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1 **Planktivorous vendace (*Coregonus albula*) utilize algae-derived fatty acids**
2 **for biomass increase and lipid deposition**

3
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15
16 **Running head:** Fatty acid accumulation in vendace

17

18

19 **Abstract**

20 Spatial and seasonal variation in the abundance and composition of phytoplankton and other
21 basal resources (bacteria, terrestrial detritus) influence the availability of essential
22 polyunsaturated fatty acids (PUFA) for upper trophic levels in lake food webs. We studied
23 accumulation, composition, and content of fatty acids in planktivorous vendace (*Coregonus*
24 *albula*) muscle tissue during the summer feeding period (May to late September) in six boreal
25 lakes. Vendace muscle fatty acid content increased from ~17.7 mg g⁻¹ DW to ~28.1 mg g⁻¹
26 DW from spring to summer, corresponding to a ~ 59 % increase in total fatty acids. PUFA
27 accounted for 45-65 % of all fatty acids, and the most abundant fatty acid was
28 docosahexaenoic acid (DHA, 22:6n-3). The DHA content remained relatively constant
29 throughout the sampling period (~7-8 mg g⁻¹ DW), but the proportion of DHA decreased
30 from ~40 % in spring to ~30 % in summer and autumn. The decrease in DHA proportion was
31 due to accumulation of other fatty acids, primarily C₁₈ PUFA (predominantly synthesized by
32 algae) and monounsaturated fatty acids. A similar fatty acid accumulation pattern was
33 observed in lipid storing copepods, an important prey for planktivorous vendace. The
34 abundance of bacterial and terrestrial biomarker fatty acids in vendace muscle and prey items
35 was low (<5% and <0.1%, respectively). The increase in weight and the deposition of internal
36 energy stores in vendace were achieved predominantly by algae-based food chain. This has
37 ecological implications, as vendace requires sufficient lipid stores for reproduction in autumn
38 and survival over winter.

39

40

41 **Key words:** algal biomarkers, bacterial biomarkers, dissolved organic carbon, pelagic food
42 web, large lakes

43

44 Introduction

45 Planktivorous fish in boreal lakes experience high seasonal variability in prey availability.
46 The ability to store energy in the form of lipid deposits is crucial for winter survival when
47 zooplankton prey is less abundant (Ågren, Muje, Hänninen, Herranen & Penttilä, 1987; Lahti
48 & Muje, 1991; Linko, Rajasilta & Hiltunen, 1992). Reproductive success in late autumn is
49 also highly dependent on lipid gain during the previous summer (Ågren et al., 1987; Lahti &
50 Muje, 1991; Linko et al., 1992). For reproduction, the abundance of polyunsaturated fatty
51 acids (PUFA) from the n-6 and n-3 families have been shown to be particularly important
52 (Sargent, Bell, McEvoy, Tocher & Estevez, 1999). Animals need PUFA for various
53 physiological processes, e.g. maintaining membrane fluidity and as precursors of local tissue
54 hormones, but most organisms lack the ability to biosynthesize these molecules *de novo* and
55 therefore must obtain them from their diet (Glencross, 2009). In aquatic food webs, algae are
56 the major source of n-3 and n-6 PUFA (Ahlgren, Lundstedt, Brett & Forsberg, 1990; Brett,
57 Kainz, Taipale & Seshan, 2009; Taipale et al., 2013, 2016). Although some fish species are
58 able to synthesize the long chained PUFA, eicosapentaenoic acid (EPA 20:5n-3),
59 docosahexaenoic acid (DHA, 22:6n-3) and arachidonic acid (ARA 20:4n-6), from shorter C₁₈
60 PUFA, this process is generally regarded as inefficient (Tocher, 2003). Dietary EPA and
61 DHA are shown to be important for optimal health and somatic growth of zooplankton and
62 fish (Ahlgren et al., 1990; Mairesse, Thomas, Gardeur & Brun-Bellut, 2007). In aquatic
63 environments, algae (phytoplankton and attached algae) are the main producers of EPA and
64 DHA, but different algal classes have very distinct fatty acid compositions and they differ in
65 their EPA and DHA content (Ahlgren et al., 1990; Taipale et al. 2013; Galloway & Winder,
66 2015). For example, cryptophytes and dinoflagellates are able to biosynthesize EPA and
67 DHA, whereas green algae and cyanobacteria contain predominantly the shorter chained C₁₈
68 PUFA (α -linolenic acid, ALA, 18:3n-3). Thus, the phytoplankton community composition

69 strongly influences the food web processes and availability of PUFA to upper trophic level
70 consumers (Brett & Müller-Navarra, 1997; Müller-Navarra, Brett, Liston & Goldman, 2000;
71 Galloway & Winder, 2015; Strandberg et al., 2015b; Taipale et al., 2016).

72 The knowledge on fatty acids in different basal resources can be applied for tracing the
73 pathways of autochthonous and allochthonous organic carbon flow in lake ecosystems
74 (Desvillettes, Bourdier, Amblard & Barth, 1997; Galloway et al., 2014). Although consumer-
75 specific differences in lipid digestion and metabolism will modify the fatty acids at different
76 trophic levels, specific fatty acids may be traced through several trophic levels (Gladyshev et
77 al., 2011; Strandberg et al., 2015a). Odd-chained and/or branched fatty acids are typically
78 characterized as actinobacterial, whereas long-chain (≥ 24 carbon) saturated fatty acids are
79 considered to originate from detrital terrestrial particulate organic matter (t-POM)
80 (Desvillettes et al. 1997; Taipale, Kankaala, Hämäläinen & Jones, 2009; Galloway et al.,
81 2014). Similarly, low n-3/n-6 ratio (< 1) in zooplankton has been suggested to indicate an
82 allochthonous diet (Brett et al., 2009; Taipale, Kainz & Brett, 2015). In addition to dietary
83 differences in FA composition, zooplankton fatty acid composition is taxon-specific (Persson
84 & Vrede, 2006, Smyntek, Teece, Schulz & Storch, 2008; Hiltunen, Strandberg, Taipale &
85 Kankaala, 2015). Variation in the importance of autotrophic (algal) vs. heterotrophic
86 (bacterial) production at the base of the food web may also influence the fatty acid
87 composition and lipid accumulation of planktivorous fish.

88 Vendace (*Coregonus albula* (L.)) is a small (total length 10-25 cm) zooplanktivorous
89 coregonid inhabiting large to medium-sized boreal lakes, and is the most important
90 commercial fish in these lakes (Viljanen, 1986; Marjomäki, Keskinen & Karjalainen, 2016).
91 Vendace is abundant in oligo-mesotrophic lakes with low to moderate humic content and
92 well-oxygenated cooler water layers (Dembiński, 1971; Rask, Viljanen, & Sarvala, 1999;
93 Winfield, Fletcher & James, 2004). It is generally not present in small shallow lakes with

94 high terrestrial dissolved organic carbon (DOC) concentration due to the absence of a well-
95 oxygenated hypolimnion. Weight gain and biomass increase of planktivorous fish is strongly
96 related to prey availability and abundance. In boreal lakes, the main feeding period for
97 planktivorous vendace is summer, when zooplankton (copepods and cladocerans) are
98 abundant (e.g. Hamrin, 1983; Viljanen, 1983; Helminen, Sarvala & Hirvonen, 1990). During
99 the feeding season, vendace accumulate lipids in their tissues to ensure spawning in autumn
100 and survival over winter (Ågren et al., 1987; Lahti & Muje, 1991; Linko et al., 1992). Almost
101 all vendace spawn for the first time during their second autumn (age 1+) and in intensively
102 harvested vendace populations, up to 80% of all spawners belong to this age group (Huusko
103 & Hyvärinen, 2005; Karjalainen et al., 2016).

104 We studied the composition and accumulation of fatty acids in pelagic food chains in large
105 (>100 km²) boreal lakes in Eastern Finland with moderate (5 – 10 mg C L⁻¹) DOC
106 concentrations (Hiltunen et al., 2015; Strandberg et al., 2015a,b). Studies on fatty acids in
107 phyto- and zooplankton in these lakes have been previously published (see Hiltunen et al.,
108 2015; Strandberg et al., 2015a,b). Here we focus on seasonal changes in vendace fatty acid
109 composition and content during the feeding period before autumn spawning. We
110 hypothesized that 1) the seasonal changes in phytoplankton community composition, and
111 concurrent changes in zooplankton fatty acid composition, modify fatty acid composition of
112 planktivorous vendace, and 2) resources of allochthonous origin play a minor role in vendace
113 nutrition leading to low abundance of terrestrial and bacterial fatty acid biomarkers in
114 vendace.

