Lipid-rich zooplankton subsidise the winter diet of benthivorous Arctic charr (*Salvelinus alpinus*) in a subarctic lake

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SUMMARY

1. Generalist fish species commonly act as important links between littoral and pelagic habitats and food-web compartments in lakes. However, diet and habitat links may depend significantly on seasonal availability of, and qualitative differences between, littoral and pelagic prey and on fish size. Despite increasing interest in food-web dynamics, little is known about the seasonal changes in, or qualitative differences between, littoral and pelagic trophic pathways supporting generalist fish species in high-latitude lakes.

2. We used stomach contents together with analyses of stable carbon and nitrogen isotopes and fatty acids to study the winter and summer diet of generalist Arctic charr and determine the qualitative differences between littoral and pelagic prey items. We were particularly interested to determine whether Arctic charr are able to utilise abundant and lipid-rich winter zooplankton resources in subarctic Lake Saanajärvi, northern Finland.

3. Arctic charr fed actively on cladoceran zooplankton in both seasons, despite the higher abundance and higher lipid content of calanoid copepods. Although the stomach contents consisted mainly of zooplankton in summer, the isotopic compositions of muscle and liver suggest Arctic charr relied more on littoral carbon sources throughout the year. Fatty acid analysis indicated that Arctic charr had lower amounts of body fat and total and essential fatty acids in winter compared with summer.

4. Observed seasonal feeding activity and dietary shifts were partly related to Arctic charr size. Small (<200 mm) Arctic charr had more empty stomachs in winter, but higher amounts of zooplankton in stomachs and of essential fatty acids in muscle tissue in summer compared with larger (>200 mm) conspecifics that had more seasonally stable feeding activity and diet.

5. Fatty acid analysis indicated that both littoral and pelagic food sources provided similar fatty acids to Arctic charr, but in general, zooplankton had higher percentages of essential fatty acids compared with zoobenthos. Pelagic *Eudiaptomus graciloides* calanoids and littoral *Gammarus lacustris* amphipods had the highest concentrations of total and essential fatty acids, but only the latter prey item was found in Arctic charr stomach contents.

6. Our study demonstrates that lipid-rich zooplankton can subsidise the predominantly benthivorous diet of top consumers (here Arctic charr) in subarctic lakes. The results also demonstrate that littoral and pelagic trophic pathways can be highly integrated in high-latitude lakes, as a result of the flexible foraging behaviour of top consumers such as Arctic charr.

Keywords: energy mobilisation, fatty acids, food-web coupling, generalist foraging, seasonality

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Introduction

Several food-web studies have demonstrated that littoral and pelagic trophic pathways in lakes are highly integrated (Schindler & Scheuerell, 2002; Vadeboncoeur, Vander Zanden & Lodge, 2002). The argument is supported by the fact that many consumers, from benthic and pelagic invertebrates up to top consumers, often show generalist foraging across lake habitat boundaries (Solomon *et al.*, 2011; Thompson, Dunne & Woodward, 2012). For example, recent evidence from laboratory and field studies shows that littoral benthic algae can subsidise the predominantly phytoplankton-based diet of zooplankton (Rautio & Vincent, 2006; Mariash *et al.*, 2011; Cazzanelli *et al.*, 2012), whereas many benthic invertebrates (particularly mussels and profundal zoobenthos) consume largely pelagic carbon sources (Premke *et al.*, 2010). However, the most apparent couplers of littoral and pelagic food-web compartments in lakes are generalist fish species that can undergo rapid changes in their feeding behaviour and habitat use (Vander Zanden & Vadeboncoeur, 2002; Rooney *et al.*, 2006). Generalist foraging by fish has been shown to be particularly evident in high-latitude lakes where consumers must adapt to seasonal changes in prey availability, light and temperature (Hecky & Hesslein, 1995; Power, Reist & Dempson, 2008). Generalist fish can also play a major role in carbon and nutrient cycling in lakes, for example by feeding on benthic prey but providing nutrients to pelagic phytoplankton in dissolved format (Vanni, 2002; Glaholt & Vanni, 2005). From a whole community perspective, generalist foraging of fish may increase food-web stability by decreasing consumer-resource oscillations (Vadeboncoeur *et al.*, 2005; Rooney *et al.*, 2006; Kratina *et al.*, 2012). Although generalist fish are increasingly used as model organisms in studies of energy flow and trophic cascades in freshwater food webs (e.g. Jeppesen *et al.*, 2003; Vander Zanden, Vadeboncoeur & Chandra, 2011), little is known about the seasonal shifts in, and qualitative differences between, littoral and pelagic trophic pathways supporting top consumers in high-latitude lakes.

Winter is a critical period for the survival of benthic and pelagic organisms in high-latitude lakes (George & Hewitt, 1999; Vincent, Hobbie & Laybourn-Parry, 2008; Karlsson & Säwström, 2009). The limited primary production and the long, dark and cold ice-cover period demand special adaptations for overwintering invertebrate and fish consumers. Some zooplankton can form resting eggs, while some may stay active by storing lipids (Lee, Hagen & Kattner, 2006; Mariash, 2012) and/or by substituting the reduced phytoplankton resources by feeding on benthic algae (Karlsson & Säwström, 2009; Mariash *et al.*, 2011; Cazzanelli *et al.*, 2012), or on heterotrophic bacteria or nanoflagellates that derive energy from allochthonous carbon via the microbial loop (Ask *et al.*, 2009; Rautio, Mariash & Forsström, 2011). In some high-latitude lakes, some zooplankton species have been observed to reach a maximum density peak in winter (Rautio, Sorvari & Korhola, 2000) and to develop high lipid stores for overwintering and reproduction (Snow, 1972; Mariash, 2012). These lipid-rich zooplankton resources may provide an important food source for generalist fish such as Arctic charr (*Salvelinus alpinus*).

While benthic invertebrates are typically larger and may dominate the energy supply to top consumers in high-latitude lakes, pelagic zooplankton may support the physiological requirements of overwintering fish by providing higher concentrations of essential fatty acids (Smyntek *et al.*, 2008; Ravet, Brett & Arhonditsis, 2010). The physiological requirements and feeding behaviour of generalist fish are highly dependent on individual ontogenetic stage, because size and maturity affect foraging efficiency, predation risk, competitive dominance and basic metabolic rate (Werner & Gilliam, 1984; Byström *et al.*, 2006; Persson & De Roos, 2007). Hence, to understand trophic dynamics and energy mobilisation in high-latitude lakes, it is important to pay particular attention to the effect of top consumer size, and the potential for seasonal changes in the quantity and the quality of littoral and pelagic food resources.

