Chapman, A.D., Schelske, C.L., 2006. Factors Controlling Seasonal Variations in
1781 Stable Isotope Composition of Particulate Organic Matter in a Soft Water Eutrophic
1782 Lake. Limnol. Oceanogr. 51, 2837–2848.
1783 Häberle, S., Fuller, B.T., Nehlich, O., Van Neer, W., Schibler, J., Plogmann, H.H., 2016. Inter-
1784 and intraspecies variability in stable isotope ratio values of archaeological freshwater
1785 fish remains from Switzerland (11th–19th centuries AD). Environmental Archaeology
1786 21, 119–132.
1787 Hanisch, J.R., Tonn, W.M., Paszkowski, C.A., Scrimgeour, G.J., 2010. δ^{13}C and δ^{15}N
1788 Signatures in Muscle and Fin Tissues: Nonlethal Sampling Methods for Stable Isotope
1789 Analysis of Salmonids. N. Am. J. Fish. Manage. 30, 1–11.
1790 Heaton T.H.E., Holmes J.A., Bridgwater N.D., 1995. Carbon and oxygen isotope variations
1791 among lacustrine ostracods: Implications for palaeoclimatic studies. The Holocene 5,
1792 428–434.
1793 Hecky, R.E., Mopper, K., Kilham, P., Degens, E.T. 1973. The amino acid and sugar
1794 composition of diatom cell walls. Marine Biology 19, 323.
1795 Heiri, O., Schilder, J., van Hardenbroek, M., 2012. Stable isotopic analysis of fossil
1796 chironomids as an approach to environmental reconstruction: State of development
1797 and future challenges. Proceedings of the 18th International Symposium on
1798 Chironomidae, Trondheim 4–6. August 2011, Fauna Norvegica 31.
1799 Hendry, K.R., Brzezinski, M.A., 2014. Using silicon isotopes to understand the role of the
1800 Southern Ocean in modern and ancient biogeochemistry and climate. Quatern. Sci. Rev.
1801 89, 13–26.
1802 Hendry, K.R., Gong, X., Knorr, G., Pike, J., Hall, I.R., 2016. Deglacial diatom production in
1803 the tropical North Atlantic driven by enhanced silicic acid supply. Earth Planet. Sci. Lett.
1804 438, 122–129.
1805 Hernández, A., Bao, R., Giralt, S., Leng, M.J., Barker, P.A., Sáez, A., Pueyo, J.J., Moreno, A.,
1806 Valero-Garcés, B.L., Sloane, H.J. 2008. The palaeohydrological evolution of Lago
1807 Chungará (Andean Altiplano, northern Chile) during the Lateglacial and early Holocene
1808 using oxygen isotopes in diatom silica. J. Quat. Sci. 23, 351–363.
1809 Hernández, A., Bao, R., Giralt, S., Barker, P.A., Leng, M.J., Sloane, H.J., Saez, A., 2011.
1810 Biogeochemical processes controlling oxygen and carbon isotopes of diatom silica in

1811 Late Glacial to Holocene lacustrine rhythmites. Palaeogeogr. Palaeoclimat. Palaeoecol. 1812 299, 413–425.
1813 Hernández, A., Bao, R., Giralt, S., Saez, A., Leng, M.J., Barker, P.A., Kendrick, C.P., Sloane, 1814 H.J., 2013. Climate, catchment runoff and limnological drivers of carbon and oxygen 1815 isotope composition of diatom frustules from the central Andean Altiplano during the 1816 Lateglacial and Early Holocene. Quatern. Sci. Rev. 66, 64–73.
1817 Hershey, A.E., Northington, R.M., Hart-Smith, J., Bostick, M., Whalen, S.C., 2015. Methane 1818 efflux and oxidation, and use of methane-derived carbon by larval Chironomini, in arctic 1819 lake sediments. Limnol. Oceanogr. 60, 276–285.
1820 Herzschuh, U., Mischke, S., Meyer, H., Plessen, B., Zhang, C., 2010a. Using variations in the 1821 stable carbon isotope composition of macrophyte remains to quantify nutrient 1822 dynamics in lakes. J. Paleolimnol. 43, 739–750.
1823 Herzschuh, U., Mischke, S., Meyer, H., Plessen, B., Zhang, C., 2010b. Lake nutrient 1824 variability inferred from elemental (C, N, S) and isotopic (δ13C, δ15N) analyses of 1825 aquatic plant macrofossils. Quaternary Sci. Rev. 29, 2161–2172.
1826 Heyng, A.M., Mayr, C., Lücke, A., Striewski, B., Wastegård, S., Wissel, H., 2012. 1827 Environmental changes in northern New Zealand since the Middle Holocene inferred 1828 from stable isotope records (δ^{13}C, δ^{15}N of Lake Pupuke. J. Paleolimnol. 48, 351–366.
1829 Hodell, D.A., Curtis, J.H., Jones, G.A., Higuera-Grundy, A., Brenner, M., Binford, M.W., 1830 Dorsey, K.T., 1991. Reconstruction of Caribbean climate change over the past 10,500 1831 years. Nature 352, 790–793.
1832 Hodell, D.A., Curtis, J.H., Brenner, M., 1995. Possible role of climate in the collapse of 1833 Classic Maya civilization. Nature 375, 391–394.
1834 Høie, H., Folkvord, A., Otterlei, E., 2003. Effect of somatic and otolith growth rate on stable 1835 isotopic composition of early juvenile cod (Gadus morhua L) otoliths. J. Exp. Mar. Biol. 1836 Ecol. 289, 41–58.
1837 Hobson, K.A., Atwell, L., Wassenaar, L.I., 1999. Influence of drinking water and diet on the 1838 stable-hydrogen isotope ratios of animal tissues. Proceedings of the National Academy 1839 of Sciences of the United States of America 96, 8003–8006.
1840 Hollander, D.J., McKenzie, J.A., 1991. CO_2 control on carbon-isotope fractionation during 1841 aqueous photosynthesis: A paleo-pCO_2 barometer. Geology 19, 929–932.
1842 Hollander, D.J., Smith, M.A., 2001. Microbially mediated carbon cycling as a control on the 1843 δ^{13}C of sedimentary carbon in eutrophic Lake Mendota (USA): new models for 1844 interpreting isotopic excursions in the sedimentary record. Geochim. Cosmochim. Acta 1845 65, 4321–4337.
1846 Holmes, J.A., 1992. Nonmarine ostracods as Quaternary palaeoenvironmental indicators. 1847 Progr. Phys.Geog. 405–431.
1848 Holmes, J.A., 1996. Trace-element and stable-isotope geochemistry of non-marine 1849 ostracod shells in Quaternary palaeoenvironmental reconstruction. J. Paleolimnol. 15, 1850 223–235.
1851 Holmes, J.A., Chivas, A.R., 2002. Ostracod shell chemistry - overview. In: Holmes, J.A., 1852 Chivas, A.R. (Eds.), The Ostracoda: Applications in Quaternary Research. Geophysical 1853 Monograph Series 131, 183–204. American Geophysical Union, Washington DC.
1854 Holmes ,J.A., Darbyshire, D.P.F., Heaton, T.H.E., 2007. Palaeohydrological significance of 1855 late Quaternary strontium isotope ratios in a tropical lake. Chem.Geol. 236, 281–290.
1856 Holmes, J.A., Arrowsmith, C., Austin, W.E.N., Fisher, E., Holme, R., Marshall, J.D., Oldfield, 1857 F., van der Post, K. 2010. Climate and atmospheric circulation changes over the past