115

116 **Materials and methods**

117 Pelagic food webs were studied in six large (area >100 km²) lake basins in eastern Finland. In
118 2011 we sampled lakes Kallavesi, Suvasvesi, Orivesi, Paasivesi, and Pyhäselkä. Each lake

119 was sampled 3 times: late May-early June (spring samples), late July- early August (summer
120 samples), and at the end of September (autumn samples). Additional samples were collected
121 from Lake Karjalan Pyhäjärvi in August 2012. Due to the lack of samples, data from the
122 latter lake is presented separately and excluded from detailed analysis. All the lakes are
123 hydrologically connected within the Vuoksi water course (see Hiltunen et al., 2015, for a map
124 of the sampling area). The studied lake basins are oligotrophic or mesotrophic with total
125 phosphorus concentrations varying between ca. 5 - 12 $\mu\text{g L}^{-1}$ and chlorophyll *a* concentration
126 ca. 2 – 6 $\mu\text{g L}^{-1}$ (Table 1). The concentration of dissolved organic carbon (DOC) ranged from
127 ca. 5 to 10 mg C L^{-1} .

128 For phyto- and zooplankton sampling, laboratory methods and detailed results see
129 Hiltunen et al. (2015) and Strandberg et al. (2015b). The fish samples here are the same as in
130 Strandberg et al. (2015a). Vendace were caught by gill netting (mesh sizes 13-14, 16-17 and
131 15-18 mm) or by trawling (mesh sizes 12 or 13 mm, trawl bottom 5 mm). For each sampling
132 day and site, at least three random fish were taken for analyses and the weight and length of
133 the fish were measured (total $n=52$). Although not systematically determined, presumably all
134 caught individuals were mature based on their size (see Table 2, Lahti & Muje, 1991,
135 Karjalainen et al., 2016). Replicates of dorsal muscle tissue were dissected and stored in -70
136 $^{\circ}\text{C}$ until fatty acid analyses. We also opportunistically analyzed roe samples. Roe samples
137 were collected from lakes Paasivesi ($n=2$), Kallavesi ($n=1$), Orivesi ($n=1$), Pyhäselkä ($n=2$),
138 and Suvasvesi ($n=1$), and stored in -70 $^{\circ}\text{C}$ until further analysis.

139 **Laboratory analyses**

140 Vendace muscle and roe samples were freeze-dried prior to lipid analyses. Muscle tissue of
141 each fish was analyzed in duplicate and the mean value was used. Only one roe sample per
142 fish was analyzed. Total lipids were extracted twice with chloroform:methanol (2:1). Fatty
143 acids were transmethylated by adding methanolic sulfuric acid (1 % v/v) and heating for 16 h

144 at 50°C. The fatty acid methyl esters were extracted twice with *n*-hexane/diethyl ether (1:1 by
145 vol), evaporated to dryness and suspended into *n*-hexane. The fatty acid methyl esters of
146 vendace samples were analyzed with a gas chromatograph (Agilent 6890N) equipped with a
147 mass spectrometer (Agilent 5973N). The GC method was as follows: inlet temperature 250
148 °C, splitless injection, initial oven temperature 60 °C maintained for 1.5 min, after which
149 oven temperature ramped 10 °C/min to 100 °C, and then 2 °C/min to 140 °C, followed by 1
150 °C/min increase to 180 °C and finally 2 °C/min to 210 °C. The final oven temperature was
151 maintained for 6 minutes. Helium was used as the carrier gas with an average velocity of 36
152 cm/sec. The column was an Agilent DB-23 (30 m x 0.25 mm x 0.15 µm). Peak identification
153 was based on mass spectra and standard FAME mixes (37 FAME mix from Sigma-Aldrich
154 and GLC-68D from Nu-Chek Prep). The GLC-68D reference standard was used for the
155 calibration curve. For fatty acids not present in the reference standard, the calibration curve
156 was calculated based on the structurally most similar fatty acid.

157 **Statistical analyses**

158 Fulton condition index of fish (K) was calculated as follows: $K=W/L^3$, where W= weight (g)
159 and L= length (cm), a scaling factor of 100 was applied to bring the value close to 1.
160 Multidimensional scaling (MDS) was used to visualize fatty acid profiles among vendace,
161 and permutational multivariate analysis of variance (PERMANOVA; Anderson, 2001) was
162 used to test for differences in vendace fatty acid composition between different seasons (fixed
163 factor) and lake basins (random factor). Vendace sampled from Karjalan Pyhjärvi only in
164 August 2012 were excluded from the analyses. Prior to the multivariate analyses, the fatty
165 acid data was arcsine square root transformed. Ordination was based on Euclidean distances,
166 where samples close to another are compositionally more similar than samples further apart.
167 Goodness of fit for the ordination is expressed as the stress value. The lower the stress, the
168 better objects are fitted in the multidimensional space. The significance of PERMANOVA

169 analyses was determined using unrestricted permutation of the raw data (9999 permutations)
170 with type III sums of squares, (Anderson, Gorley & Clarke, 2008). Multivariate analyses
171 (MDS, PERMANOVA) were done with PRIMER 6.0 with the PERMANOVA + add on.
172 Other statistical analyses were done with IBM SPSS Statistics 23 program package.
173 Correlations between the total fatty acid content and the proportions of PUFA,
174 monounsaturated fatty acids (MUFA), BAFA and the n-3/n-6 ratio were evaluated, and
175 bootstrapping (1000 bootstrap samples) was applied to estimate the confidence interval (95 %
176 biased corrected) for the correlation coefficient. Seasonal differences (spring, summer,
177 autumn) in the relative proportions of different fatty acid classes in seston, zooplankton and
178 vendace was investigated with ANOVA using Games-Howell post hoc test. Seasonal
179 differences in total fatty acid content of vendace were tested with Wilcoxon signed rank test
180 and proportional differences of specific fatty acids between vendace muscle and roe were
181 analyzed with Mann-Whitney U-test.

182

183 **Results**

184 The total amount of fatty acids in the vendace muscle tissue increased significantly from 17.7
185 mg g⁻¹ DW in spring to 28.1 mg g⁻¹ DW in summer in accordance with the simultaneous
186 increase in the condition index (Wilcoxon signed rank test, p<0.05, Table 2, Figure 1). The
187 fatty acid content of vendace in autumn did not differ from that of summer samples. The most
188 abundant fatty acids in the vendace dorsal muscle were 14:0, 16:0, 16:1n-7, 18:0, 18:1n-9,
189 18:1n-7, 18:2n-6, 18:3n-3, 18:4n-3, 20:4n-6 (ARA), 20:5n-3 (EPA), 22:5n-6, 22:5n-3 and
190 22:6n-3 (DHA), accounting for >89% of all fatty acids (Supplement 1). Polyunsaturated fatty
191 acids were the most abundant group of fatty acids, accounting for 45-65% of all fatty acids.
192 Of these, DHA and EPA were the most abundant ones accounting for ~ 34% and ~ 9% of all
193 fatty acids, respectively. Saturated fatty acids (SFA) accounted for ~33% of all fatty acids

194 during all seasons. The general fatty acid composition of vendace did not differ between the
195 lake basins (PERMANOVA, Pseudo- $F_{5,46} = 1.759$, $P = 0.1$).