Although stable isotope studies suggest that top consumers like Arctic charr rely heavily on littoral carbon sources in high-latitude lakes (Karlsson & Byström, 2005; Eloranta, Kahilainen & Jones, 2010), lipid-rich zooplankton may supplement the limited littoral food resources in winter. For example, previous studies from subarctic lakes have shown that zooplankton can be an important dietary item for Arctic charr in autumn/early winter, particularly for small fish (Klemetsen *et al.*, 2003; Amundsen, Knudsen & Klemetsen, 2008; Amundsen & Knudsen, 2009). Nevertheless, the extent to which littoral and pelagic trophic pathways support top consumers in high-latitude lakes may largely depend on seasonal food availability and on qualitative differences between littoral and pelagic prey items as sources of physiologically important fatty acids.

Stable isotope analysis (SIA) has been widely used to study the reliance of freshwater invertebrate and fish consumers on littoral and pelagic carbon (e.g. Hecky & Hesslein, 1995; Sierszen, McDonald & Jensen, 2003; Vander Zanden *et al.*, 2011). While stomach contents analysis

(SCA) provides information about recent foraging with high taxonomic resolution, SIA reflects the main long-term carbon sources assimilated into body tissues (McCarthy *et al.*, 2004; Davis *et al.*, 2012). In turn, while SIA can be used as an effective tool to study the trophic pathways supporting top consumers, the method does not reflect qualitative differences between littoral and pelagic food sources. To compensate for the shortcomings of SCA and SIA, fatty acid analysis (FAA) can be used to quantify the physiological status of top consumers and the qualitative differences between consumed littoral and pelagic prey items (Brett *et al.*, 2009; Iverson, 2009).

To supplement the limited knowledge of winter trophic pathways in high-latitude lakes, we studied the winter and summer diet of Arctic charr in the Finnish subarctic Lake Saanajärvi, where zooplankton have been observed to develop high lipid concentrations and to occur in relatively high abundances during winter (Rautio *et al.*, 2000; Syväranta & Rautio, 2010). We used stomach contents and stable carbon and nitrogen isotope analyses to investigate the recent and long-term reliance of Arctic charr on littoral and pelagic food resources. We also analysed the fatty acid compositions of the fish and their putative food sources to investigate seasonal and size-related differences in fish physiological status and the quality of littoral and pelagic prey items. Previous studies from Saanajärvi (Eloranta *et al.*, 2010) and other Arctic charr lakes (L'Abée-Lund *et al.*, 1993; Riget *et al.*, 2000; Amundsen & Knudsen, 2009) have indicated that Arctic charr longer than 200 mm are not typically preyed upon by cannibalistic conspecifics or other piscivorous fish and therefore may undergo significant seasonal habitat and dietary shifts. In contrast, fish smaller than 200 mm typically avoid predation by remaining near benthic areas throughout the year. Hence, we compared the SCA, SIA and FAA data for Arctic charr size groups of total length <200 mm and >200 mm. We focussed on three main study questions: (i) Do Arctic charr feed on lipid-rich zooplankton in winter? (ii) Do the diet and fatty acids composition of Arctic charr depend on fish size? and (iii) Do littoral and pelagic food sources have different fatty acid compositions indicative of potential qualitative differences between the main pelagic and littoral trophic pathways?

Methods

Study area

Lake Saanajärvi (69°05′N; 20°87′E) is a small (0.7 km^2), dimictic, ultraoligotrophic lake with a small catchment area (460 ha) and a maximum depth of 24 m, situated above the treeline (679 m a.s.l) in the subarctic Kilpisjärvi region of north-western Finland (see Fig. S1 in Supporting Information). The littoral and pelagic zones in Saanajärvi extend down to around 15 m [i.e. the compensation depth to which 1% of photosynthetically available surface radiation penetrates; measured using LI-1400 light meter (wavelength 400–700 nm; Li-Cor Inc., Lincoln, NE, U.S.A.)]. The lake is typically ice-free between late June and mid-October, and the maximum surface water temperature (13–15 °C) is usually reached in August, while the water temperature is 0–4 °C under ice (Rautio *et al.*, 2000; Forsström *et al.*, 2005). Saanajärvi contains a monomorphic population of Arctic charr that were introduced to the lake in 1993 and low numbers of brown trout (*Salmo trutta*) of unknown origin. *Eudiaptomus graciloides*, *Cyclops abyssorum* and *Daphnia umbra* are present in the water column throughout the year, although *E. graciloides* make up more than 40% of the zooplankton community under the ice in Saanajärvi (Rautio *et al.*, 2000, 2011). In summer, the zooplankton community is more diverse, with both copepod and several cladoceran species (*D. umbra*, *Bosmina* sp., *Holopedium gibberum* and *Polyphemus pediculus*). The cladoceran zooplankton (*D. umbra* and *H. gibberum*) density peaks in late spring and/or late summer (Rautio *et al.*, 2000, 2011).

Sampling and sample preparation

Arctic charr were caught between 29 January and 9 February and between 18 and 20 August 2011 from Saanajärvi using seasonally dependent sampling protocols. In winter, 1.8 × 30 m gillnets with 30–35-mm (knot to knot) mesh were pulled under ice on the lake bottom at depths of 5–10 m. Logistical considerations prevented attempts to set pelagic or profundal zone nets, but Arctic charr are known to prefer littoral habitat in winter (Klemetsen *et al.*, 2003; Amundsen & Knudsen, 2009). A more comprehensive sampling procedure was employed in August when three sets of gillnets (12–60-mm mesh) were set in the littoral and profundal zones (benthic gill nets; 0–10 m and 15–24 m, respectively) and in the uppermost pelagic zone (floating gill nets; set to the surface above 15–24 m depth) of the lake. However, in August, most Arctic charr were caught from the littoral zone (88%), while only 4% and 8% were caught from the pelagic and profundal zones, respectively. The total gillnet catch from Saanajärvi included 33 Arctic charr in winter, whereas a total of 37 Arctic charr were randomly chosen in summer to balance summer and winter

sample sizes. Although different seasonal sampling strategies were used, mean total length of subsampled Arctic charr (range from 109 to 503 mm) did not differ significantly between seasons ($U = 534$, $n = 70$, $P = 0.37$). Arctic charr caught from the pelagic zone in August were slightly larger (mean ± SD total length: 191 ± 77 mm) than the littoral-caught conspecifics (161 ± 61 mm; $U = 1634$, $n = 496$, $P = 0.03$).