1858 1000 years reconstructed from oxygen isotopes in lake-sediment carbonate from
1859 Ireland. The Holocene 20, 1105–1111.
1860 Holmes, J.A., Tindall, J., Roberts, N., Marshall, W., Marshall, J.D., Bingham, A., Feeser, I.,
1861 O'Connell, M., Atkinson, T., Jourdan, A-L., March, A., Fisher, E.H., 2016. Lake isotope
1862 records of the 8200-year cooling event in western Ireland: Comparison with model
1863 simulations. Quat. Sci. Rev. 131, 341–349.
1864 Horn, M.G., Beucher, C.P., Robinson, R.S., Brzezinski, M.A., 2011. Southern ocean nitrogen
1865 and silicon dynamics during the last deglaciation. Earth. Planet. Sci. Lett. 310, 334–339.
1866 Horton, T.W., Defliese, W.F., Tripati, A.K., Oze, C., 2016. Evaporation induced ^{18}O and ^{13}C
1867 enrichment in lake systems: A global perspective on hydrologic balance effects. Quat.
1868 Sci. Rev. 131, 365–379.
1869 Huang, Y., Street-Perrott, F.A., Perrott, R.A., Metzger, P., Eglinton, G., 1999. Glacial-
1870 interglacial environmental changes inferred from molecular and compound-specific
1871 δ^{13}C analyses of sediments from Sacred Lake, Mt. Kenya. Geochim. Cosmochim Acta 63,
1872 1383–1404.
1873 Hurrell, E.R., Barker, P.A., Leng, M.J., Vane, C.H., Wynn, P., Kendrick, C.P., Verschuren, D.,
1874 Street-Perrott, F.A. 2011. Developing a methodology for carbon isotope analysis of
1875 lacustrine diatoms. Rapid Commun. Mass Sp. 25, 1567–1574.
1876 Iglesias, C., Meerhoff, M., Johansson, L.S., González-Bergonzoni, I., Mazzeo, N., Pacheco,
1877 J.P., Mello, F.T.-d., Goyenola, G., Lauridsen, T.L., Søndergaard, M., Davidson, T.A.,
1878 Jeppesen, E., 2017. Stable isotope analysis confirms substantial differences between
1879 subtropical and temperate shallow lake food webs. Hydrobiologia 784, 111–123.
1880 Janz, H., Vennemann, T.W., 2005. Isotopic composition (O, C, Sr, and Nd) and trace
1881 element ratios (Sr/Ca, Mg/Ca) of Miocene marine and brackish ostracods from North
1882 Alpine Foreland deposits (Germany and Austria) as indicators for palaeoclimate.
1883 Palaeogeogr., Palaeoclimatol., Palaeoecol. 225, 216–247.
1884 Jex, C.N., Pate, G.H., Blyth, A.J., Spencer, R.G.M., Hernes, P.J., Khan, S.J., Baker, A., 2014.
1885 Lignin biogeochemistry: from modern processes to Quaternary archives. Quaternary
1886 Sci. Rev. 87, 46–59.
1887 Jeziorski, A., Yan, N.D., 2006. Species identity and aqueous calcium concentrations as
1888 determinants of calcium concentrations of freshwater crustacean zooplankton. Can. J.
1889 Fish. Aquat. Sci. 63, 1007–1013.
1890 Jeziorski, A., Paterson, A.M., Yan, N.D., Smol, J.P., 2008. Calcium levels in *Daphnia* ephippia
1891 cannot provide a useful paleolimnological indicator of historical lakewater Ca
1892 concentrations. J. Paleolimnol. 39, 421–425.
1893 Jones, M.D., Dee, S., Anderson, L., Baker, A., Bowen, G., Noone, D.C., 2016a. Water isotope
1894 systematics: Improving our palaeoclimate interpretations. Quaternary Sci. Rev. 131,
1895 243–249.
1896 Jones, M.D., Cuthbert, M.O., Leng, M.J., McGowan, S., Mariethoz, G., Arrowsmith, C.,
1897 Sloane, H.J., Humphrey, K.K., Cross, I., 2016b. Comparisons of observed and modelled
1898 lake δ^{18}O variability. Quaternary Sci. Rev. 131, 329–340.
1899 Jones, R.I., Grey, J., 2011. Biogenic methane in freshwater food webs. Freshwat. Biol. 56,
1900 213–229.
1901 Jones, R.I., Carter, C.E., Kelly, A., Ward, S., Kelly, D.J., Grey, J., 2008. Widespread
1902 contribution of methane-cycle bacteria to the diets of lake profundal chironomid larvae.
1903 Ecology 89, 857–864.

1904 Juillet-Leclerc, A., Labeyrie, L., 1987. Temperature dependence of the oxygen isotopic
1905 fractionation between diatom silica and water. Earth Planet. Sci. Lett. 84: 69–74.
1906 Junker, J.R., Cross, W.F., 2014. Seasonality in the trophic basis of a temperate stream
1907 invertebrate assemblage: Importance of temperature and food quality. Limnol.
1908 Oceanogr. 59, 507–518.
1909 Kaminski, M., 1984. Food composition of three bryozoan species (Bryozoa,
1910 Phylactolaemata) in a mesotrophic lake. Polish Archive of Hydrobiology 31, 45–53.
1911 Kamjunke, N., Benndorf, A., Wilbert, C., Opitz, M., Kranich, J., Bollenbach, M., Benndorf, J.,
1912 1999. Bacteria ingestion by Daphnia galeata in a biomanipulated reservoir: a
1913 mechanism stabilizing biomanipulation? Hydrobiologia 403, 109–121.
1914 Kankaala, P., Taipale, S., Li, L., Jones, R., 2010. Diets of crustacean zooplankton, inferred
1915 from stable carbon and nitrogen isotope analyses, in lakes with varying allochthonous
1916 dissolved organic carbon content. Aquat. Ecol. 44, 781–795.
1917 Karlsson, J., Berggren, M., Ask, J., Byström, P., Jonsson, A., Laudon, H., Jansson, M., 2012.
1918 Terrestrial organic matter support of lake food webs: Evidence from lake metabolism
1919 and stable hydrogen isotopes of consumers. Limnol. Oceanogr. 57, 1042–1048.
1920 Kattel, G., Gell, P., Perga, M.-E., Jeppesen, E., Grundell, R., Weller, S., Zawadzki, A., Barry,
1921 L., 2015. Tracking a century of change in trophic structure and dynamics in a floodplain
1922 wetland: integrating palaeoecological and palaeoisotopic evidence. Freshwat. Biol. 60,
1923 711–723.
1924 Kawabe, I.1978. Calculation of oxygen isotope fractionation in quartz-water system with
1925 special reference to the low temperature fractionation. Geochim. Cosmochim. Acta 42,
1926 613–621.
1927 Keatings, K.W., 1999. The basis for ostracod shell chemistry in palaeoclimate
1928 reconstruction. Unpublished Ph. D. thesis, Kingston University, UK.
1929 Keatings, K.W., Heaton, T.H.E., Holmes, J.A., 2002a. Carbon and oxygen isotope
1930 fractionation in non-marine ostracods: Results from a 'natural culture' environment.
1931 Geochim. Cosmochim. Acta 66, 1701–1711.
1932 Keatings, K.W., Heaton, T.H.E., Holmes, J.A., 2002b. The effects of diagenesis on the trace
1933 element and stable isotope geochemistry of non-marine ostracod valves. J. Paleolimnol.
1934 28, 245–252.
1935 Keatings, K.W., Holmes, J.A., Heaton, T.H.E., 2006. Effects of pre-treatment on ostracod
1936 valve chemistry. Chem. Geol. 235, 250–261.
1937 Kelly, M.H., Hagar, W.G., Jardine, T.D., Cunjak, R.A., 2006. Nonlethal Sampling of Sunfish
1938 and Slimy Sculpin for Stable Isotope Analysis: How Scale and Fin Tissue Compare with
1939 Muscle Tissue. N. Am. J. Fish. Manage. 26, 921–925.
1940 Kelts, K., Talbot, M. 1990. Lacustrine carbonates as geochemical archives of environmental
1941 change and biotic/abiotic interactions. In: Tilzer, M.M., Serruya, C. (Eds.), Large Lakes:
1942 Ecology, Structure and Function. Science and Technology Publishers, Madison, pp. 288–
1943 315.
1944 Kim, S.T., O’Neil, J.R., 1997 Equilibrium and nonequilibrium oxygen isotope effects in
1945 synthetic carbonates Geochim. Cosmochim. Acta, 61, 3461–3475.
1946 Kim, S.T., O'Neil, J.R., Hillaire-Marcel, C., Mucci, A., 2007. Oxygen isotope fractionation
1947 between synthetic aragonite and water: influence of temperature and Mg^{2+}
1948 concentration. Geochim. Cosmochim. Acta 71, 4704–4715.
1949 Knauth, L.P., Epstein, S.1982. The nature of water in hydrous silica. American Mineralogy
1950 67, 510–520.