196 The seasonal differences in the fatty acid composition, especially between spring and
197 summer/autumn (Fig. 2), of vendace were significant (PERMANOVA, Pseudo- $F_{2,49} = 13.224$,
198 $P < 0.001$). The proportion of C_{22} PUFA decreased in vendace muscle as the total fatty acid
199 content increased ($r = -0.842$, $P < 0.001$, 95 % CI $-0.896 - -0.782$; Fig. 3). This was mainly
200 due to decreasing proportion of DHA, from $\sim 40 \pm 4$ % in spring, to 29 ± 7 % in summer and
201 32 ± 4 % in autumn, although the DHA content in vendace muscle remained at the same level
202 (spring 6.9 ± 1.5 , summer 7.3 ± 1.6 , autumn 8.0 ± 1.2 mg g^{-1} DW). The seasonal increase in the
203 total fatty acid content in the muscle tissue was mainly due to accumulation of
204 monounsaturated fatty acids (sum of C_{14} , C_{16} and C_{18} MUFA), C_{18} and C_{20} PUFA, where
205 proportions correlated positively with increasing fatty acid content (Fig. 3). The n-3/n-6
206 PUFA ratio in vendace muscle varied between 3.2 and 10.0 (mean 6.0 ± 1.3), decreasing as
207 total fatty acid content increased ($r = -0.584$, $P < 0.001$).

208 Proportion of solely bacterial fatty acids (BAFA, branched or odd-chain) in vendace
209 muscle varied from less than 0.5 to 4 % and correlated positively with the total fatty acid
210 content of fish ($r = 0.864$, $P < 0.001$, Fig 3). Low proportions (≤ 0.5 %) of isoprenoid fatty
211 acids, 4,8,12- trimethyltridecanoic (4,8,12-TMTD) and 3,7,11,15-tetramethylhexadecenoic
212 (phytanic) acids were also detected. Trace values of very long chained PUFA (≥ 24 carbons)
213 were found in the summer samples (mean of all summer samples 0.1%). Traces (< 0.1 %) of
214 lignoceric acid (24:0) was detected in the muscle of only 15% of the studied fish, while in
215 most fish 24:0 was not found at all.

216 The most abundant fatty acids in vendace roe, sampled in autumn, were generally the
217 same as in the muscle of adult fish (Supplement 2). PUFA accounted for about 48 % of all
218 fatty acids in the roe, but the proportion of DHA (~ 16 %) was significantly lower (Mann-

219 Whitney U test, $P < 0.001$) than what was analyzed from the muscle tissue in autumn. The
220 total fatty acid content in roe was about 10 times higher than total fatty acid content of the
221 muscle tissue in the autumn. ($279 \pm 35 \text{ mg g}^{-1} \text{ DW}$). The mean DHA content of roe was ~
222 $44 \pm 8 \text{ mg g}^{-1} \text{ DW}$.

223

224 **Discussion**

225 The content and composition of fatty acids in vendace muscle tissue changed significantly
226 from spring to autumn. The low fatty acid content in spring can be explained by depletion of
227 internal energy stores during spawning in autumn and by limited food availability and in
228 winter (Ågren et al., 1987). Summer is the main feeding period for vendace, and the 1.6-fold
229 increase in total fatty acid content from spring to summer reflects the accumulation of lipids
230 and building of internal energy stores in preparation for reproduction and overwintering
231 (Ågren et al., 1987; Lahti & Muje, 1991). Fatty acids that most contributed to the increase in
232 total fatty acid content were MUFA (particularly 16:1n-7, 18:1n-9) and C₁₈ PUFA (18:2n-6,
233 18:3n-3, 18:4n-3), whereas long chain polyunsaturated DHA declined from ca. 40 % in
234 spring to 30 % in summer. Similar trends of declining proportion of C₂₂ PUFA and increasing
235 proportion of C₁₈ PUFA was also seen in calanoid copepods (*Limnocalanus macrurus*,
236 *Eudiaptomus* spp.) in the study lakes (Hiltunen et al., 2015, Table 3).

237 The seasonal trends in fatty acid composition of vendace and zooplankton corresponded
238 with changes in the phytoplankton community composition (Strandberg et al., 2015b). In
239 seston (fraction 0.22 – 50 µm), the most abundant group of fatty acids were the C₁₈ PUFA, ~
240 30 % of all fatty acids during all the seasons (Strandberg et al., 2015b, Table 3). C₁₈ PUFA is
241 common in a wide variety of algal groups, including cryptophytes and chrysophytes, as well
242 as green algae and cyanobacteria (Taipale et al., 2013), thus explaining the abundance and
243 availability of C₁₈ PUFA in the pelagic food chain throughout the sampling season.

244 Cryptophytes and diatoms were the most abundant phytoplankton groups in all the lakes and
245 seasons, but the proportion of dinoflagellates decreased and the proportion of cyanobacteria
246 and green algae increased from spring to fall (Strandberg et al., 2015b). Dinoflagellates are
247 abundant source of DHA and the decrease in the abundance of dinoflagellates from spring to
248 summer coincided with the decrease of DHA in seston (Strandberg et al., 2015b) as well as in
249 zooplankton and vendace (Table 3), thus, supporting our first hypothesis.

250 Zooplankton are an important link for the transfer of energy and nutrients from producers
251 to upper trophic level consumers. In pelagic food webs, EPA and DHA originating from
252 seston accumulate in zooplankton and fish (Kainz, Arts & Mazumder, 2004; Strandberg et
253 al., 2015a). The general fatty acid composition of vendace muscle tissue was characterized by
254 a high proportion of PUFA (45-65 % of all fatty acids), which was similar in magnitude to
255 copepod zooplankton, but greater than in cladocerans and seston (Hiltunen et al., 2015;
256 Strandberg et al. 2015a, Table 3). Taxonomic differences in fatty acid composition between
257 cladocerans and copepods might be an important factor determining the transfer of fatty acids
258 to vendace. Cladocerans are known to efficiently retain C₂₀ PUFA (EPA and ARA), but
259 almost completely lack C₂₂ PUFA, whereas copepods are abundant in DHA (Persson &
260 Vrede, 2006; Smyntek et al., 2008; Hiltunen et al., 2015).

261 Stomach content analyses have indicated that vendace predominately prey on cladocerans
262 in summer (Hamrin, 1983; Viljanen, 1983), corresponding with the decline of DHA
263 proportion in vendace from spring to summer. The dominance of cladocerans in vendace diet
264 in summer was also observed in our study lakes, Pyhäselkä and Kallavesi, where ~ 90% of
265 prey remains in vendace guts (n=13) consisted of cladocerans (Viljanen & Lehtovaara,
266 unpublished). In autumn, vendace diet has been suggested to shift more to copepods
267 (Northcote & Hammar, 2006). This shift could be expected to increase the proportion and
268 content of DHA in the muscle; however, we did not observe any such pattern. Allocation of

269 DHA to reproduction may also partly explain why copepod diet was not clearly reflected in
270 the muscle fatty acid composition (Dabrowski, 1982; Pickova, Kiessling, Pettersson & Dutta,
271 1999; Tocher, 2003). Nevertheless, the high proportion of DHA (on average 34 % of all fatty
272 acids) in vendace muscle suggests that copepods are an important prey item. Alternatively,
273 vendace may have elongated and desaturated shorter chain n-3 PUFA (e.g. 18:3n-3 and/or
274 20:5n-3) to produce DHA. Coregonid *Coregonus lavaretus maraena* has been shown to
275 biosynthesize DHA from shorter chained analogues (Watanabe et al., 1989), suggesting that
276 vendace may also elongate shorter chained fatty acids to DHA. However, it remains
277 questionable if this metabolic pathway would be efficient enough to result in high proportions
278 of DHA, analyzed in vendace muscle (Kaitaranta, 1980; Muje, Ågren, Lindqvist & Hänninen,
279 1989). Additionally, the very long chained polyunsaturated fatty acids (C₂₄-C₂₆ PUFA),
280 which are specific biomarkers for the large copepod *Limnocalanus macrurus* (Hiltunen,
281 Strandberg, Keinänen, Taipale & Kankaala, 2014), were found in vendace muscle,
282 demonstrating a clear dietary link between the copepod and vendace.

283 Reproductive success is highly dependent on lipid reserves in many fish because the
284 formation of gonads is very energy intensive and the produced eggs need to be provisioned
285 with lipids (Marshall, Yaragina, Lambert & Kjesbu, 1999; Pickova et al., 1999, Tocher,
286 2003). The fatty acid content of vendace roe was high ($279 \pm 35 \text{ mg g}^{-1}$) demonstrating that
287 reproduction requires substantial allocation of lipids, particularly in females (Lahti & Muje,
288 1991). About 50% of fatty acids in roe were PUFA, and although the proportion of DHA in
289 roe was lower than in the muscle tissue, due to the high total fatty acid content, the amount of
290 DHA in roe was about 6 times higher than in the muscle tissue.