Nets were checked daily after 21–27 h soaking time, and fish were removed, euthanised and transported on ice to the laboratory, where total length (mm) and mass (g) were recorded for each fish. Stomachs were removed for immediate SCA. Finally, an approximate 20×20 mm piece of fresh dorsal muscle tissue was dissected from each fish, frozen at -20 °C and freeze-dried for stable carbon and nitrogen isotope (SIA) and fatty acid (FAA) analyses that reflect long-term food source assimilation and physiological status of individual fish, respectively. In addition, an approximate 10×10 mm piece of fresh liver was dissected from each fish for SIA, because liver has a more rapid turn-over rate and thus reflects the diet over a shorter period than the muscle tissue (Perga & Gerdeaux, 2005). Dried muscle samples for FAA were immediately stored at -80 °C, while the samples for SIA were stored at -20 °C.

Removed stomachs were opened, and the total fullness was visually determined on a percentage scale ranging from empty (0%) to completely full (100%). Prey items were identified to species, genus or family, and their contribution to total stomach fullness was estimated following Amundsen, Gabler & Staldvik (1996). In the final presentation of SCA data, the observed 16 prey taxa were grouped into: (1) cladoceran zooplankton (*D. umbra*, *Bosmina* sp.), (2) predatory cladocerans (*Bythotrephes longimanus*, *P. pediculus*), (3) copepods (*C. abyssorum*), (4) *Gammarus lacustris* amphipods, (5) chironomid larvae, (6) other benthos (*Pisidium* sp. mussels, Valvatidae and *Lymnaea* sp. snails, trichopteran larvae, plecopteran nymphs, unidentified insects), (7) pleuston (chironomid pupae, adult terrestrial insects) and (8) fish (Arctic charr). The dietary similarity between all Arctic charr caught in winter and summer and between small (<200 mm) and large (>200 mm) Arctic charr caught in each season was tested with a nested analysis of similarities (ANOSIM) using PAST 2.17 program (Hammer, Harper & Ryan, 2001). Similarity percentages (SIMPER) were also calculated in PAST to determine whether a particular prey taxon was most important in determining the potential seasonal or size-related differences in Arctic charr diet. In addition, the dietary niche width of all winter and summer Arctic

charr and of small and large individuals was calculated using Levins' (1968) index (*B*).

Qualitative samples of littoral (depth <5 m) benthic macroinvertebrates (hereafter zoobenthos) were collected in August 2011 for SIA and FAA using an Ekman grab in deep (1–5 m) and a kick-net or hand-picking in shallow (0–1 m) sites. Ekman and kick-net samples were sieved through a 500-μm mesh, and zoobenthos were separated into taxonomic groups. Zooplankton were collected in January/February and July/August 2008 and 2011 with replicate vertical hauls from 20 m depth to surface using a 50 μm net. Samples were later sieved through 100-μm and 200-μm meshes to retain only large zooplankton. After separation, both zoobenthos and zooplankton were left in cool tap water for a few hours for animals to void their guts. Finally, the samples were frozen at -20 °C, freeze-dried and stored at -20 °C (SIA) or at -80 °C (FAA) for later preparation.

All freeze-dried fish muscle and liver tissues, zoobenthos (only soft body tissue from molluscs) and zooplankton samples were homogenised and weighed (0.5–0.6 mg of powder) prior to SIA of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) conducted as described in Eloranta, Knudsen & Amundsen (2013). The Stable Isotope Analysis in R, version 4.1.3 (SIAR; Parnell *et al.*, 2010) package was used to compute the relative contribution of littoral carbon in the long-term diet of Arctic charr. Comparisons of Arctic charr littoral reliance between seasons and size groups were conducted by comparing the 95% Bayesian credibility intervals which were considered significantly different when showing no overlap. Only those zoobenthos and zooplankton taxa found in Arctic charr stomachs were chosen to calculate the littoral and pelagic baselines, respectively (see Table S1 in Supporting Information). Comparisons of Arctic charr long-term dietary (or *isotopic*) niche width between seasons were completed using the Stable Isotope Bayesian Ellipses in R (SIBER; Jackson *et al.*, 2011) method in the SIAR package. The SIBER metrics are reformulated in a Bayesian approach to incorporate uncertainties related to sampling and sample sizes (Jackson *et al.*, 2011). However, no niche width comparisons were performed between Arctic charr size groups due to insufficient sample sizes (Syväranta *et al.*, 2013). The commonly used trophic fractionation factors of 0.4 ± 1.3‰ for $\delta^{13}C$ and 3.4 ± 1.0‰ for $\delta^{15}N$ were used (Post, 2002). The fish muscle and liver $\delta^{13}C$ were not corrected for lipids due to recently demonstrated uncertainties related to commonly used lipid-normalisation procedures (e.g. Syväranta & Rautio, 2010; Boecklen *et al.*, 2011; Fagan *et al.*, 2011).

Fatty acid analysis

Fatty acids were analysed using a three-step extraction-methylation protocol. Ideal dry weights for analysis were as follows: 26–29 mg fish, 3–6 mg zoobenthos and 0.5–1.5 mg zooplankton. Freeze-dried and weighed samples were extracted using a chloroform-methanol wash cycle and determined to µg lipid per mg dry mass by gravimetry (Mettler Toledo XP26DR; Mettler Toledo AG, Greidensee, Switzerland). The extracted lipids were methylated using toluene and H_2SO_4-methanol and then solubilised in hexane. Fatty acids (C14–C24) were identified as fatty acid methyl esters (FAME) using a gas chromatograph equipped with a Supelco™ SP-2560 column (100 m, 25 mm i.d., 0.2 µm film thickness; Sigma-Aldrich Co., St. Louis, MO, U.S.A.) and a flame ionisation detector (TRACE GC-FID THERMO™; Thermo Fisher Scientific Corporation, Waltham, MA, U.S.A.). Total fatty acid concentrations were calculated using calibration curves based on known standard concentrations and are reported as µg FAMEmg C^{-1}. Only those values exceeding a 1 µg mg C^{-1} threshold are used in the results. The concentration of essential fatty acids (EFA) is the sum of linoleic acid (LIN; 18 : 2ω6), α-linolenic acid (ALA; 18 : 3ω3), stearidonic acid (SDA; 18 : 4ω3), arachidonic acid (ARA; 20 : 4ω6), eicosapentaenoic acid (EPA; 20 : 5ω3) and docosahexaenoic acid (DHA; 22 : 6ω3). EPA, ARA and DHA are considered as the three most physiologically important fatty acids for fish (Ahlgren, Vrede & Goedkoop, 2009).