1951 Korniłłowicz-Kowalska, T., Bohacz, J., 2011. Biodegradation of keratin waste: Theory and
1952 practical aspects. Waste Manage. 31, 1689–1701.
1953 Kroger, N., Poulsen, N. 2008. Diatoms-from cell wall biogenesis to nanotechnology. Annual
1954 Review of Genetics 42, 83–107.
1955 Labeyrie, L.D., 1974. New approach to surface seawater paleotemperatures using
1956 (18)O/(16)O ratios in silica of diatom frustules. Nature 248, 40–42.
1957 Labeyrie, L.D., Juillet, A., 1982. Oxygen isotopic exchangeability of diatom valve silica;
1958 interpretation and consequences for palaeoclimatic studies. Geochim. Cosmochim. Acta
1959 46, 967–975.
1960 Lamb, A.L., Leng, M.J., Sloane, H.J., Telford, R.J. 2005. A comparison of $\delta^{18}O$ data from
1961 calcite and diatom silica from early Holocene in a small crater lake in the tropics.
1962 Palaeogeog. Palaeoclimat. Palaeoecol. 223, 290–30.
1963 Lane, C.S., Cullen, V.L., White, D., Bramham-Law, C.W.F., Smith, V.C., 2014. Cryptotephra
1964 as a dating and correlation tool in archaeology. J. Archaeol. Sci. 42, 42–50.
1965 Larsen, T., Taylor, D.L., Leigh, M.B., O'Brien, D.M., 2009. Stable isotope fingerprinting: a
1966 novel method for identifying plant, fungal, or bacterial origins of amino acids. Ecology,
1967 90(12): 3526–3535.
1968 Larsen, T., Ventura, M., Andersen, N., O'Brien, D.M., Piatkowski, U., McCarthy, M.D., 2013.
1969 Tracing Carbon Sources through Aquatic and Terrestrial Food Webs Using Amino Acid
1970 Stable Isotope Fingerprinting. PLOS ONE 8, e73441.
1971 Lasher, G.E., Axford, Y., McFarlin, J.M., Kelly, M.A., Osterberg, E.C., Berkelhammer, M.B.,
1972 2017. Holocene temperatures and isotopes of precipitation in Northwest Greenland
1973 recorded in lacustrine organic materials. Quaternary Sci. Rev. 170, 45–55.
1974 Laws, E.A., Popp, B.N., Bidigare, R.R., Kennicutt, M.C., Macko, S.A., 1995. Dependence of
1975 phytoplankton carbon isotopic composition on growth rate and $[CO_2]_{aq}$: Theoretical
1976 considerations and experimental results. Geochim. Cosmochim. Acta 59, 1131–1138.
1977 Layman, C.A., Araujo, M.S., Boucek, R., Hammerschlag-Peyer, C.M., Harrison, E., Jud, Z.R.,
1978 Matich, P., Rosenblatt, A.E., Vaudo, J.J., Yeager, L.A., Post, D.M., Bearhop, S., 2012.
1979 Applying stable isotopes to examine food-web structure: an overview of analytical
1980 tools. Biological Reviews 87, 545–562.
1981 Leavitt, P.R., Brock, C.S., Ebel, C., Patoine, A., 2006. Landscape-scale effects of urban
1982 nitrogen on a chain of freshwater lakes in central North America. Limnol. Oceanogr. 51,
1983 2262–2277.
1984 Leng, M.J., Barker, P., Greenwood, P, Roberts, N., Reed, J.M. 2001. Oxygen isotope analysis
1985 of diatom silica and authigenic calcite from Lake Pinarbasi, Turkey. Journal of
1986 Paleolimnology, 25, 343–349.
1987 Leng, M.J., Marshall, J.D., 2004. Palaeoclimate interpretation of stable isotope data from
1988 lake sediment archives. Quat. Sci.Rev. 23, 811–831.
1989 Leng, M. J., Barker, P.A., 2006. A review of the oxygen isotope composition of lacustrine
1990 diatom silica for palaeoclimate reconstruction. Earth Sci. Rev. 75, 5–27.
1991 Leng, M.J., Sloane, H.J., 2008. Combined oxygen and silicon isotope analysis of biogenic
1992 silica. J. Quat. Sci. 23, 313–319.
1993 Leng, M.J., Swann, G.E.A. 2010. Stable Isotopes in Diatom Silica. In: J.P. Smol and E.F.
1994 Stoermer. The Diatoms: applications for the Environmental and Earth Sciences,
1995 Cambridge Press, pp 667.
1996 Leng, M.J., Henderson, A.C.G., 2013. Recent advances in isotopes as palaeolimnological
1997 proxies. J. Paleolimnol. 49, 481–496.

1998 Leng, M.J., Lewis, J.P., 2016. Oxygen isotopes in Molluscan shell: applications in
1999 environmental archaeology. Env. Archaeology 21, 295–306.
2000 Leng, M.J., Lamb, A.L., Lamb, H.F., Telford, R.J. 1999. Palaeoclimatic implications of isotopic
2001 data from modern and early Holocene shells of the freshwater snail *Melanoides*
2002 *tuberculata*, from lakes in the Ethiopian Rift Valley. J. Paleolimnol. 21, 97–106.
2003 Leng, M.J., Swann, G.E.A., Hodson, M.J., Tyler, J.J., Patwardhan, S.V., Sloane, H.J., 2009.
2004 The potential use of silicon isotope composition of biogenic silica as a proxy for
2005 environmental change. Silicon 1, 65–77.
2006 Leschine, S.B., 1995. Cellulose Degradation in Anaerobic Environments. Annu. Rev.
2007 Microbiol. 49, 399–426.
2008 Li, X., Liu, W., 2014. Water salinity and productivity recorded by ostracod assemblages and
2009 their carbon isotopes since the early Holocene at Lake Qinghai on the northeastern
2010 Qinghai–Tibet Plateau, China. Palaeogeogr., Palaeoclimatol., Palaeoecol. 407, 25–33
2011 Lombino, A., 2014. The systematics of oxygen isotopes in chironomids (Insecta: Diptera): a
2012 tool for reconstructing past climate. PhD Thesis, Department of Geography, University
2013 College London.
2014 Luoto, T.P., Ojala, A.E.K., Arppe, L., Brooks, S.J., Kurki, E., Oksman, M., Wooller, M.J.,
2015 Zajączkowski, M., 2018. Synchronized proxy-based temperature reconstructions reveal
2016 mid- to late Holocene climate oscillations in High Arctic Svalbard. J. Quaternary Sci. 33,
2017 93–99.
2018 Mackay, A.W., Swann, G.E.A., Brewer, T.S., Leng, M.J., Morley, D.W., Piotrowska, N.,
2019 Rioual, P., White, D. 2011. A reassessment of late glacial - Holocene diatom oxygen
2020 isotope record from Lake Baikal using a geochemical mass-balance approach. J. Quat.
2021 Sci. 26, 627–634.
2022 Magozzi, S., Yool, A., Vander Zanden, H.B., Wunder, M.B., Trueman, C.N., 2017. Using
2023 ocean models to predict spatial and temporal variation in marine carbon isotopes.
2024 Ecosphere 8, e01763.
2025 Maier, E., Chapligin, B., Abelmann, A., Gersonde, R., Esper, O., Ren, J., Friedrichsen, H.,
2026 Meyer, H., Tiedemann, R., 2013. Combined oxygen and silicon isotope analysis of
2027 diatom silica from a deglacial subarctic Pacific record. J Quat. Sci. 28, 571–581.
2028 Mariash, H.L., Cazzanelli, M., Rautio, M., Hamerlik, L., Wooller, M.J., Christoffersen, K.S., in
2029 press. Changes in food web dynamics of low arctic ponds with varying content of
2030 dissolved organic carbon. Arct. Antarct. Alp. Res.
2031 Matheney, R.K., Knauth, L.P., 1989. Oxygen-isotope fractionation between marine biogenic
2032 silica and seawater. Geochim. Cosmochim. Acta 53, 3207–3214.
2033 Mayr, C., Laprida, C., Lücke, A., Martín, R.S., Massaferro, J., Ramón-Mercau, J., Wissel, H.,
2034 2015. Oxygen isotope ratios of chironomids, aquatic macrophytes and ostracods for
2035 lake-water isotopic reconstructions – Results of a calibration study in Patagonia. J.
2036 Hydrol. 529, 600–607.
2037 McCutchan Jr, J.H., Lewis Jr, W.M., Kendall, C., McGrath, C.C., 2003. Variation in trophic
2038 shift for stable isotope ratios of carbon, nitrogen, and sulphur. Oikos 102, 378–390.
2039 Menicucci, A.J., Spero, H.J., Matthews, J., Parikh, S.J. 2017. Influence of exchangeable
2040 oxygen on biogenic silica oxygen isotope data. Chem. Geol. 466, 710–721.
2041 Mering, J.A., 2015. New constraints on water temperature at Lake Bonneville from
2042 carbonate clumped isotopes. MSc thesis, Geochemistry, University of California Los
2043 Angeles.