291 We could not find any between-lake differences in the fatty acid composition and content
292 of vendace. This may be partly due to low number of analyzed individuals per lake, but
293 presumably also due to the similarity of the sampled lakes, which are interconnected in the

294 Vuoksi water course (Table 1). Although some differences in phytoplankton community
295 structure in relation to DOC concentration was observed in the study lakes, taxonomic
296 identity was the most important determinant (62%) of zooplankton fatty acid composition and
297 the role of environmental factors was minor (Hiltunen et al., 2015). The proportion of DHA
298 was much lower (~ 13%, Linko et al., 1992) in vendace of mesotrophic Säkylän Pyhäjärvi
299 (SW Finland) compared to vendace in our study lakes suggesting lower availability of DHA
300 from phytoplankton via zooplankton to vendace. Lake trophic state and increased nutrient
301 concentrations with concurrent changes in phytoplankton community composition is
302 connected to decreasing EPA and DHA in a predatory fish, the Eurasian Perch (*Perca*
303 *fluviatilis*, Taipale et al., 2016). In contrast to our study lakes, Lake Säkylän Pyhäjärvi has
304 suffered from eutrophication and experiences occasionally cyanobacteria blooms (Ventelä et
305 al., 2011), which may influence the availability of DHA to vendace.

306 Phytoplankton are the ultimate source of PUFA in pelagic food web, but other basal
307 resources may be important in providing energy and specific fatty acids. For instance,
308 branched and odd-chained fatty acids are thought to originate from bacterial production
309 (Taipale et al., 2009), which utilize both phytoplankton-derived and terrestrial-origin DOC as
310 a carbon source (e.g. Simon, Cho & Azam, 1992; Pérez & Sommaruga, 2006). The
311 proportion of BAFA in the seston was ~ 2% (Strandberg et al., 2015b) and in zooplankton
312 ~1-5% (Hiltunen et al., 2015), indicating that bacterial fatty acids are incorporated into the
313 pelagic food web but have a low contribution. This suggests that BAFA are not markedly
314 utilized for biomass increase and lipid deposition in vendace. However, the proportion of
315 BAFA increased with increasing lipid content in vendace, indicating direct dietary input.
316 Since fatty acid content in actinobacteria is relatively low (Taipale et al., 2014), it is not
317 surprising that the proportion of BAFA in vendace was minimal (0.5-4%) in comparison to
318 algae-derived PUFA. In an oligotrophic clear-water alpine lake, Lake Annecy (area 28 km²),

319 whitefish (*Coregonus lavaretus*) larvae contained similar proportions (2-4 %) of BAFA
320 (Perga, Bec & Anneville, 2009). Various fatty acid biomarkers have been applied to estimate
321 autochthonous and allochthonous sources of fatty acids in aquatic biota. Increased
322 proportions of n-6 PUFA (e.g. 18:2n-6) and lower n-3/n-6 ratio may indicate the abundance
323 of terrestrially derived fatty acids, as n-6 PUFA are generally more abundant in terrestrial
324 than aquatic environments (Hixson, Sharma, Kainz, Wacker & Arts, 2015). A low n-3/n-6
325 ratio (< 1) in fish and zooplankton may indicate a significant contribution of terrestrial fatty
326 acids (Ahlgren, Sonesten, Boberg & Gustafsson, 1996; Taipale et al., 2015). In the current
327 study, the mean n-3/n-6 ratio in vendace was high; varying from 3 to 10, suggesting that
328 allochthonous resources would not directly contribute to vendace diet. Another suggested
329 indicator for terrestrial fatty acids is lignoceric acid (C24:0; Perga et al., 2009). In the current
330 study only 15% of the studied fish contained trace levels of 24:0 (<0.1% of total fatty acids).
331 Thus, our second hypothesis was supported. In Lake Annecy, zooplankton and whitefish
332 larvae contained higher, but still small, proportions of 24:0 (0.5-2.9% of total fatty acids;
333 Perga et al., 2009). However, laboratory feeding experiments indicate that 24:0 is not
334 efficiently assimilated by cladocerans or copepods, suggesting that this fatty acid is a poor
335 indicator of terrestrial contribution in consumer diets (Taipale et al., 2015; Taipale et al.,
336 unpublished).

337 Our results indicate that the commercially important planktivorous fish, vendace, is
338 dependent on the algae-based food chain for deposition of internal energy stores in summer.
339 Compared to algal fatty acids, the proportion of terrestrial and microbial fatty acid
340 biomarkers were low suggesting that the utilization of terrestrial resources in pelagic food
341 webs of large boreal lakes is limited. The importance of algae-derived fatty acids in biomass
342 increase and fatty acid accumulation in vendace is ecologically significant because the
343 survival of coregonids in winter depends on body condition and presence of sufficient lipid

344 stores (Pangle, Sutton, Kinnunen & Hoff, 2004). Additionally, reproductive success is linked
345 with the accumulation of these lipids, particularly DHA (Sargent et al., 1999).

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357 **References:**

358

359 Ågren, J., Muje, P., Hänninen, O., Herranen, J., & Penttilä, I. (1987). Seasonal variations of
360 lipid fatty acids of boreal freshwater fish species. *Comparative Biochemistry and*
361 *Physiology B*, 88: 905-909.

362

363 Ahlgren, G., Lundstedt, L. Brett, M., & Forsberg, C. (1990). Lipid composition and food
364 quality of some freshwater phytoplankton for cladoceran zooplankters. *Journal of Plankton*
365 *Research*, 13: 809-818.

366

367 Ahlgren, G., Sonesten, L., Boberg, M., & Gustafsson I.-B. (1996). Fatty acid content of some
368 freshwater fish in lakes of different trophic levels – a bottom-up effect? *Ecology of*
369 *Freshwater Fish*, 5: 15-27.

370

371 Anderson, M. J. (2001). A new method for non-parametric multivariate analysis of variance.
372 *Austral Ecology*, 26: 32-46.

373

374 Anderson, M. J., Gorley, R. N., & Clarke, K. R. (2008). PERMANOVA+ for PRIMER: guide
375 to software and statistical methods. PRIMER-E Ltd, Plymouth, UK.

376

377 Brett, M., & Müller-Navarra, D. (1997). The role of highly unsaturated fatty acids in the
378 aquatic foodweb processes. *Freshwater Biology*, 38: 483-499.

379

- 380 Brett, M.T., Kainz, M.J., Taipale, S.J., & Seshan, H. (2009). Phytoplankton, not
381 allochthonous carbon, sustains herbivorous zooplankton production. *Proceedings of the*
382 *National Academy of Sciences of the United States of America*, 106: 21197-21201.
- 383
- 384 Dabrowski, K. R. (1982). Reproductive cycle of vendace (*Coregonus albula* L.) in relation to
385 some chemical and biochemical changes in the body. *Hydrobiologia*, 94: 3-15.
- 386
- 387 Dembiński, W. (1971). Vertical distribution of vendace *Coregonus albula* L. and other
388 pelagic fish species in some Polish lakes. *Journal of Fish Biology*, 3: 341-357.
- 389
- 390 Desvillettes C.H., Bourdier, G., Amblard, C.H., & Barth, B. (1997). Use of fatty acids for the
391 assessment of zooplankton grazing on bacteria, protozoans and microalgae. *Freshwater*
392 *Biology*, 38: 629–637.
- 393
- 394 Galloway, A.W.E., Taipale, S.J., Hiltunen, M., Peltomaa, E., Strandberg, U., Brett, M.T., &
395 Kankaala, P. (2014). Diet-specific biomarkers show that high-quality phytoplankton fuels
396 herbivorous zooplankton in large boreal lakes. *Freshwater Biology*, 59: 1902-1915.
- 397
- 398 Galloway, A.W.E., & Winder, M. (2015). Partitioning the relative importance of phylogeny
399 and environmental conditions on phytoplankton fatty acids. *PloS One*, doi:
400 10.1371/journal.pone.0130053.
- 401
- 402 Gladyshev, M.I., Sushchik, N.N., Anishchenko, O.V., Makhutova, O.N., Kolmakov, V.I.,
403 Kalachova, G.S., Kolmakova, A.A., & Dubovskaya, O.P. (2011). Efficiency of transfer of