We compared the FAME composition and concentration in Arctic charr between seasons and between individuals in the small and large size groups within each season. To evaluate the qualitative differences between littoral and pelagic prey items, we compared concentrations of total FAMEs and EFAs, proportion of body fat and EFAs as well as ratios of ω3 : ω6 and DHA : ARA. High ω3 : ω6 and DHA : ARA ratios usually indicate high food quality and are more often observed in carnivorous/benthivorous fish and in aquatic food sources than in herbivorous/omnivorous fish or in terrestrial food (Ahlgren *et al.*, 2009). The littoral prey included six families of zoobenthos (*G. lacustris*, chironomid, trichopteran and tipulid larvae, *Lymnaea* sp. and Oligochaeta), while the pelagic prey included the most common taxa from the summer zooplankton community (*E. graciloides*, *D. umbra*, *Bosmina* sp., *B. longimanus*, *C. abyssorum* and Rotifera *Kellicottia longispina* and *Asplanchna* sp.).

Statistics

Depending on the normality and homoscedasticity of the data, either a *t*-test or Mann–Whitney *U*-test was used to compare the stomach fullness, proportions of zooplankton and zoobenthos in stomach contents, concentrations of total FAMEs and EFAs, proportions of body fat and EFAs and the $\delta^{13}C$ and $\delta^{15}N$ values of all Arctic charr and individuals in the small and large size ranges between seasons and between size groups within each season. The same statistical tests were used to compare the concentrations of total FAMEs and EFAs and the proportions of body fat and EFAs in littoral and pelagic food sources. To reveal possible ontogenetic dietary shifts, linear regression was used to examine the significance of relationships between Arctic charr total length and muscle and liver $\delta^{13}C$ and $\delta^{15}N$ values. All statistical analyses were performed using PASW Statistics 18 for Windows (SPSS Inc., Chicago, IL, U.S.A.) and R 2.14.2 (R Development Core Team, 2012).

Results

Winter feeding of Arctic charr

Stomach content analysis showed that Arctic charr were actively feeding under the ice. Only two of 33 winter captured Arctic charr had completely empty stomachs (Table 1). Arctic charr showed significant difference in diet between the seasons (ANOSIM: $R = 0.17$, $P < 0.001$), and fed more actively and on more diverse prey in summer than in winter, as indicated by significantly higher stomach fullness and Levins' index values (Tables 1, 2 & 3). No *E. graciloides* calanoid copepods were found in Arctic charr stomachs in either season, whereas cladoceran zooplankton (particularly *D. umbra*) were an important food source in both seasons. According to SCA, Arctic charr fed significantly more on zooplankton in summer and significantly more on zoobenthos in winter (Tables 1, 2 & 3). The seasonal dietary difference resulted from the dominance of *G. lacustris* in winter and *D. umbra* and *B. longimanus* in summer (SIMPER contributions: 31, 26 and 17%, respectively).

In comparison, the SIAR results indicated that Arctic charr relied more on littoral than on pelagic carbon in both seasons and showed only a slightly (17–23%) higher littoral reliance in winter compared with summer (Table 1; Fig. 1). Correspondingly, the $\delta^{13}C$ values of Arctic charr muscle and liver tissue showed no statistically significant differences between the seasons

Table 1 Summary table of stomach contents (SCA), stable isotope (SIA) and fatty acid (FAA) data obtained from all, <200 and >200 mm Arctic charr caught from Lake Saanajärvi in winter and summer 2011

	Winter All	<200 mm	>200 mm	Summer All	<200 mm	>200 mm
SCA						
n	33	14	19	37	18	19
Empty stomachs	2	2	0	0	0	0
Stomach fullness (%)	32 ± 26	13 ± 11	46 ± 25	49 ± 26	57 ± 30	42 ± 20
Levins' B	2.5	2.8	2.4	4.0	2.3	3.5
Zooplankton proportion (%)	34 ± 20	36 ± 45	33 ± 42	63 ± 40	78 ± 32	50 ± 43
Zoobenthos proportion (%)	62 ± 44	64 ± 45	61 ± 44	34 ± 39	22 ± 33	45 ± 43
SIA						
n	33	14	19	29	18	15
LP_{muscle} (%)	84 (76–92)	75 (54–93)	87 (77–98)	67 (56–79)	67 (52–83)	67 (51–83)
LP_{liver} (%)	81 (71–90)	65 (45–84)	86 (75–97)	58 (47–69)	51 (38–64)	64 (48–81)
SEA_{muscle}	4.7 (3.2–6.3)	–	–	4.5 (3.1–6.1)	–	–
SEA_{liver}	7.8 (5.3–10.6)	–	–	6.6 (4.5–8.9)	–	–
FAA						
n	20	9	11	19	12	7
Body fat (%)	6 ± 1	6 ± 1	6 ± 1	8 ± 4	8 ± 4	9 ± 4
FAME concentration ($\mu g\ mg\ C^{-1}$)	59 ± 17	56 ± 21	63 ± 13	95 ± 43	80 ± 24	121 ± 57
EFA concentration ($\mu g\ mg\ C^{-1}$)	28 ± 13	24 ± 15	31 ± 10	46 ± 17	42 ± 15	52 ± 19
EFA (%)	44 ± 16	41 ± 19	47 ± 13	48 ± 13	50 ± 15	45 ± 7

All values are given as mean $\pm$ SD except Levins' B dietary niche indices, and the Stable Isotope Analysis in R littoral proportion (LP) and Stable Isotope Bayesian Ellipses in R niche width (SEA_B; Standard Ellipse Area) estimates presented as means with 95% Bayesian credibility intervals (in parentheses). No SEA niche width comparisons were performed between Arctic charr size groups due to insufficient sample sizes, Abbreviations in FAA refer to total fatty acid methyl esters (FAME) and essential fatty acids (EFA).