2044 Merritt, R.W., Cummins, K.W., Berg, M.B., 2008. An introduction to the Aquatic Insects of
2045 North America, 4th ed. Kendall and Hunt, Dubuque, p. 1158.
2046 Meyers, P.A., Ishiwatari, R., 1993. Lacustrine organic geochemistry-an overview of
2047 indicators of organic matter sources and diagenesis in lake sediments. Org. Geochem.
2048 20, 867–900.
2049 Meyers, P.A., Teranes, J.L., 2001. Sediment Organic Matter, in: Last, W.M., Smol, J.P. (Eds.),
2050 Tracking environmental change using lake sediments. Volume 2: Physical and
2051 Geochemical Techniques. Kluwer Academic Publishers, Dordrecht, pp. 239–269.
2052 Middelburg, J.J., 2014. Stable isotopes dissect aquatic food webs from the top to the
2053 bottom. Biogeosciences 11, 2357-2371.
2054 Mikkelsen N., Labeyrie L., Berger W.H., 1978. Silica oxygen isotopes in diatoms: A 20,000 yr
2055 record in deep-sea sediments. Nature 271, 536–538.
2056 Miller, R.F., 1991. Chitin Paleoecology. Biochem. Syst. Ecol. 19, 401–411.
2057 Milligan, A.J., Varela, D.E., Brzezinski, M.A., Morel, F.O.M.M., 2004. Dynamics of silicon
2058 metabolism and silicon isotopic discrimination in a marine diatom as a function of pCO_2.
2059 Limnol. Oceanogr. 49, 322–329.
2060 Mopper K., Garlick G.D., 1971. Oxygen isotope fractionation between biogenic silica and
2061 ocean water. Geochimica et Cosmochimica Acta 35, 1185–1187.
2062 Morley, D.W., Leng, M.J., Mackay, A.W., Sloane, H.J., Rioual, P., Battarbee, R.W., 2004.
2063 Cleaning of lake sediment samples for diatom oxygen isotope analysis. J. Paleolimnol.
2064 31, 391–401.
2065 Morlock, M.A., Schilder, J., van Hardenbroek, M., Szidat, S., Wooller, M.J., Heiri, O., 2017.
2066 Seasonality of cladoceran and bryozoan resting stage δ13C values and implications for
2067 their use as palaeolimnological indicators of lacustrine carbon cycle dynamics. J.
2068 Paleolimnol. 57, 141–156.
2069 Moschen, R., Lücke, A., Schleser, G., 2005. Sensitivity of biogenic silica oxygen isotopes to
2070 changes in surface water temperature and palaeoclimatology. Geophys. Res. Lett. 32:
2071 L07708, doi:10.1029/2004GL022167.
2072 Moschen, R., Lücke, A. , Parplies, J., Radtke, U., Schleser, G.H., 2006. Transfer and early
2073 diagenesis of biogenic silica oxygen isotope signals during settling and sedimentation of
2074 diatoms in a temperate freshwater lake (Lake Holzmaar, Germany). Geochim.
2075 Cosmochim. Acta 70, 4367–4379.
2076 Nation, J.L., 2002. Integument, in: Nation, J.L. (Ed.), Insect physiology and biochemistry.
2077 CRC Press, Boca Raton, p. 485.
2078 Nielson, K.E., Bowen, G.J., 2010. Hydrogen and oxygen in brine shrimp chitin reflect
2079 environmental water and dietary isotopic composition. Geochim. Cosmochim. Acta 74,
2080 1812–1822.
2081 Oehlerich, M., Mayr, C., Gussone, N., Hahn, A., Hölzl, S., Lücke, A., Ohlendorf, C., Rummel,
2082 S., Teichert, B.M.A., Zolitschka, B., 2015. Lateglacial and Holocene climatic changes in
2083 south-eastern Patagonia inferred from carbonate isotope records of Laguna Potrok Aike
2084 (Argentina). Quaternary Sci. Rev. 114, 189–202.
2085 Opfergelt, S., Delmelle, P. 2012. Silicon isotopes and continental weathering processes:
2086 Assessing controls on Si transfer to the ocean. Comptes Rendus Geoscience 344, 723–
2087 738.
2088 Opfergelt, S., Eiriksdottir, E.S., Burton, K.W., Einarsson, A., Siebert, C., Gislason, S.R.,
2089 Halliday, A.N., 2011. Quantifying the impact of freshwater diatom productivity on

2090 silicon isotopes and silicon fluxes: Lake Myvatn, Iceland. Earth Planet. Sci. Lett. 305, 73–
2091 82.
2092 Panizzo, V.N., Crespin, J., Crosta, X., Shemesh, A., Masse, G., Yam, R., Mattielli, N., Cardinal,
2093 D., 2014. Sea ice diatom contributions to Holocene nutrient utilization in East
2094 Antarctica. Paleoceanography 29, 328–342.
2095 Panizzo, V.N., Swann, G.E.A., Mackay, A.W., Vologina, E., Sturm, M., Pashley, V.,
2096 Horstwood, M.S.A., 2016. Insights into the transfer of silicon isotopes into the sediment
2097 record. Biogeosciences 13, 147–157.
2098 Panizzo, V.N., Swann, G.E., Mackay, A.W., Vologina, E.G., Pashley, V.H., Horstwood, M.S.A.,
2099 2017. Constraining modern-day silicon cycling in Lake Baikal. Global Biogeochem. Cycles
2100 2017, 556–574.
2101 Panizzo, V.N., Roberts, S., Swann, G.A.A., McGowan, S., Mackay, A.W., Vologina, E.,
2102 Pashley, V., Horstwood, M.S.A., 2018a. Spatial differences in dissolved silicon utilisation
2103 in Lake Baikal, Siberia: disentangling the effects of high diatom biomass events and
2104 eutrophication. Limnol. Oceanogr. doi: 10.1002/lno.10792
2105 Panizzo, V.N., Swann, G.E.A., Mackay, A.W., Pashley, V., Horstwood, M.S.A., 2018b.
2106 Modelling silicon supply during the Last Interglacial (MIS 5e) at Lake Baikal. Quatern.
2107 Sci. Rev. 190, 114-122.
2108 Parnell, A.C., Phillips, D.L., Bearhop, S., Semmens, B.X., Ward, E.J., Moore, J.W., Jackson,
2109 A.L., Grey, J., Kelly, D.J., Inger, R., 2013. Bayesian stable isotope mixing models.
2110 Environmetrics 24, 387–399.
2111 Patterson, W.P., Smith, G.R., Lohmann, K.C., 1993. Continental paleothermometry and
2112 seasonality using the isotopic composition of aragonitic otoliths of freshwater fishes.
2113 Geophys. Monogr. 78, 191–202.
2114 Pentecost, A., Andrews, J.E., Dennis, P.F., Marca-Bell, A., Dennis, S., 2006. Charophyte
2115 growth in small temperate water bodies: Extreme isotopic disequilibrium and
2116 implications for the palaeoecology of shallow marl lakes. Palaeogeogr., Palaeoclimatol.,
2117 Palaeoecol. 240, 389–404.
2118 Perga, M.-E., 2010. Potential of $\delta^{13}C$ and $\delta^{15}N$ of cladoceran subfossil exoskeletons for
2119 paleo-ecological studies. J. Paleolimnol. 44, 387–395.
2120 Perga, M.-E., 2011. Taphonomic and early diagenetic effects on the C and N stable isotope
2121 composition of cladoceran remains: implications for paleoecological studies. J.
2122 Paleolimnol. 46, 203–213.
2123 Perga, M.E., Gerdeaux, D., 2003. Using the $\delta^{13}C$ and $\delta^{15}N$ of whitefish scales for
2124 retrospective ecological studies: changes in isotope signatures during the restoration of
2125 Lake Geneva, 1980–2001. J. Fish Biol. 63, 1197–1207.
2126 Perga, M.E., Gerdeaux, D., 2006. Seasonal variability in the $\delta^{13}C$ and $\delta^{15}N$ values
2127 of the zooplankton taxa in two alpine lakes. Acta Oecol. 30, 69–77.
2128 Perga, M.-E., Desmet, M., Enters, D., Reyss, J.-L., 2010. A century of bottom-up and top-
2129 down driven changes on a lake planktonic food web: A paleoecological and
2130 paleoisotopic study of Lake Annecy, France. Limnol. Oceanogr. 55, 803–816.
2131 Perga, M.-E., Maberly, S.C., Jenny, J.-P., Alric, B., Pignol, C., Naffrechoux, E., 2016. A
2132 century of human-driven changes in the carbon dioxide concentration of lakes. Glob.
2133 Biogeochem. Cycles 30, 93–104.
2134 Phillips, D.L., Inger, R., Bearhop, S., Jackson, A.L., Moore, J.W., Parnell, A.C., Semmens, B.X.,
2135 Ward, E.J., 2014. Best practices for use of stable isotope mixing models in food-web
2136 studies. Canadian Journal of Zoology 92, 823–835.