- 404 essential polyunsaturated fatty acids versus organic carbon from producers to consumers
405 in a eutrophic reservoir. *Oecologia*, 165: 521-531.
- 406
- 407 Glencross, B.D. (2009). Exploring the nutritional demand for essential fatty acids by
408 aquaculture species. *Reviews in Aquaculture*, 1: 71-124.
- 409
- 410 Hamrin, S.F. (1983). The food preference of vendace (*Coregonus albula*) in South Swedish
411 lakes including the predation effect on zooplankton populations. *Forest Water Ecosystems*,
412 13: 121-128.
- 413
- 414 Helminen, H., Sarvala, J., & Hirvonen, A. 1990. Growth and food consumption of vendace
415 (*Coregonus albula* (L.)) in Lake Pyhäjärvi, SW Finland: a bioenergetics modeling
416 analysis. *Hydrobiologia*, 200/20:, 511-522.
- 417
- 418 Hiltunen, M., Strandberg, U., Keinänen, M., Taipale, S., & Kankaala, P. (2014). Distinctive
419 lipid composition of the copepod *Limnocalanus macrurus* with a high abundance of
420 polyunsaturated fatty acids. *Lipids*, 49: 919-932.
- 421
- 422 Hiltunen, M., Strandberg, U., Taipale, S.J., & Kankaala, P. (2015). Taxonomic identity and
423 phytoplankton diet affect fatty acid composition of zooplankton in large lakes with
424 differing dissolved organic carbon concentration. *Limnology and Oceanography*, 60: 303-
425 317. doi: 10.1002/lno 10028
- 426
- 427 Hixson, S.M., Sharma, B., Kainz, M.J., Wacker, A., & Arts, M.T. (2015). Production,
428 distribution, and abundance of long-chain omega-3 polyunsaturated fatty acids: A

- 429 fundamental dichotomy between freshwater and terrestrial ecosystems. *Environmental*
430 *Reviews*, 23: 414-424.
- 431
- 432 Huusko, A., & Hyvärinen, P. 2005. A high harvest rate induces a tendency to generation
433 cycling in a freshwater fish population. *Journal of Animal Ecology*, 74: 525–531.
- 434
- 435 Kainz, M., Arts, M.T., & Mazumder, A. (2004). Essential fatty acids in the planktonic food
436 web and their ecological role for higher trophic levels. *Limnology and Oceanography*, 49:
437 1784-1793.
- 438
- 439 Kaitaranta, J.K. (1980). Lipids and fatty acids of a whitefish (*Coregonus albula*) flesh and
440 roe. *Journal of the Science of Food and Agriculture*, 31: 1303-1308.
- 441
- 442 Karjalainen, J., Urpanen, U., Keskinen, T. Huuskonen, H., Sarvala, J., Valkeajärvi, P., &
443 Marjomäki, T. (2016). Phenotypic plasticity in growth and fecundity induced by strong
444 population fluctuations affects reproductive traits of female fish. *Ecology and Evolution*,
445 6: 779–790.
- 446
- 447 Lahti, E., & Muje, P. (1991). Egg quality and female condition in vendace (*Coregonus albula*
448 L.) before and during spawning. *Hydrobiologia*, 209: 175-182.
- 449
- 450 Linko, R.R., Rajasilta, M., & Hiltunen, R. (1992). Comparison of lipid and fatty acid
451 composition in vendace (*Coregonus albula* L.) and available plankton feed. *Comparative*
452 *Biochemistry and Physiology A*, 103: 205-212.

- 453 Mairesse, G., Thomas, M., Gardeur, J.-N., & Brun-Bellut, J. (2007). Effects of dietary
454 factors, stocking biomass and domestication on the nutritional and technological quality of
455 the Eurasian perch *Perca fluviatilis*. *Aquaculture*, 262: 86–94.
- 456
- 457 Marjomäki T.J., Keskinen, T., & Karjalainen, J. (2016). The potential ecologically
458 sustainable yield of vendace (*Coregonus albula*) from large Finnish lakes. *Hydrobiologia*,
459 780: 125-134. doi: 10.1007/s10750-016-2783-x
- 460
- 461 Marshall, C.T., Yaragina, N.A., Lambert, Y., & Kjesbu, O.S. 1999. Total lipid energy as a
462 proxy for total egg production by fish stocks. *Nature*, 402: 288-290.
- 463
- 464 Muje, P., Ågren, J.J., Lindqvist, O.V., & Hänninen, O. (1989). Fatty acid composition of
465 vendace (*Coregonus albula* L.) muscle and its plankton feed. *Comparative Biochemistry*
466 *and Physiology B*, 92: 75-79.
- 467
- 468 Müller-Navarra, D.C., Brett, M.T., Liston, A.M., & Goldman, C.R. (2000). A highly
469 unsaturated fatty acid predicts carbon transfer between primary producers and consumers.
470 *Nature*, 403: 74-77.
- 471
- 472 Northcote, T.G., & Hammar, J. (2006). Feeding ecology of *Coregonus albula* and *Osmerus*
473 *eperlanus* in the limnetic waters of Lake Mälaren, Sweden. *Boreal Environment Research*,
474 11: 229-246.
- 475

- 476 Pangle, K.L., Sutton, T.M., Kinnunen, R.E., & Hoff, M.H. (2004). Overwintering survival of
477 juvenile lake herring in relation to body size, physiological condition, energy stores, and
478 food ration. *Transactions of the American Fisheries Society*, 133: 1235-1246.
479
- 480 Pérez, M.T., & Sommaruga, R. (2006). Differential effect of algal- and soil-derived dissolved
481 organic matter on alpine lake bacterial community composition and activity. *Limnology
482 and Oceanography*, 51: 2527-2537.
483
- 484 Perga, M.-E., Bec, A., & Anneville, O. (2009). Origins of carbon sustaining the growth of
485 whitefish *Coregonus lavaretus* early larval stages in Lake Annecy: insights from fatty acid
486 biomarkers. *Journal of Fish Biology*, 74: 2-17.
487
- 488 Persson, J., & Vrede, T. (2006). Polyunsaturated fatty acids in zooplankton: variation due to
489 taxonomy and trophic position. *Freshwater Biology*, 51: 887–900.
490
- 491 Pickova, J., Kiessling, A., Pettersson, A., & Dutta, P.C. (1999). Fatty acid and carotenoid
492 composition of eggs from two nonanadromous Atlantic salmon stocks of cultured and wild
493 origin. *Fish Physiology and Biochemistry*, 21: 147–156.
494
- 495 Rask, M., Viljanen, M., & Sarvala, J. (1999). Humic lakes as fish habitat. In: Keskitalo, J. &
496 Eloranta, P. (Eds.) *Limnology of humic waters* (pp. 209-224). Backhuys Publishers,
497 Leiden, the Netherlands.
498
- 499 Sargent, J., Bell, G., McEvoy, L., Tocher, D., & Estevez, A. (1999). Recent developments in
500 the essential fatty acid nutrition of fish. *Aquaculture*, 177: 191-199.

501

502 Simon, M., Cho, B.C., & Azam, F. (1992). Significance of bacterial biomass in lakes and the
503 ocean: comparison to phytoplankton biomass and biogeochemical implications. *Marine*
504 *Ecology Progress Series*, 86: 103-110.

505

506 Smyntek, P.M., Teece, M.A., Schulz, K.L., & Storch, A.J. (2008). Taxonomic differences in
507 the essential fatty acid composition of groups of freshwater zooplankton relate to
508 reproductive demands and generation time. *Freshwater Biology*, 53: 1768-1782.

509

510 Strandberg, U., Hiltunen, M., Jelkänen, E., Taipale, S.J., Kainz, M.J., Brett, M.T., &
511 Kankaala, P. (2015a). Selective transfer of polyunsaturated fatty acids from phytoplankton
512 to planktivorous fish in large boreal lakes. *Science of the Total Environment*, 536: 858-
513 865. doi: 10.1016/j.scitotenv.2015.07.010

514

515 Strandberg, U., Taipale, S.J., Hiltunen, M., Galloway, A.W.E., Brett, M.T., & Kankaala, P.
516 (2015b). Inferring phytoplankton community composition with a fatty acid mixing model.
517 *Ecosphere*, 61. doi: 10.1890/ES14-00382.1

518

519 Taipale, S.J., Kankaala P., Hämäläinen H., & Jones R.I. (2009). Seasonal shifts in the diet of
520 lake zooplankton revealed by phospholipid fatty acid analysis. *Freshwater Biology*, 54:
521 90-104.