Table 2 Relative proportion (volume) of different prey items in the stomach contents of all Arctic charr and individuals in the <200 and >200 mm size ranges caught in winter and summer

	n	Cladocera	Pred. clad.	Copepoda	*Gammarus lacustris*	Chiron. l.	Other zoob.	Pleuston	Arctic charr
Winter									
All	33	31	0	0	54	5	2	0	8
<200 mm	14	31	0	0	46	21	1	0	0
>200 mm	19	31	0	0	56	1	2	0	10
Summer									
All	37	44	26	<1	18	2	8	2	0
<200 mm	18	72	14	0	5	2	6	<1	0
>200 mm	19	8	42	<1	35	<1	10	4	0

Corresponding Levins' B dietary niche width estimates are given in Table 1. Abbreviations refer to predatory cladocerans (Pred. clad.), chironomid larvae (Chiron. l.) and other zoobenthos (Other zoob.).

(Table 3). The muscle $\delta^{15}N$ values were statistically, although not biologically (on average 0.6‰), significantly higher in summer than in winter (Table 3). The SIBER isotopic niche width estimates showed no significant differences between the seasons, but the liver tissue showed slightly higher isotopic variation than the muscle tissue (Table 1). The FAA data indicated Arctic charr had both higher percentage of body fat and concentrations of total FAMEs and EFAs in summer compared with winter (Tables 1 & 3). However, neither the relative proportion of EFAs (Tables 1 & 3) nor FA composition (Fig. 2) differed between seasons.

Size-related differences in Arctic charr seasonal feeding

When looking at the SCA, SIA and FAA results regardless of season, there were no significant differences ($P > 0.05$) in stomach fullness, proportion of zooplankton and zoobenthos in stomach contents, muscle $\delta^{13}C$ and $\delta^{15}N$ values, or proportions of EFA or body fat

between small and large Arctic charr (Tables 1 & 4; Fig. 1). In contrast, small Arctic charr had significantly lower liver $\delta^{13}C$ and $\delta^{15}N$ values (on average 1.9 and
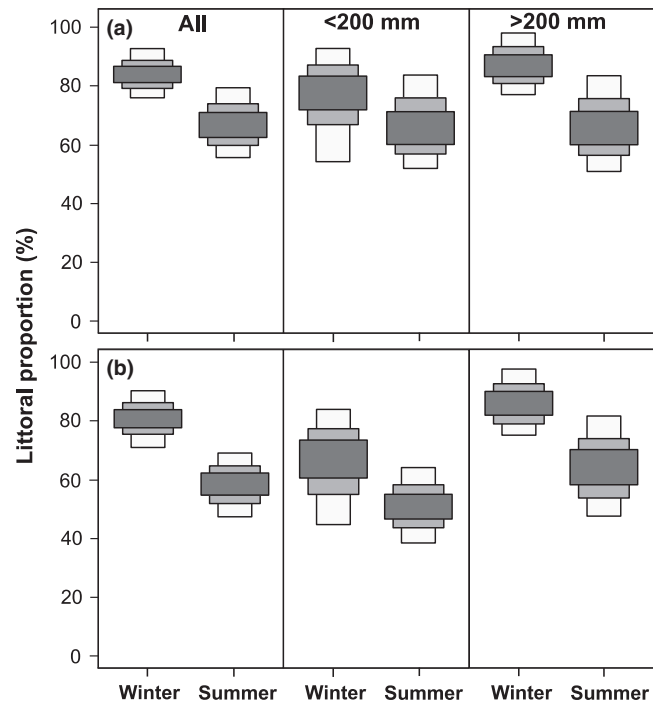

Fig. 1 Proportion of littoral dietary carbon in (a) muscle and (b) liver of all Arctic charr and individuals in the <200 and >200 mm size ranges caught in winter and summer. The boxes indicate the 95, 75 and 50% Bayesian credibility intervals for estimates based on the Stable Isotope Analysis in R isotopic mixing model that compares the $\delta^{13}C$ and $\delta^{15}N$ values from fish tissues to the $\delta^{13}C$ and $\delta^{15}N$ values of littoral zoobenthos and pelagic zooplankton.

0.5‰, respectively) compared with larger conspecifics (Table 4). The linear regressions between Arctic charr total length and muscle and liver $\delta^{13}C$ values provided no evidence of an ontogenetic shift from pelagic to littoral diet or *vice versa* ($P > 0.05$), whereas the liver $\delta^{15}N$ values indicated a shift to higher trophic position ($P < 0.05$) with increasing total length (see Fig. S2 in Supporting Information). Overall, both $\delta^{13}C$ and $\delta^{15}N$ showed marked individual variation regardless of Arctic charr total length (Fig. S2).

Seasonal feeding activity and diet composition depended on Arctic charr size, as only individuals smaller than 200 mm had significantly emptier stomachs and lower proportions of zooplankton and higher proportions of zoobenthos in stomach contents in winter compared with summer. No similar seasonal changes were observed in larger Arctic charr (Tables 1 & 3). In summer, the small fish had significantly higher proportions of EFA than their larger conspecifics (Tables 1 & 4). However, the large fish relied slightly more on littoral carbon in winter compared with summer, whereas there were less evident seasonal differences in the muscle and liver SIAR estimates of small Arctic charr (Table 1; Fig. 1). The two Arctic charr size groups showed similar diets in winter (ANOSIM: $R = 0.06$, $P = 0.12$), although the small fish showed a slightly lower littoral reliance and a higher consumption of chironomid larvae, while a single large Arctic charr (total length 330 mm) had fed on two smaller (100 and 112 mm) conspecifics (Fig. 1; Tables 2 & 4). Despite insignificant differences in dietary proportions of zooplankton and zoobenthos (Table 4)

Table 3 Statistical comparisons between seasons of stomach fullness, proportion of zooplankton and zoobenthos in stomach contents, muscle and liver $\delta^{13}C$ and $\delta^{15}N$ values, and concentrations of total fatty acid methyl esters (FAME) and essential fatty acids (EFAs) and proportions of body fat and EFAs in muscle of all, <200 and >200 mm Arctic charr