2137 Pinnegar, J.K., Polunin, N.V.C., 1999. Differential fractionation of $\delta^{13}C$ and $\delta^{15}N$ among fish
2138 tissues: implications for the study of trophic interactions. Funct. Ecol. 13, 225–231.
2139
2140 Reynolds, B.C., Aggarwal, J., André, L., Baxter, D., Beucher, C., Brzezinski, M.A., Engström,
2141 E., Georg, R.B., Land, M., Leng, M.J., Opfergelt, S., Rodushkin, I., Sloane, H.S., van den
2142 Boorn, S.H.J.M., Vroon, P.Z., Cardinal, D. 2007. An inter-laboratory comparison of Si
2143 isotope reference materials. JAAS 22, 561–568.
2144 Rings, A., Lücke, A., Schleser, G.H., 2004. A new method for the quantitative separation of
2145 diatom frustules from lake sediments. Limnol. Oceanogr. Meth. 2, 25–34.
2146 Rinta, P., van Hardenbroek, M., Jones, R.I., Kankaala, P., Rey, F., Szidat, S., Wooller, M.J.,
2147 Heiri, O., 2016. Land Use Affects Carbon Sources to the Pelagic Food Web in a Small
2148 Boreal Lake. PLOS ONE 11, e0159900.
2149 Roberts, L., Holmes, J. A., Horne, D. J., Leng, M. J. In prep. Effects of cleaning methods
2150 upon preservation of isotopes and trace elements in ostracods. Quat. Sci. Rev. 189,
2151 197–209.
2152 Robinson, R.S., Brunelle, B.G., Sigman, D.M., 2004. Revisiting nutrient utilization in the
2153 glacial Antarctic: Evidence from a new method for diatom-bound N isotopic analysis.
2154 Paleoceanography 19, doi:10.1029/2003PA000996.
2155 Rozanski, K., Klisch, M.A., Wachniew, P., Gorczyca, Z., Goslar, T., Edwards, T.W.D.,
2156 Shemesh, A., 2010. Oxygen-isotope geothermometers in lacustrine sediments: New
2157 insights through combined $\delta^{18}O$ analyses of aquatic cellulose, authigenic calcite and
2158 biogenic silica in Lake Gosciaz, central Poland. Geochim. Cosmochim. Acta 74, 2957–
2159 2969.
2160 Sancetta, C., Heusser L., Labeyrie L., Sathy Naidu A., Robinson S.W., 1985. Wisconsin -
2161 Holocene paleoenvironment of the Bering Sea: evidence from diatoms, pollen, oxygen
2162 isotopes and clay minerals. Mar. Geol. 62, 55–68.
2163 Sanseverino, A.M., Bastviken, D., Sundh, I., Pickova, J., Enrich-Prast, A., 2012. Methane
2164 Carbon Supports Aquatic Food Webs to the Fish Level. PLoS ONE 7, e42723.
2165 Schilder, J., Tellenbach, C., Möst, M., Spaak, P., van Hardenbroek, M., Wooller, M.J., Heiri,
2166 O., 2015a. The stable isotopic composition of *Daphnia* ephippia reflects changes in $\delta^{13}C$
2167 and $\delta^{18}O$ values of food and water. Biogeosciences 12, 3819–3830.
2168 Schilder, J., Bastviken, D., van Hardenbroek, M., Leuenberger, M., Rinta, P., Stötter, T.,
2169 Heiri, O., 2015b. The stable carbon isotopic composition of Daphnia ephippia in small,
2170 temperate lakes reflects in-lake methane availability. Limnol. Oceanogr. 60, 1064–1075.
2171 Schilder, J., van Hardenbroek, M., Bodelier, P., Kirilova, E.P., Leuenberger, M., Lotter, A.F.,
2172 Heiri, O., 2017. Trophic state changes can affect the importance of methane-derived
2173 carbon in aquatic food webs. Proceedings of the Royal Society B: Biological Sciences
2174 284.
2175 Schilder, J., van Roij, L., Reichart, G.-J., Sluijs, A., Heiri, O., 2018. Variability in $\delta^{13}C$ values
2176 between individual *Daphnia* ephippia: Implications for palaeo-studies. Quatern. Sci.
2177 Rev. 189: 127–133.
2178 Schimmelmann, A., 2011. Carbon, Nitrogen and Oxygen Stable Isotope Ratios in Chitin, in:
2179 Gupta, N.S. (Ed.), Chitin. Springer Netherlands, pp. 81–103.
2180 Schimmelmann, A., DeNiro, M.J., 1986a. Stable isotopic studies on chitin. II. The 13C/12C
2181 and $^{15}N/^{14}N$ ratios in arthropod chitin. Contrib. Mar. Sci. 29, 113–130.
2182 Schimmelmann, A., DeNiro, M.J., 1986b. Stable isotopic studies on chitin. III. The D/H and
2183 $^{18}O/^{16}O$ ratios in arthopod chitin. Geochim. Cosmochim. Acta 50, 1485–1496.

2184 Schimmelmann, A., DeNiro, M.J., Poulicek, M., Voss-Foucart, M.-F., Goffinet, G., Jeuniaux,
2185 C., 1986. Stable Isotopic Composition of Chitin from Arthropods Recovered in
2186 Archaeological Contexts as Palaeoenvironmental Indicators. J. Archaeol. Sci. 13, 553–
2187 566.
2188 Schleser, G.H., Lücke, A., Moschen, R., Rings, A., 2001. Separation of diatoms from
2189 sediment and oxygen isotope extraction from their siliceous valves – a new approach.
2190 Terra Nostra, 2001/3. Schriften der Alfred-Wegener-Stiftung (6th workshop of the
2191 European lake drilling programme, Potsdam.), pp. 187–191.
2192 Schmidt, M., Botz, R., Stoffers, P., Anders, T., Bohrmann, G., 1997. Oxygen isotopes in
2193 marine diatoms: A comparative study of analytical techniques and new results on the
2194 isotopic composition of recent marine diatoms. Geochim. Cosmochim. Acta 61, 2275–
2195 2280.
2196 Schmidt, M., Botz, R., Rickert, D., Bohrmann, G., Hall, S.R., Mann, S., 2001. Oxygen isotope
2197 of marine diatoms and relations to opal-A maturation. Geochim. Cosmochim. Acta 65,
2198 201–211.
2199 Schneider, S. C., Garcia, A., Martin-Closas, C., Chivas, A. R., 2015. The role of charophytes
2200 (Charales) in past and present environments: an overview. Aquatic Botany 120, 2–6.
2201 Schwalb, A., Lister, G.S., Kelts, K., 1994. Ostracode carbonate δ^{18}O- and δ^{13}C-signatures of
2202 hydrological and climatic changes affecting Lake Neuchâtel (Switzerland) since the Late
2203 Pleistocene. J. Paleolimnol. 11, 3–17.
2204 Schwalb, A., Dean, W., Güde, H., Hanisch, S., Sobek, S., Wessels, M., 2013. Benthic
2205 ostracode δ^{13}C as sensor for early Holocene establishment of modern circulation
2206 patterns in Central Europe. Quaternary Sci. Rev. 66, 112–122.
2207 Seddon, A.W.R., Froyd, C.A., Witkowski, A., Willis, K.J., 2014. A quantitative framework for
2208 analysis of regime shifts in a Galápagos coastal lagoon. Ecology 95, 3046–3055.
2209 Semmens, B.X., Ward, E.J., Moore, J.M., Drummond, C.T., 2009. Quantifying inter- and
2210 intra-population niche variability using hierarchical Bayesian stable isotope mixing
2211 models. PloS One 4, e6187.
2212 Shanahan, T.M., Pigati, J.S., Dettman, D.L., Quade, J., 2005. Isotopic variability in the
2213 aragonite shells of freshwater gastropods living in springs with nearly constant
2214 temperature and isotopic composition. Geochim. Cosmochim. Acta 69, 3949–3966.
2215 Shemesh, A., Charles, C.D., Fairbanks R.G., 1992. Oxygen isotopes in biogenic silica: global
2216 changes in ocean temperature and isotopic composition. Science 256, 1434–1436.
2217 Shemesh, A., Burckle, L.H., Hays, J.D., 1995. Late Pleistocene oxygen isotope records of
2218 biogenic silica from the Atlantic sector of the Southern Ocean. Paleoceanography 10,
2219 179–196.
2220 Shemesh, A., Rietti-Shati, M., Rioual, P., Battarbee, R.W., de Beaulieu, J-L., Reille, M.,
2221 Andrieu, V., Svobodova, H., 2001 An oxygen isotope record of lacustrine opal from a
2222 European Maar indicates climatic stability during the last interglacial. Geophys. Res.
2223 Lett. 28, 2305–2308.
2224 Siegenthaler, U., Eicher, U., 1986. Stable oxygen and carbon isotope analyses, in: Berglund,
2225 B.E. (Ed.), Handbook of Holocene Palaeoecology and Palaeohydrology. John Wiley &
2226 Sons Ltd., New York, pp. 407–421.
2227 Singer, A.J., Shemesh, A., 1995. Climatically Linked Carbon-Isotope Variation during the
2228 Past 430,000 Years in Southern-Ocean Sediments. Paleoceanography 10, 171–177.