522

523 Taipale, S.J., Strandberg, U., Peltomaa, E., Galloway, A.W.E., Ojala, A., & Brett, M.T.
524 (2013). Fatty acid composition as biomarkers of freshwater microalgae: analysis of 37

- 525 strains of microalgae in 22 genera and in 7 classes. *Aquatic Microbial Ecology*, 71: 165-
526 178.
- 527
- 528 Taipale, S.J., Brett, M. T., Hahn, M. W., Martin-Creuzburg, D., Yeung, S., Hiltunen, M.,
529 Strandberg, U., & Kankaala, P. (2014). Differing *Daphnia magna* assimilation efficiencies
530 for terrestrial, bacterial, and algal carbon and fatty acids. *Ecology*, 95: 563-576.
- 531
- 532 Taipale, S.J., Kainz, M.J., & Brett, M.T. (2015). A low ω -3: ω -6 ratio in *Daphnia* indicates
533 terrestrial resource utilization and poor nutritional condition. *Journal of Plankton*
534 *Research*, 37: 596–610.
- 535
- 536 Taipale, S.J., Vuorio, K., Strandberg, U., Kahilainen, K.K., Järvinen, M., Hiltunen, M.,
537 Peltomaa, E., & Kankaala, P. (2016). Lake eutrophication and brownification downgrade
538 availability and transfer of essential fatty acids for human consumption. *Environment*
539 *International*, 96: 156-166. doi: 10.1016/j.envint.2016.08.018
- 540
- 541 Tocher, D.R. (2003). Metabolism and functions of lipids and fatty acids in teleost fish.
542 *Reviews in Fisheries Science* 11: 107-184. *Journal of Plankton Research*, 37: 596–610.
- 543
- 544 Viljanen, M. (1983). Food and food selection of cisco (*Coregonus albula* L.) in a dystrophic
545 lake. *Hydrobiologia*, 101: 129-138.
- 546
- 547 Viljanen, M. (1986). Biology, propagation, exploitation and management of vendace
548 (*Coregonus albula* L.) in Finland. *Archiv für Hydrobiologie – Beiheft Ergebnisse der*
549 *Limnologie*, 22: 73-97.

550

551 Watanabe, T., Thongrod, S., Takeuchi, T., Satoh, S., Kubota, S.S., Fujimaki, Y. Cho, C.Y.,
552 (1989). Effect of dietary n-6 and n-3 fatty acids on growth, fatty acid composition and
553 histological changes of white fish *Coregonus lavaretus maraena*. *Nippon Suisan*
554 *Gakkaishi* 55: 1977-1982.

555

556 Ventelä, A.-M., Kirkkala, T., Lendasse, M., Tarvainen, M., Helminen, H. & Sarvala, J.,
557 (2011). Climate-related challenges in long-term management of Säkylän Pyhäjärvi (SW
558 Finland). *Hydrobiologia*, 660: 49-58.

559

560 Winfield, I.J., Fletcher, J.M., & James, B.J. (2004). Conservation ecology of the vendace
561 (*Coregonus albula*) in Bassenthwaite Lake and Deerwent Water, U.K. *Annales Zoologici*
562 *Fennici*, 41:155-164.

563 Table 1. Location, mean depth and total lake area of the studied lakes as well as mean (\pm SD) water color, Secchi depth, and concentrations of
 564 DOC, total nitrogen, total phosphorus, and chlorophyll *a* at the sampling sites in six lake basins in the eastern Finland. The values for color are
 565 from 1 m depth and the others pooled for 0-4 m depth sampled in May-June, early August and late September 2011, with the exception of Lake
 566 Karjalan Pyhäjärvi, which was sampled in August 2012.

567

Lake basin	Coordinates	Mean depth (m)	Surface area (km ²)	Colour (mg Pt L ⁻¹)	Secchi depth (m)	DOC (mg L ⁻¹)	totN (μg L ⁻¹)	totP (μg L ⁻¹)	Chl <i>a</i> (μg L ⁻¹)
Karjalan Pyhäjärvi	61°48'N; 29°52'E	8.8	247	15	5.1±0.3	5.3±0.5	228±20	4.9±0.9	2.2±0.3
Orivesi	62°10'N; 29°43'E	9	601	28±10	2.1±0.3	7.0±0.3	304±32	11.8±1.0	6.3±2.6
Paasivesi	62° 8'N; 29°26'E	21	124	43±6	2.5±0.5	8.4±0.4	349±27	5.7±3.2	2.0±1.1
Suvasvesi	62°40'N; 28°12'E	10.4	233	32±3	3.1±0.5	8.8±0.4	490±42	10.0±7.3	2.8±1.0
Pyhäselkä	62°28'N; 29°48'E	8.8	361	57±6	1.9±0.0	9.5±0.5	399±48	9.3±0.6	1.5±0.8
Kallavesi	62°49'N; 27°52'E	8.6	478	45±5	2.3±0.6	10.2±0.2	639±107	12.1±7.1	3.4±1.3

568

569 Table 2. Length, weight and fatty acid content of vendace sampled from five large boreal
 570 lakes in spring (n=15), summer (n=18) and autumn (n=15). Values are mean (\pm SD).
 571 Approximate lipid content (Lipid %) in the muscle tissue was calculated assuming that fatty
 572 acids comprise 70 % of all lipids in fish muscle (Ahlgren et al. 1996). The fatty acid content
 573 of the muscle tissue and the weight of vendace as well as condition index (Fulton's K) were
 574 significantly lower in spring than in summer or autumn (marked with +; Wilcoxon signed
 575 rank test, $P < 0.05$). The length of vendace did not differ between seasons.

	Length (mm)	Weight (g)	FA content (mg g ⁻¹ DW)	Lipid %	Condition index (K)
Spring	139 \pm 25	17.2 \pm 9.6 ⁺	17.7 \pm 4.3 ⁺	2.5	0.58 \pm 0.07 ⁺
Summer	148 \pm 13	23.0 \pm 6.1	28.1 \pm 13.1	4.0	0.69 \pm 0.07
Autumn	146 \pm 19	23.6 \pm 11.0	25.7 \pm 6.1	3.7	0.70 \pm 0.07

576

577 Table 3. Average (\pm SD) proportion (%) of saturated fatty acids (SFA), monounsaturated
 578 fatty acids (MUFA) and polyunsaturated fatty acids (separately for C₁₈, C₂₀ and C₂₂ PUFA) in
 579 seston (0.22 – 50 μ m) PLFA (from Strandberg et al., 2015b), and total fatty acids of major
 580 zooplankton taxa (from Hiltunen et al., 2015) and in vendace during spring, summer and
 581 autumn in five lakes (names in Table 1) in Eastern Finland (n = 21 – 48 in each). Different
 582 letters (a-c) denote significant between-season difference in the relative proportion of the
 583 specific FA class within each group/taxa (ANOVA, Games-Howell post hoc test).