	Both seasons	Winter	Summer
SCA			
Stomach fullness	$U = 480$, $n = 70$, $P = 0.13$	$U = 29$, $n = 33$, $P < \mathbf{0.01}$	$t = 1.83$, $n = 37$, $P = 0.08$
Zooplankton proportion	$U = 452$, $n = 68$, $P = 0.13$	$U = 107$, $n = 31$, $P = 0.80$	$U = 109$, $n = 37$, $P = 0.06$
Zoobenthos proportion	$U = 489$, $n = 68$, $P = 0.30$	$U = 99$, $n = 31$, $P = 0.56$	$U = 120$, $n = 37$, $P = 0.13$
SIA			
Muscle $\delta^{13}C$	$t = -1.28$, $n = 66$, $P = 0.21$	$t = -1.76$, $n = 33$, $P = 0.09$	$t = 0.119$, $n = 33$, $P = 0.91$
Muscle $\delta^{15}N$	$t = 0.37$, $n = 66$, $P = 0.71$	$t = -1.27$, $n = 33$, $P = 0.21$	$U = 124$, $n = 33$, $P = 0.71$
Liver $\delta^{13}C$	$U = 350$, $n = 64$, $P = \mathbf{0.03}$	$t = -2.35$, $n = 31$, $P = \mathbf{0.03}$	$U = 102$, $n = 33$, $P = 0.24$
Liver $\delta^{15}N$	$U = 308$, $n = 64$, $P < \mathbf{0.01}$	$t = -1.74$, $n = 31$, $P = 0.09$	$t = -1.86$, $n = 33$, $P = 0.07$
FAA			
FAME concentration	$U = 164$, $n = 37$, $P = 0.85$	$U = 33$, $n = 19$, $P = 0.40$	$U = 26$, $n = 18$, $P = 0.29$
EFA concentration	$U = 159$, $n = 37$, $P = 0.73$	$U = 40$, $n = 19$, $P = 0.78$	$U = 33$, $n = 18$, $P = 0.66$
Body fat (%)	$U = 139$, $n = 37$, $P = 0.34$	$t = 0.39$, $n = 19$, $P = 0.71$	$U = 34$, $n = 18$, $P = 0.72$
EFA (%)	$U = 122$, $n = 37$, $P = 0.14$	$U = 40$, $n = 19$, $P = 0.78$	$t = 3.34$, $n = 18$, $P < \mathbf{0.01}$

Test statistics for *t*-tests and Mann–Whitney *U*-tests are shown and significant differences ($P < 0.05$) are marked in bold.

and littoral carbon (Fig. 1), the two Arctic charr size groups showed a significant diet segregation ($R = 0.30$, $P < 0.001$) in summer, when small fish fed mainly on cladoceran zooplankton, while larger conspecifics fed on larger prey such as *G. lacustris* and *B. longimanus* predatory cladocerans (Table 2). *Daphnia umbra*, *B. longimanus* and *G. lacustris* were the most important prey taxa contributing to the dietary differences between small and

Fig. 2 Fatty acid concentrations (mean ± SE) and ratios of ω3 : ω6 and DHA : ARA of (a) Arctic charr caught in winter ($n = 20$) and summer ($n = 19$), and of (b) littoral (zoobenthos, $n = 8$) and pelagic (zooplankton, $n = 11$) food sources sampled in summer 2011 from Lake Saanajärvi. Note the different scales on the (a) and (b) y-axes.

large Arctic charr (SIMPER contributions: 28, 23 and 18%, respectively).

Qualitative differences between littoral and pelagic food sources

Both littoral and pelagic food sources provided all FAMEs and in higher concentrations than was found in Arctic charr (Fig. 2). Littoral and pelagic food sources did not differ significantly in concentrations of total FAMEs ($t = 0.99$, $n = 19$, $P = 0.34$) or EFAs ($U = 21$, $n = 19$, $P = 0.06$), or in percentages of body fat ($U = 23$, $n = 19$, $P = 0.09$). However, the pelagic food sources had significantly higher proportions of EFAs ($t = 4.86$, $n = 19$, $P < 0.001$) and ratios of ω3 : ω6 ($U = 0$, $n = 19$, $P < 0.001$) and DHA : ARA ($U = 2$, $n = 19$, $P < 0.001$) than the littoral prey items (Table 5). All zoobenthos except *G. lacustris* had lower than detection limit DHA values and thus zero DHA : ARA ratios. *G. lacustris* had the highest FAME concentrations of the zoobenthos, with concentrations exceeding those of all zooplankton except *E. graciloides*, which had more than double the FAME and EFA concentrations of all prey items (Table 5). To summarise, the FAA results suggest that both pelagic and littoral food sources can generally provide similar EFAs but pelagic zooplankton and benthic crustaceans may be nutritionally superior prey for fish.

Seasonal FA comparisons were carried out only for zooplankton because no zoobenthos were sampled in winter. *Eudiaptomus graciloides* had higher FAME and EFA concentrations in summer (mean ± SE: FAME:
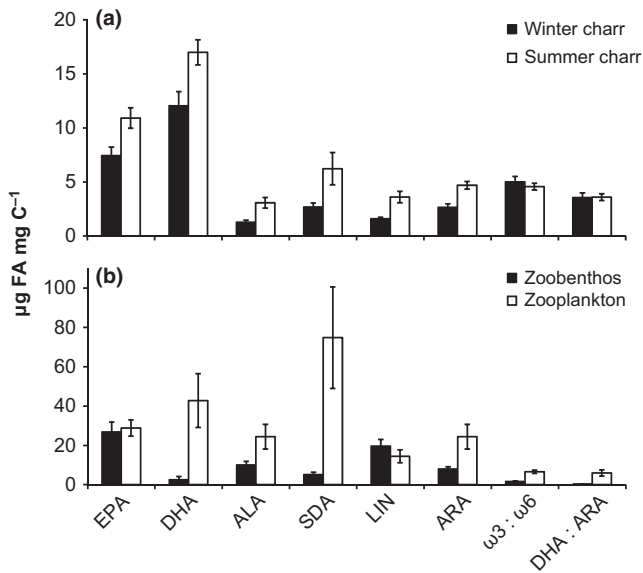
Table 4 Statistical comparisons between <200 and >200 mm Arctic charr of stomach fullness, proportion of zooplankton and zoobenthos in stomach contents, muscle and liver $\delta^{13}C$ and $\delta^{15}N$ values, and concentrations of total fatty acid methyl esters (FAME) and essential fatty acids (EFAs) and proportions of body fat and EFAs in muscle tissue in both seasons and separately in winter and summer