2229 Smyntek, P.M., Maberly, S.C., Grey, J., 2012. Dissolved carbon dioxide concentration
2230 controls baseline stable carbon isotope signatures of a lake food web. Limnol.
2231 Oceanogr. 57, 1292–1302.
2232 Snelling, A.M., Swann, G.E.A., Leng, M.J., Pike J. 2013. A Micro-manipulation technique for
2233 the purification of diatoms for isotope and geochemical analysis. Silicon 5, 13–17.
2234 Solomon, C., Cole, J., Doucett, R., Pace, M., Preston, N., Smith, L., Weidel, B., 2009. The
2235 influence of environmental water on the hydrogen stable isotope ratio in aquatic
2236 consumers. Oecologia 161, 313–324.
2237 Soto, D.X., Wassenaar, L.I., Hobson, K.A., 2013. Stable hydrogen and oxygen isotopes in
2238 aquatic food webs are tracers of diet and provenance. Funct. Ecol. 27, 535–543.
2239 Soulié-Märsche, I., Bieda, S., Lafond, R., Maley, J., M'Baitoudji, Vincent, P.M., Faure, H.,
2240 2010. Charophytes as bio-indicators for lake level high stand at “Trou au Natron”,
2241 Tibesti, Chad, during the Late Pleistocene. Global Planet. Change 72, 334–340.
2242 Spero, H.J., Lea, D.W., 1993. Intraspecific stable isotope variability in the planktonic
2243 foraminifera Globigerinoides sacculifer: results from laboratory experiments. Marine
2244 Micropaleontolgy 22, 221–234.
2245 Spero, H.J. Lea, D.W., 1996. Experimental determination of stable isotope variability in
2246 Globigerina bulloides: implications for paleoceanographic reconstructions. Marine
2247 Micropaleontology 28, 231–246.
2248 Spero, H. J., Bijma, J., Lea, D.W., Bemis, B., 1997. Effect of seawater carbonate chemistry
2249 on planktonic foraminiferal carbon and oxygen isotope values. Nature 390, 497–500.
2250 Stankiewicz, B.A., Van Bergen, P.F., Duncan, I.J., Carter, J.F., Briggs, D.E., Evershed, R.P.,
2251 1996. Recognition of Chitin and Proteins in Invertebrate Cuticles Using Analytical
2252 Pyrolysis/Gas Chromatography and Pyrolysis/Gas Chromatography/Mass Spectrometry.
2253 Rapid Commun. Mass Sp. 10, 1747–1757.
2254 Stankiewicz, B.A., Mastalerz, M., Hof, C.H.J., Bierstedt, A., Flannery, M.B., Briggs, D.E.G.,
2255 Evershed, R.P., 1998. Biodegradation of the chitin-protein complex in crustacean
2256 cuticle. Org. Geochem. 28, 67–76.
2257 Stötter, T., Bastviken, D., Bodelier, P.L.E., van Hardenbroek, M., Rinta, P., Schilder, J.,
2258 Schubert, C.J., Heiri, O., 2018. Abundance and $\delta^{13}C$ values of fatty acids in lacustrine
2259 surface sediments: Relationships with in-lake methane concentrations. Quaternary Sci.
2260 Rev. 191, 337-347.
2261 Street-Perrott, F.A., Holmes J.A., Waller, M.P., Allen, M.J., Barber, N.G.H., Fothergill, P.A.,
2262 Harkness, D.D., Ivanovich, M., Kroon, D., Perrott, R.A., 2000. Drought and dust
2263 deposition in the West African Sahel: A 5500-year record from Kajemarum Oasis,
2264 northeastern Nigeria. The Holocene, 10, 293–302.
2265 Street-Perrott, F.A., Barker, P.A., Leng, M.J., Sloane, H.J., Wooller, M.J., Ficken, K.J., Swain,
2266 D.L., 2008. Towards an understanding of late Quaternary variations in the continental
2267 biogeochemical cycle of silicon: multi-isotope and sediment-flux data for Lake Rutundu,
2268 Mt Kenya, East Africa, since 38 ka BP. J Quat. Sci 23, 375–387.
2269 Street-Perrott, F.A., Holmes, J.A., Robertson, I., Ficken, K.J., Koff, T., Loader, N.J., Marshall,
2270 J.D., Martma, T., 2018. The Holocene isotopic record of aquatic cellulose from Lake
2271 Äntu Sinijärv, Estonia: Influence of changing climate and organic-matter sources.
2272 Quaternary Sci. Rev. 193, 68-83.
2273 Struck, U., Voss, M., Von Bodungen, B., Mumm, N., 1998. Stable Isotopes of Nitrogen in
2274 Fossil Cladoceran Exoskeletons: Implications for Nitrogen Sources in the Central Baltic
2275 Sea During the Past Century. Naturwissenschaften 85, 597–603.