FA class	Group/taxa	Spring	Summer	Autumn
SFA	Seston	26.6 \pm 4.0 ^a	36.9 \pm 6.8 ^b	29.7 \pm 9.1 ^{ab}
	Cladocera	36.8 \pm 3.9 ^a	36.5 \pm 4.9 ^a	30.3 \pm 2.0 ^b
	<i>Limnocalanus</i>	30.2 \pm 4.9 ^a	12.1 \pm 3.2 ^b	14.0 \pm 3.3 ^b
	<i>Eudiaptomus</i>	31.3 \pm 4.7 ^a	40.4 \pm 6.1 ^b	28.1 \pm 1.6 ^a
	Cyclopoida	35.6 \pm 5.4 ^a	45.3 \pm 7.1 ^b	29.8 \pm 8.2 ^a
	Vendace	33.6 \pm 3.8	33.0 \pm 2.6	32.6 \pm 2.3
MUFA	Seston	16.4 \pm 6.3	19.0 \pm 2.2	20.8 \pm 3.9
	Cladocera	24.2 \pm 6.3	24.0 \pm 4.2	27.6 \pm 4.8
	<i>Limnocalanus</i>	12.9 \pm 3.7	15.0 \pm 3.0	15.9 \pm 2.0
	<i>Eudiaptomus</i>	13.9 \pm 3.0 ^a	7.6 \pm 0.6 ^b	9.5 \pm 1.4 ^c
	Cyclopoida	13.8 \pm 3.2	10.6 \pm 4.2	14.0 \pm 6.6
	Vendace	7.2 \pm 2.2 ^a	12.2 \pm 4.0 ^b	10.8 \pm 2.8 ^b
C ₁₈ PUFA	Seston	29.5 \pm 7.0	30.5 \pm 5.1	31.4 \pm 5.4
	Cladocera	18.8 \pm 3.6	19.0 \pm 3.6	17.7 \pm 5.1
	<i>Limnocalanus</i>	7.5 \pm 1.6 ^a	14.2 \pm 2.9 ^b	17.1 \pm 2.1 ^c
	<i>Eudiaptomus</i>	16.6 \pm 4.3 ^a	16.2 \pm 4.2 ^a	27.9 \pm 3.8 ^b
	Cyclopoida	16.5 \pm 6.0 ^a	15.7 \pm 3.1 ^a	23.0 \pm 9.5 ^b
	Vendace	2.9 \pm 2.4 ^a	7.2 \pm 3.5 ^b	5.8 \pm 2.0 ^b
C ₂₀ PUFA	Seston	16.1 \pm 2.5 ^a	7.1 \pm 3.9 ^b	10.6 \pm 2.4 ^b
	Cladocera	15.7 \pm 5.6	16.9 \pm 6.1	19.6 \pm 2.1
	<i>Limnocalanus</i>	19.1 \pm 4.6 ^a	25.3 \pm 2.1 ^b	23.6 \pm 1.4 ^b
	<i>Eudiaptomus</i>	13.2 \pm 2.2	13.0 \pm 2.3	13.6 \pm 1.0
	Cyclopoida	13.1 \pm 2.9 ^a	9.5 \pm 2.7 ^b	13.2 \pm 2.7 ^a
	Vendace	13.4 \pm 1.7 ^a	15.1 \pm 1.6 ^b	15.5 \pm 1.7 ^b
C ₂₂ PUFA	Seston	9.5 \pm 3.6 ^a	2.4 \pm 2.7 ^b	2.8 \pm 1.7 ^b
	Cladocera	0.7 \pm 0.4	0.5 \pm 0.6	0.4 \pm 0.7
	<i>Limnocalanus</i>	28.5 \pm 3.3 ^a	23.2 \pm 3.4 ^b	23.2 \pm 2.8 ^b
	<i>Eudiaptomus</i>	19.5 \pm 4.6 ^a	19.5 \pm 2.2 ^a	13.7 \pm 2.1 ^b

	Cyclopoida	16.9 ± 3.6	15.7 ± 2.6	14.8 ± 3.0
	Vendace	42.4 ± 4.1 ^a	31.3 ± 6.8 ^b	34.5 ± 4.2 ^b
BAFA	Seston	1.5 ± 0.9	2.2 ± 1.1	2.3 ± 0.8
	Cladocera	4.2 ± 1.8	4.6 ± 1.1	5.0 ± 0.3
	<i>Limnocalanus</i>	3.4 ± 1.8 ^a	1.2 ± 0.7 ^b	1.2 ± 0.3 ^b
	<i>Eudiaptomus</i>	5.0 ± 1.6 ^a	3.4 ± 0.7 ^b	4.3 ± 0.5 ^a
	Cyclopoida	5.4 ± 2.6	5.4 ± 1.3	4.8 ± 1.8
	Vendace	0.9 ± 0.4	1.5 ± 0.9	1.0 ± 0.3

584

585

586 **FIGURE LEGENDS**

587

588 Figure 1. Mean content of saturated fatty acids (SFA), monounsaturated fatty acids (MUFA),
589 C₁₈, C₂₀ and C₂₂ polyunsaturated fatty acids (PUFA) and bacterial fatty acids (BAFA) during
590 different seasons in the muscle of vendace from different lake basins (n=3-5 in each lake and
591 season). Error bars represent SD of the total FA content.

592

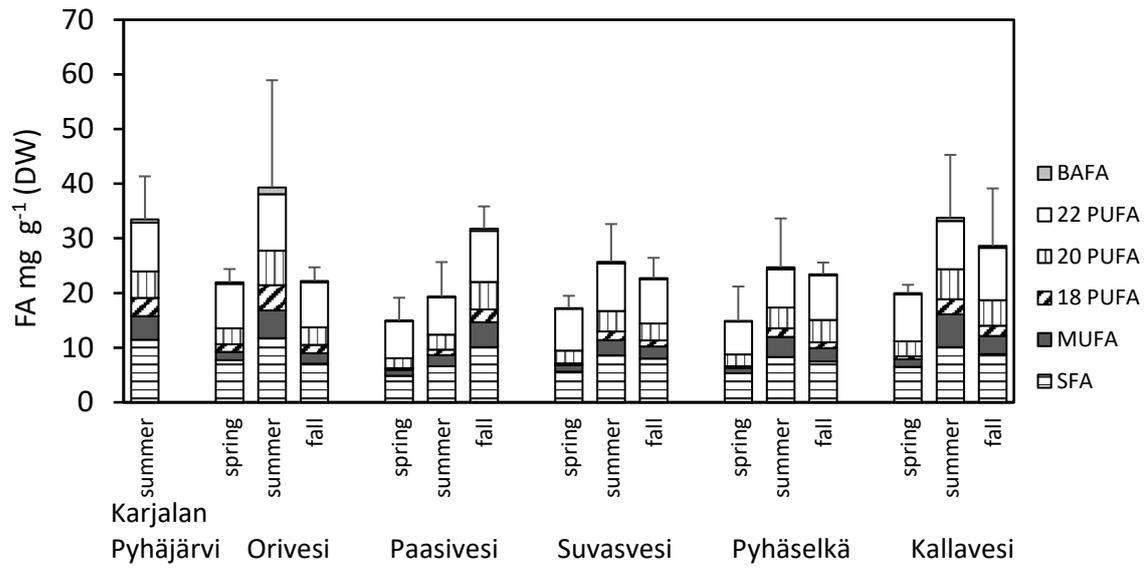
593 Figure 2. Multidimensional scaling ordination of fatty acid proportions (42 fatty acids) in the
594 dorsal muscle of vendace in different lake basins and seasons. Euclidean distance was used
595 for the scaling (2D stress: 0.05). Seasonal differences of the fatty acid composition of
596 vendace is statistically significant (see text), specifically the spring samples differ from
597 summer and autumn samples (results of the PERMANOVA post hoc test in the figure).

598

599 Figure 3. Correlation (with 95 % confidence intervals) of the total fatty acid content of
600 vendace muscle and the proportion of A) MUFA (mainly C₁₆ and C₁₈), B) C₁₈ PUFA, C)
601 C₂₂PUFA, and D) BAFA. Note the different scales on the y-axis.