	Both seasons	Winter	Summer
SCA			
Stomach fullness	$U = 480$, $n = 70$, $P = 0.13$	$U = 29$, $n = 33$, $P < \textbf{0.01}$	$t = 1.83$, $n = 37$, $P = 0.08$
Zooplankton proportion	$U = 452$, $n = 68$, $P = 0.13$	$U = 107$, $n = 31$, $P = 0.80$	$U = 109$, $n = 37$, $P = 0.06$
Zoobenthos proportion	$U = 489$, $n = 68$, $P = 0.30$	$U = 99$, $n = 31$, $P = 0.56$	$U = 120$, $n = 37$, $P = 0.13$
SIA			
Muscle $\delta^{13}C$	$t = -1.28$, $n = 66$, $P = 0.21$	$t = -1.76$, $n = 33$, $P = 0.09$	$t = 0.119$, $n = 33$, $P = 0.91$
Muscle $\delta^{15}N$	$t = 0.37$, $n = 66$, $P = 0.71$	$t = -1.27$, $n = 33$, $P = 0.21$	$U = 124$, $n = 33$, $P = 0.71$
Liver $\delta^{13}C$	$U = 350$, $n = 64$, $P = \textbf{0.03}$	$t = -2.35$, $n = 31$, $P = \textbf{0.03}$	$U = 102$, $n = 33$, $P = 0.24$
Liver $\delta^{15}N$	$U = 308$, $n = 64$, $P < \textbf{0.01}$	$t = -1.74$, $n = 31$, $P = 0.09$	$t = -1.86$, $n = 33$, $P = 0.07$
FAA			
FAME concentration	$U = 164$, $n = 37$, $P = 0.85$	$U = 33$, $n = 19$, $P = 0.40$	$U = 26$, $n = 18$, $P = 0.29$
EFA concentration	$U = 159$, $n = 37$, $P = 0.73$	$U = 40$, $n = 19$, $P = 0.78$	$U = 33$, $n = 18$, $P = 0.66$
Body fat (%)	$U = 139$, $n = 37$, $P = 0.34$	$t = 0.39$, $n = 19$, $P = 0.71$	$U = 34$, $n = 18$, $P = 0.72$
EFA (%)	$U = 122$, $n = 37$, $P = 0.14$	$U = 40$, $n = 19$, $P = 0.78$	$t = 3.34$, $n = 18$, $P < \textbf{0.01}$

Test statistics for t-tests and Mann–Whitney U-tests are shown, and significant differences ($P < 0.05$) are marked in bold.

Table 5 Mean ± SD percentages of body fat and essential fatty acid (EFA), concentrations of total fatty acid methyl esters (FAME) and EFAs, and ratios of ω3 : ω6 and DHA : ARA in pelagic and littoral prey items sampled from Lake Saanajärvi in summer 2011

		Proportion (%)		Concentration (µg mg C^{-1})		Ratio	
	n	Body fat	EFA	FAME	EFA	ω3 : ω6	DHA : ARA
Pelagic	11	34 ± 0.2	0.5 ± 0.1	334 ± 250	193 ± 171	6.6 ± 2.7	6.0 ± 5.2
Eudiaptomus graciloides	3	67 ± 0.1	0.6 ± 0.0	716 ± 21	455 ± 17	10 ± 0.1	13.0 ± 0.2
Daphnia umbra	2	23	0.6	243	137	4.7	1.2
Bythotrephes longimanus	1	20	0.4	186	83	2.7	0.18
Cyclops abyssorum	1	30	0.4	317	137	7.5	5.87
Bosmina sp.	1	18	0.4	138	54	3.5	1.0
Rotifera	2	15	0.5	125	65	7.4	8.8
Littoral	8	19 ± 0.1	0.3 ± 0.1	239 ± 150	72 ± 29	1.6 ± 0.7	0.2 ± 0.4
Gammarus lacustris	2	31	0.2	467	105	2.4	0.8
Chironomidae	2	19	0.4	224	85	1.1	0
Lymnaea sp.	1	17	0.3	108	30	1.5	0
Oligochaeta	1	12	0.4	127	47	0.8	0
Tipulidae	1	9	0.4	106	42	1.1	0
Trichoptera	1	16	0.4	187	76	2.7	0

715 ± 12 µg mg C^{-1}; EFA: 455 ± 10 µg mg C^{-1}) than in winter (570 ± 205 µg mg C^{-1}; EFA: 334 ± 132 µg mg C^{-1}; see Fig. S3 in Supporting Information). In contrast, *D. umbra* had almost twice the FAME and EFA concentrations in winter (FAME: 422 ± 26 µg mg C^{-1}; EFA: 225 ± 16 µg mg C^{-1}) than in summer (FAME: 243 µg mg C^{-1}; EFA: 137 µg mg C^{-1}). Both species had higher DHA: ARA ratios in summer than in winter (mean ± SE: *D. umbra*: 1.2 versus 0.8 ± 0.1; *E. graciloides*: 12.6 ± 0.1 versus 7.7 ± 0.3). *Eudiaptomus graciloides* had also higher ω3 : ω6 ratios in summer than in winter (10.0 ± 0.1 versus 7.3 ± 0.4), while *D. umbra* had seasonally unvarying ω3 : ω6 ratios (4.7 versus 4.9 ± 0.1). To summarise the species-specific seasonal trends, *E. graciloides* had higher concentrations of DHA and ARA in summer and more than *D. umbra* at any time, while *D. umbra* had higher concentration of ALA, LIN and EPA in winter (Fig. S3).

Discussion

Our results show that Arctic charr feed actively on zooplankton in winter, but generally rely more on littoral than on pelagic carbon sources all year round. The seasonal feeding activity and dietary shifts of Arctic charr were partly explained by fish size. Small Arctic charr (<200 mm) shifted to a predominantly zooplanktivorous diet and had significantly fuller stomachs and slightly higher concentrations of EFAs in muscle tissue in summer compared with winter. In contrast, larger (>200 mm) Arctic charr showed less seasonally variable stomach contents, feeding activity and levels of EFAs in muscle. Overall, Arctic charr had significantly higher body fat proportions and total FAME and EFA concentrations in summer, probably due to generally more active feeding and retention of FA for overwintering storage and/or reproduction. The FA analyses indicated that littoral and pelagic food sources provided similar EFAs to Arctic charr and did not differ in body fat or concentrations of total FAMEs and EFAs. However, zooplankton had significantly higher percentages of EFA and ratios of ω3 : ω6 and DHA : ARA and thus were a qualitatively superior food for Arctic charr compared with zoobenthos. FAME concentrations were highest in pelagic *E. graciloides* copepods and in littoral *G. lacustris* amphipods, but only the latter was an important food source for Arctic charr.

Do Arctic charr feed on lipid-rich zooplankton in winter?