2276 Sturm, C., Zhang, Q., Noone, D., 2010. An introduction to stable water isotopes in climate
2277 models: benefits of forward proxy modelling for paleoclimatology. Clim. Past 6, 115–
2278 129.
2279 Sun, X.L., Andersson, P.S., Humborg, C., Pastuszak, M., Morth, C.M., 2013. Silicon isotope
2280 enrichment in diatoms during nutrient-limited blooms in a eutrophied river system.
2281 Journal of Geochemical Exploration 132, 173–180.
2282 Sutton, J.N., Varela, D.E., Brzezinski, M.A., Beucher, C.P., 2013. Species-dependent silicon
2283 isotope fractionation by marine diatoms. Geochim. Cosmochim. Acta 104, 300–309.
2284 Sutton, J.N., André, L., Cardinal, D., Conley, D.J., de Souza, G.F., Dean, J., Dodd, J., Ehlert,
2285 C., Ellwood, M.J., Frings, P.J., Grasse, P., Hendry, K., Leng, M.J., Michalopoulos, P.,
2286 Panizzo, V.N., Swann, G.E.A., 2018. A Review of the Stable Isotope Bio-geochemistry of
2287 the Global Silicon Cycle and Its Associated Trace Elements. Front Earth Sci 5. UNSP 112
2288 doi: 10.3389/feart.2017.00112
2289 Swann, G.E.A., Leng, M.J. 2009. A review of diatom $\delta^{18}O$ in palaeoceanography. Quatern.
2290 Sci. Rev. 28, 384–398.
2291 Swann, G.E.A., Leng, M.J., Juschus, O., Melles, M., Brigham-Grette, J., Sloane, H.J., 2010. A
2292 combined oxygen and silicon diatom isotope record of Late Quaternary change in Lake
2293 El'gygytgyn, North East Siberia. Quatern. Sci. Rev. 29, 774–786.
2294 Swann, G.E.A., Mackay, A.W., Vologina, E., Jones, M.D., Panizzo, V.N., Leng, M.J., Sloane,
2295 H.J., Snelling, A.M., Sturm, M., 2018. Lake Baikal isotope records of Holocene Central
2296 Asian precipitation. Quaternary Sci. Rev. 189, 210–222.
2297 Taipale, S.J., Peltomaa, E., Hiltunen, M., Jones, R.I., Hahn, M.W., Biasi, C., Brett, M.T., 2015.
2298 Inferring Phytoplankton, Terrestrial Plant and Bacteria Bulk $\delta^{13}C$ Values from Compound
2299 Specific Analyses of Lipids and Fatty Acids. PLOS ONE 10, e0133974.
2300 Templeton, A.S., Chu, K.-H., Alvarez-Cohen, L., Conrad, M.E., 2006. Variable carbon
2301 isotope fractionation expressed by aerobic CH_4-oxidizing bacteria. Geochim.
2302 Cosmochim. Acta 70, 1739–1752.
2303 Thorp, J.H., Rogers, D.C., 2015. Freshwater Invertebrates: Ecology and General Biology.
2304 Academic Press, Boston.
2305 Turney, C.S.M., 1999. Lacustrine Bulk Organic δ13C in the British Isles during the Last
2306 Glacial-Holocene Transition (14–9 ka 14C BP). Arct. Antarct. Alp. Res. 31, 71–81.
2307 Tyler, J.J., Leng, M.J., Sloane, H.J., 2007. The effects of organic removal treatment on the
2308 integrity of $\delta^{18}O$ measurements from biogenic silica. J. Paleolimnol. 37, 491–497.
2309 Tyler, J.J., Sloane, H.J., Rickaby, R.E.M., Cox, E.J., Leng, M.J. 2017. Post-mortem oxygen
2310 isotope exchange within cultured diatom silica. Rapid Commun. Mass Sp. doi:
2311 10.1002/rcm.7954
2312 Urban, M.A., Nelson, D.M., Jiménez-Moreno, G., Châteauneuf, J.J., Pearson, A., Hu, F.S.,
2313 2010. Isotopic evidence of C4 grasses in southwestern Europe during the Oligocene–
2314 Middle Miocene. *Geology* 38: 1091–1094.
2315 Urban, M.A., Nelson, D.M., Kelly, R., Ibrahim, T., Dietze, M., Pearson, A., Hu, F.S., 2013. A
2316 hierarchical Bayesian approach to the classification of C3 and C4 grass pollen based on
2317 SPIRAL $\delta^{13}C$ data. *Geochim Cosmochim Acta* 121: 168–176.
2318 van Bennekom, A.J., van der Gaast, S.J., 1976. Possible clay structures in frustules of living
2319 diatoms. Geochim. Cosmochim. Acta 40, 1–6.
2320 van Hardenbroek, M., Heiri, O., Lotter, A., 2010a. Efficiency of different mesh sizes for
2321 isolating fossil chironomids for stable isotope and radiocarbon analyses. J. Paleolimnol.
2322 44, 721–729.

2323 van Hardenbroek, M., Heiri, O., Grey, J., Bodelier, P., Verbruggen, F., Lotter, A., 2010b.
2324 Fossil chironomid $\delta^{13}C$ as a proxy for past methanogenic contribution to benthic food
2325 webs in lakes? J. Paleolimnol. 43, 235–245.
2326 van Hardenbroek, M., Lotter, A.F., Bastviken, D., Duc, N.T., Heiri, O., 2012. Relationship
2327 between $\delta^{13}C$ of chironomid remains and methane flux in Swedish lakes. Freshwat. Biol.
2328 57, 166–177.
2329 van Hardenbroek, M., Gröcke, D., Sauer, P., Elias, S., 2013a. North American transect of
2330 stable hydrogen and oxygen isotopes in water beetles from a museum collection. J.
2331 Paleolimnol. 48, 461–470.
2332 van Hardenbroek, M., Heiri, O., Parmentier, F.J.W., Bastviken, D., Ilyashuk, B.P., Wiklund,
2333 J.A., Hall, R.I., Lotter, A.F., 2013b. Evidence for past variations in methane availability in
2334 a Siberian thermokarst lake based on $\delta^{13}C$ of chitinous invertebrate remains.
2335 Quaternary Sci. Rev. 66, 74–84.
2336 van Hardenbroek, M., Lotter, A.F., Bastviken, D., Andersen, T.J., Heiri, O., 2014. Taxon-
2337 specific δ13C analysis of chitinous invertebrate remains in sediments from Strandsjön,
2338 Sweden. J. Paleolimnol. 52, 95–105.
2339 van Hardenbroek, M., Leuenberger, M., Hartikainen, H., Okamura, B., Heiri, O., 2016.
2340 Bryozoan stable carbon and hydrogen isotopes: relationships between the isotopic
2341 composition of zooids, statoblasts and lake water. Hydrobiologia 765, 209–223.
2342 van Hardenbroek, M., Rinta, P., Wooller, M.J., Schilder, J., Stötter, T., Heiri, O., 2018.
2343 Flotsam samples can help explain the $\delta^{13}C$ and $\delta^{15}N$ values of invertebrate resting
2344 stages in lake sediment records. Quatern. Sci. Rev. 189: 187–196.
2345 Varela, D.E., Pride, C.J., Brzezinski, M.A., 2004. Biological fractionation of silicon isotopes in
2346 Southern Ocean surface waters. Global Biogeochem. Cycles 18.
2347 Ventura, M., Jeppesen, E., 2010. Evaluating the need for acid treatment prior to $\delta^{13}C$ and
2348 $\delta^{15}N$ analysis of freshwater fish scales: effects of varying scale mineral content, lake
2349 productivity and CO2 concentration. Hydrobiologia 644, 245–259.
2350 Verbruggen, F., Heiri, O., Reichart, G.J., De Leeuw, J., Nierop, K., Lotter, A., 2010a. Effects
2351 of chemical pretreatments on $\delta^{18}O$ measurements, chemical composition, and
2352 morphology of chironomid head capsules. J. Paleolimnol. 43, 857–872.
2353 Verbruggen, F., Heiri, O., Reichart, G.J., Lotter, A.F., 2010b. Chironomid $\delta^{18}O$ as a proxy for
2354 past lake water $\delta^{18}O$: a Lateglacial record from Rotsee (Switzerland). Quaternary Sci.
2355 Rev. 29, 2271–2279.
2356 Verbruggen, F., Heiri, O., Reichart, G.J., Blaga, C., Lotter, A.F., 2011. Stable oxygen isotopes
2357 in chironomid and cladoceran remains as indicators for lake-water $\delta^{18}O$. Limnol.
2358 Oceanogr. 56, 2071–2079.
2359 Visconti, A., Volta, P., Fadda, A., Di Guardo, A., M., 2014. Seasonality, littoral versus pelagic
2360 carbon sources, and stepwise ^{15}N-enrichment of pelagic food web in a deep subalpine
2361 lake: the role of planktiverous fish. Can. J. Fish. Aquat. Sci. 71, 436–446.
2362 von Grafenstein, U., 2002. Oxygen-isotope studies of ostracods from deep lakes. In:
2363 Holmes, J.A., Chivas, A.R. (Eds.), The Ostracoda: Applications in Quaternary Research.
2364 Geophysical Monograph Series 131, 249–266. American Geophysical Union,
2365 Washington DC.
2366 von Grafenstein, U., Erlenkeuser, H., Brauer, A., Jouzel, J., Johnsen, S.J., 1999a. A mid-
2367 European decadal isotope-climate record from 15,500 to 5000 years BP. Science 284,
2368 1654–1657.