602

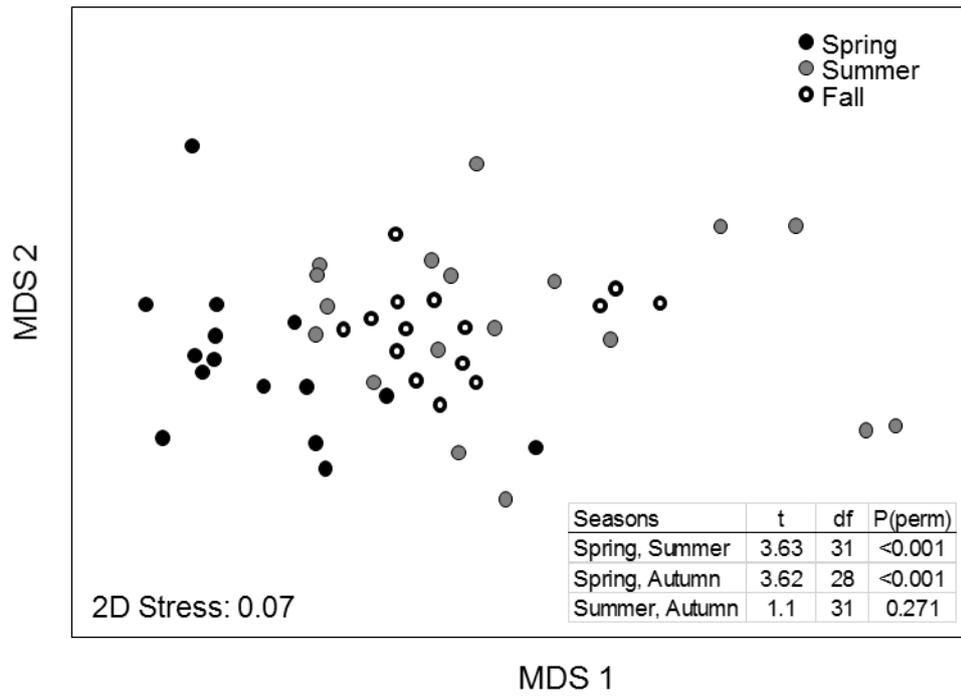
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604

605

606 Figure 1.



607

608 Figure 2.

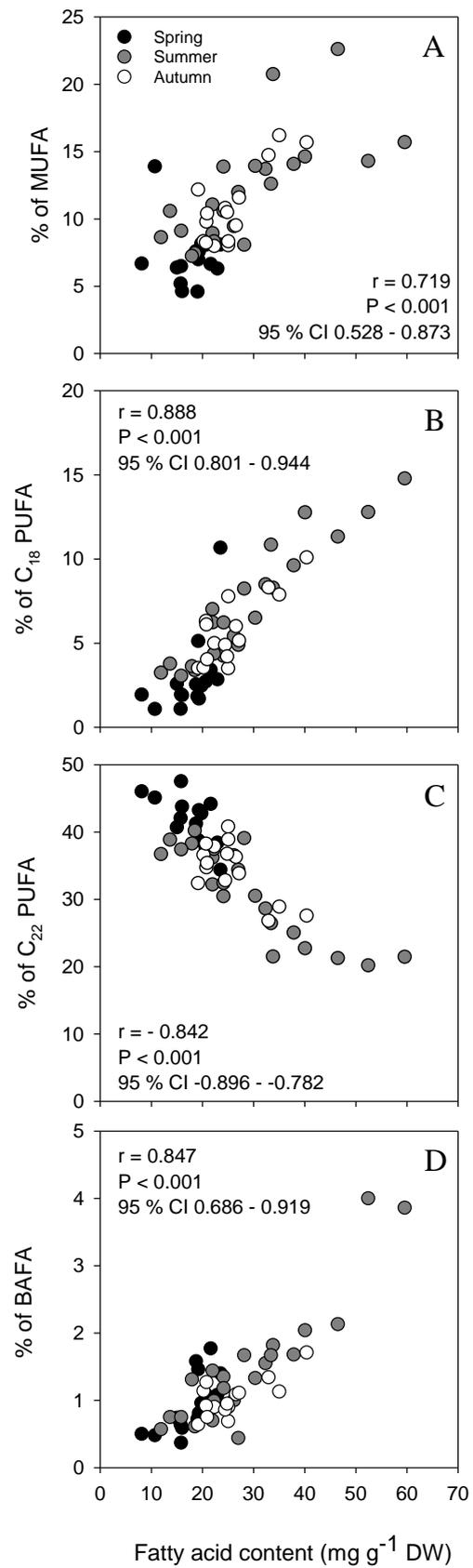


Figure 3.

Supplement 1. Fatty acid profile of vendace muscle (% of all fatty acids). Samples from 5 lake basins were combined for each season: spring (n=15), summer (n=18), fall (n=15), because no significant differences were found in the fatty acid profile between lakes (PERMANOVA, $P=0.178$). See the text for details. Samples from Lake Karjalan Pyhäjärvi are presented separately due to lack of samples from spring and fall (n=4).

	Spring		Summer		Fall		Karjalan Pyhäjärvi	
	Mean	<i>SD</i>	Mean	<i>SD</i>	Mean	<i>SD</i>	Mean	<i>SD</i>
14:0	1.0	0.5	2.0	1.3	1.7	0.8	3.6	1.0
4.8.12-TMTD	0.1	0.1	0.1	0.1	0.1	0.1	0.2	0.1
i15:0	0.0	0.0	0.2	0.2	0.1	0.1	0.3	0.1
ai15:0	0.0	0.0	0.1	0.1	0.0	0.0	0.1	0.1
15:0	0.3	0.2	0.3	0.2	0.3	0.1	0.3	0.1
i16:0	0.0	0.0	0.0	0.1	0.0	0.1	0.1	0.0
16:0	24.8	3.3	23.6	3.3	24.6	2.2	25.6	2.0
16:1n-9	0.1	0.0	0.2	0.1	0.1	0.1	0.4	0.1
16:1n-7	0.7	0.4	2.4	1.4	1.9	0.7	2.4	1.0
16:1n-5	0.2	0.1	0.3	0.1	0.2	0.1	0.4	0.1
i17:0	0.1	0.1	0.2	0.1	0.1	0.0	0.2	0.1
ai17:0	0.1	0.1	0.2	0.2	0.1	0.0	0.3	0.1
16:2n-4	0.0	0.0	0.1	0.1	0.0	0.0	0.0	0.0
17:0	0.4	0.2	0.4	0.1	0.4	0.1	0.4	0.1
Phytanate	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0
17:1	0.0	0.0	0.1	0.1	0.0	0.0	0.0	0.0
18:0	7.0	0.9	6.2	1.2	5.7	0.4	4.6	0.5
18:1n-9	4.7	1.6	6.9	2.5	6.4	1.9	6.7	1.4
18:1n-7	1.6	0.4	2.3	0.6	2.1	0.4	2.6	0.4
18:2n-6	1.1	0.7	2.0	1.0	1.7	0.6	2.2	0.7
18:3n-6	0.0	0.0	0.1	0.1	0.0	0.0	0.1	0.1
18:3n-3	1.3	0.8	2.8	1.1	2.5	0.9	3.0	1.1
18:4n-3	0.5	0.9	2.0	1.5	1.5	0.7	4.0	1.7
20:0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0
20:1n-9	0.0	0.0	0.1	0.1	0.0	0.1	0.0	0.0
20:2n-6	0.0	0.0	0.1	0.1	0.0	0.1	0.0	0.0
20:4n-6	5.0	1.0	4.5	0.7	4.9	1.1	4.5	0.3
20:3n-3	0.0	0.0	0.1	0.1	0.0	0.1	0.2	0.0
20:4n-3	0.3	0.3	0.7	0.5	0.6	0.3	1.0	0.1
20:5n-3	8.1	1.3	9.9	1.3	10.0	1.3	8.7	0.7
22:4n-6	0.0	0.0	0.1	0.1	0.0	0.0	0.0	0.0
22:5n-6	1.8	0.65	1.8	0.6	1.5	0.5	1.5	0.3
22:5n-3	0.9	0.4	1.3	0.4	1.0	0.3	1.3	0.1
22:6n-3	39.7	4.3	28.9	7.1	32.0	4.1	25.2	6.3
24:1n-9	0.2	0.2	0.1	0.1	0.1	0.1	0.0	0.0

Supplement 2. Fatty acid composition of vendace roe (n=7). Only the quantitatively most important fatty acids are presented (mean proportion > 1%). These 14 fatty acids account for 92.0-95.6% of all fatty acids in the roe. Values are mean (\pm SD).

Fatty acid	w%	Fatty acid	w%	Fatty acid	w%
14:0	2.81 (0.49)	18:2n-6	3.81 (1.10)	22:5n-3	1.94 (0.13)
16:0	17.85 (1.63)	18:3n-3	5.53 (1.05)	22:6n-3	16.31 (3.17)
18:0	2.01 (0.27)	18:4n-3	4.34 (1.01)	n3/n6	4.04 (0.69)
16:1n-7	5.27 (1.12)	20:4n-6	4.85 (0.70)	sumSFA	22.66 (1.49)
18:1n-9	13.22 (3.18)	20:4n-3	1.97 (0.45)	sumMUFA	22.36 (4.35)
18:1n-7	3.87 (0.47)	20:5n-3	9.89 (0.71)	sumPUFA	48.65 (4.07)