2369 von Grafenstein, U., Erlernkeuser, H., Trimborn, P., 1999b. Oxygen and carbon isotopes in
2370 modern fresh-water ostracod valves: assessing vital offsets and autecological effects of
2371 interest for palaeoclimate studies. Palaeogeog. Palaeoclimatol. Palaeoecol.148, 133–
2372 152.
2373 Vuorio, K., Meili, M., Sarvala, J., 2006. Taxon-specific variation in the stable isotopic
2374 signatures ($\delta^{13}C$ and $\delta^{15}N$) of lake phytoplankton. Freshwat. Biol. 51, 807–822.
2375 Wang, C-H., Yeh, H-W., 1985. Oxygen isotope compositions of DSDP Site 480 diatoms:
2376 Implications and applications. Geochim. Cosmochim. Acta 49, 1469–1478.
2377 Wang, Y., Francis, D., O'Brien, D., Wooller, M., 2008. A protocol for preparing subfossil
2378 chironomid head capsules (Diptera: Chironomidae) for stable isotope analysis in
2379 paleoclimate reconstruction and considerations of contamination sources. J.
2380 Paleolimnol. 40, 771–781. Wolfe, B.B., Falcone, M.D., Clogg-Wright, K.P., Mongeon,
2381 C.L., Yi, Y., Brock, B.E., St. Amour, N.A., Mark, W.A., Edwards, T.W.D., 2007.
2382 Progress in isotope paleohydrology using lake sediment cellulose. J. Paleolimnol. 37,
2383 221-231.
2384 Wang, Y., O'Brien, D., Jenson, J., Francis, D., Wooller, M., 2009. The influence of diet and
2385 water on the stable oxygen and hydrogen isotope composition of Chironomidae
2386 (Diptera) with paleoecological implications. Oecologia 160, 225–233.
2387 Webb, M., Barker, P.A., Wynn, P.M., Heiri, O., van Hardenbroek, M., Pick, F., Russell, J.M.,
2388 Stott, A.W., Leng, M.J., 2016. Interpretation and application of carbon isotope ratios in
2389 freshwater diatom silica. J. Quat. Sci. 31, 300–309.
2390 Wefer, G., Berger, W.H., 1991. Isotope paleontology: growth and composition of extant
2391 calcareous species. Mar. Geol. 100, 207–248.
2392 Wetzel, F., de Souza, G.F., Reynolds, B.C., 2014. What controls silicon isotope fractionation
2393 during dissolution of diatom opal? Geochim. Cosmochim. Acta 131, 128–137.
2394 Willis, J. H., 1999. Cuticular proteins in insects and crustaceans. American Zoologist 39,
2395 600–609.
2396 Wilson, K.E., Leng, M.J., Mackay, A.W. (2014) The use of multivariate statistics to resolve
2397 multiple contamination signals in the oxygen isotope analysis of biogenic silica. J. Quat.
2398 Sci. 29, 641–649.
2399 White, R.M.P., Dennis, P.F., Atkinson, T.C. 1999. Experimental calibration and field
2400 investigation of the oxygen isotopic fractionation between biogenic aragonite and
2401 water. Rapid Comm. Mass Spectrom. 13, 1242–1247.
2402 Wolfe, B.B., Falcone, M.D., Clogg-Wright, K.P., Mongeon, C.L., Yi, Y., Brock, B.E., St. Amour,
2403 N.A., Mark, W.A., Edwards, T.W.D., 2007. Progress in isotope paleohydrology using lake
2404 sediment cellulose. J. Paleolimnol. 37, 221-231.
2405 Woodland, R.J., Rodríguez, M.A., Magnan, P., Glémet, H., Cabana, G., 2012. Incorporating
2406 temporally dynamic baselines in isotopic mixing models. Ecology 93, 131–144.
2407 Wooller, M.J., Francis, D., Fogel, M.L., Miller, G.H., Walker, I.R., Wolfe, A.P., 2004.
2408 Quantitative paleotemperature estimates from $\delta^{18}O$ of chironomid head capsules
2409 preserved in arctic lake sediments. J. Paleolimnol. 31, 267–274.
2410 Wooller, M.J., Wang, Y., Axford, Y., 2008. A multiple stable isotope record of Late
2411 Quaternary limnological changes and chironomid paleoecology from northeastern
2412 Iceland. J. Paleolimnol. 40, 63–77.
2413 Wooller, M., Pohlman, J., Gaglioti, B., Langdon, P., Jones, M., Walter Anthony, K., Becker,
2414 K., Hinrichs, K.-U., Elvert, M., 2012. Reconstruction of past methane availability in an

2415 Arctic Alaska wetland indicates climate influenced methane release during the past
2416 ~12,000 years. J. Paleolimnol. 48, 27–42.
2417 Wooller, M.J., Gaglioti, B., Fulton, T.L., Lopez, A., Shapiro, B., 2015. Post-glacial dispersal
2418 patterns of Northern pike inferred from an 8800 year old pike (*Esox* cf. *lucius*) skull from
2419 interior Alaska. Quaternary Sci. Rev. 120, 118–125.
2420 Xia, J., Ito, E., Engstrom, D.R., 1997a. Geochemistry of ostracode calcite: Part 1. An
2421 experimental determination of oxygen isotope fractionation. Geochim. Cosmochim.
2422 Acta 61, 377–382.
2423 Xia J, Haskell BJ, Engstrom DR, Ito E., 1997b. Holocene climate reconstructions from
2424 tandem trace-element and stable isotope composition of ostracodes from Coldwater
2425 Lake, North Dakota, USA. J. Paleolimnol. 17, 85–100.
2426 Yasuno, N., Shikano, S., Muraoka, A., Shimada, T., Ito, T., Kikuchi, E., 2012. Seasonal
2427 increase of methane in sediment decreases $\delta^{13}C$ of larval chironomids in a eutrophic
2428 shallow lake. Limnology 13, 107–116.
2429 Zhang, A., Zhang, J., Zhang, R., Xue, Y., 2014. Modified enrichment and purification
2430 protocol for dissolved silicon-isotope determination in natural waters. JAAS 29, 2414–
2431 2418.
2432 Zhao, Y., Nelson, D.M., Clegg, B.F., An, C.-B., Hu, F.S. 2017. Isotopic analysis on nanogram
2433 quantities of carbon from dissolved insect cuticle: a method for paleoenvironmental
2434 inferences. Rapid Commun. Mass Sp. 31, 1825–1834.
2435 Zhu, J., Lücke, A., Wissel, H., Mayr, C., Ohlendorf, C., Zolitschka, B., 2014. Characterizing
2436 oxygen isotope variability and host water relation of modern and subfossil aquatic
2437 mosses. Geochim. Cosmochim. Acta 130, 212–228.
2438

2439 **Supplementary Table:** *Mean weight and standard deviation of individual invertebrate*
2440 *remains. Data are derived from surface and down core sediments after chemical*
2441 *processing with 10 % HCl and 10 % KOH (van Hardenbroek et al. 2010b, 2012, 2013; van*
2442 *Hardenbroek, Heiri and Wooller unpublished). Note that values are means for samples*
2443 *containing different sizes of remains, including head capsules from different larval instars.*

	Surface samples weight (µg)	SD	n	Down core samples weight (µg)	SD	n
Chironomid head capsules						
Chironomus anthracinus-type	2.5	3.4	18			
Chironomus plumosus-type	2.8	2.7	14			
Chironomus spp.	2.1	2.1	81	1.2	0.4	30
Dicrotendipes	1.2	0.7	6			
Endochironomus	1.0	0.5	6			
Glyptotendipes	2.0	2.3	6			
Microtendipes	1.0	0.4	5			
Polypedilum	0.6	0.2	5			
Sergentia	0.9	0.3	1			
Chironomini (including all taxa)	1.6	2.2	241	1.0	0.4	76
Corynocera ambigua				1.3	0.1	7
Tanytarsini	0.6	0.5	122	0.5	0.2	39
Orthocladiinae	0.7	0.5	105	0.9	0.5	42
Diamesinae	1.9	0.9	3			
Tanypodinae	1.0	0.7	89	1.0	0.3	17
Cladoceran ephippia						
Daphnia spp.	2.8	2.9	148	2.8	2.7	51
Simocephalus vetulus	2.8	1.4	11			
Ceriodaphnia spp.	0.8	0.7	45			
Leydigia	1.1	0.8	5			
Chydorid	0.9	0.5	26			
Bryozoan statoblasts						
Plumatella spp.	1.2	0.7	116	0.6	0.3	23
Cristatella mucedo	30.3	28.1	66	21.0	8.1	15
Lophopus crystallina	4.6		1			
Pectinatella magnifica	18.1		2			
Other remains						
Ephemeroptera (mandible)	0.6	0.3	32			
Turbularia (coccoon)				1.2	0.2	18
Ostracoda (shell lining)	1.8	1.5	56			
Trichoptera (frontoclypeus/mandible)	2.2	3.7	32			
Sialis (frontoclypeus/mandible)	2.5	1.6	23			
Chaoborus (mandible)	1.5	1.0	37			
Chaoborus (thoracic horn)	0.9	0.2	7			

2444
2445
2446