Our results provide strong evidence for active feeding of Arctic charr in winter. This is supported by previous field and laboratory studies that showed Arctic charr are well adapted to feed in extremely cold and dark conditions under the winter ice (Klemetsen *et al.*, 2003; Svenning, Klemetsen & Olsen, 2007; Amundsen & Knudsen, 2009; Elliott, 2011; Helland *et al.*, 2011). The FAA data suggest that Saanajärvi Arctic charr gained body fat and retained high concentrations of EFAs in summer, which probably promotes survival through winter. Similar trends were found in overwintering zooplankton (*D. umbra* and *E. graciloides*), which had highest lipid concentrations in the month prior to ice cover, with concentration gradually declining throughout winter (Syväranta & Rautio, 2010). The consistently high

amount of EFA (>40%) observed in both seasons and Arctic charr size groups indicates that there is a minimal threshold required to maintain metabolic function independent of external environmental stresses such as food limitation and temperature.

Based on our results from SCA, SIA and FAA, lipid-rich zooplankton supplement the predominantly benthivorous winter diet of Arctic charr. Correspondingly, previous studies conducted in Takvatn and Fjellfrøsvatn, northern Norway, have shown that zooplankton can be an important prey in autumn/early winter, especially for small Arctic charr (Klemetsen et al., 2003; Amundsen et al., 2008; Amundsen & Knudsen, 2009). The seasonal shift of Arctic charr to a zooplanktivorous diet in autumn/early winter has been commonly suggested to result from reduced littoral food resources and subsequently increased resource competition in the littoral zone (Amundsen, 1995; Eloranta et al., 2010). Despite the extremely cold and dark conditions, Arctic charr in Saanajärvi were found to be selective in their zooplankton feeding, preying mainly on cladocerans despite the previously observed winter predominance of E. graciloides calanoids (Rautio et al., 2000, 2011). Consistent with previous studies (Smyntek et al., 2008), the FAA data indicated that E. graciloides calanoids have more EFAs than the cladocerans and thus would provide a more nutritious food for fish. Hence, E. graciloides would be an ideal food source for Saanajärvi Arctic charr in both quantity and quality. However, copepods have a superior ability to escape predation by planktivorous fish due to their rapid movements (Kiørboe, 2011). Selective predation of Arctic charr on cladocerans over copepods is a well-reported phenomenon and has in some cases been found to affect zooplankton community composition and size structure (Nilsson & Pejler, 1973; Langeland, 1982; Dahl-Hansen, Rubach & Klemetsen, 1994).

The SIA data indicated minor seasonal changes in the long-term diet or niche width of Arctic charr, while the SCA suggested a shift to a largely zooplanktivorous diet and to a wider dietary niche in summer compared with winter, when Arctic charr stomach contents were dominated by littoral G. lacustris. The high and seasonally invariable littoral reliance of Arctic charr may be due to more efficient energy transfer through the littoral food-web compartment that results from the larger size of zoobenthos relative to small pelagic zooplankton (Vander Zanden et al., 2006). The apparent seasonal constancy of Arctic charr muscle and liver $\delta^{13}C$ may also result from the effects of prevailing cold water temperatures on metabolic rate and the associated reduction in

isotopic fractionation in winter. For example, Power, Guiguer & Barton (2003) noted significantly lower carbon fractionation in Daphnia magna raised at lower temperatures and Barnes et al. (2007) have noted similar fractionation effects in fish. Perga & Gerdeaux (2005) studied the seasonal changes in $\delta^{13}C$ and $\delta^{15}N$ values of planktivorous whitefish (Coregonus lavaretus) and found that the isotopic composition of liver responded more quickly and consistently to seasonal changes in zooplankton $\delta^{13}C$ and $\delta^{15}N$ values compared with muscle tissue that has longer isotopic turnover rates. In addition, they found that the whitefish muscle $\delta^{13}C$ and $\delta^{15}N$ values only reflected the main food sources consumed during the spring and summer growth period. Thus, the muscle isotopic composition of Saanajärvi Arctic charr may largely reflect the main dietary sources throughout the open water seasons when most growth occurs (Klemetsen et al., 2003). Consistent with some other studies (Amundsen et al., 2008; Amundsen & Knudsen, 2009), Arctic charr in Saanajärvi have been observed to feed mainly on littoral zoobenthos (particularly chironomid larvae and G. lacustris) from early spring to midsummer (Eloranta et al., 2010), which could largely explain the high $\delta^{13}C$ values observed here in August 2011. Alternatively, the dominance of littoral carbon in Arctic charr muscle tissue may result from differential allocation of littoral and pelagic food resources as a result of isotopic routing (Wolf, Carleton & Martinez del Rio, 2009; Boecklen et al., 2011). For example, large zoobenthos might provide carbon for growth, while small zooplankton may be more quickly assimilated for basic maintenance. The relatively faster turnover and higher assimilation of pelagic carbon to basic metabolism may also explain the slightly higher isotopic variation and lower $\delta^{13}C$ and $\delta^{15}N$ values observed in liver tissue of Saanajärvi Arctic charr.

Do the diet and fatty acids composition depend on Arctic charr size?

Size is a critical factor for fish foraging behaviour and survival as it affects the size range of potential prey items, competitive dominance, predation risk and metabolic rate (e.g. Werner & Gilliam, 1984; Persson & De Roos, 2007). In some subarctic lakes, small Arctic charr have been shown to face a trade-off between using seasonally abundant zooplankton resources and avoiding predators in the pelagic zone (L'Abée-Lund et al., 1993; Damsgård & Ugedal, 1997). Small Arctic charr also have a higher ratio of energy levels to metabolic rate and thus have a higher risk of winter starvation mortal-

ity compared with larger conspecifics (Byström *et al.*, 2006). The results from Saanajärvi suggest that in winter, Arctic charr smaller than 200 mm show a significantly lower feeding activity (emptier stomachs) and a slightly lower concentration of EFAs compared with larger conspecifics or to similar-sized Arctic charr caught in summer. In contrast, larger (>200 mm) Arctic charr showed a less seasonally variable stomach fullness and EFA percentage, suggesting more active foraging throughout the year, possibly due to lower predation risk and competitive superiority over smaller conspecifics. The active winter feeding of larger Arctic charr probably promotes temporally stable isotopic turnover and thus may explain the observed slight seasonal shift in muscle and liver $\delta^{13}C$ values resulting from a shift from benthivorous winter diet to partial zooplanktivory in summer.

Although the SIA data provided no evidence of ontogenetic dietary shifts or seasonal changes in the long-term diet of smaller Arctic charr, the SCA data indicated a significantly higher consumption of zooplankton in summer. While the noted low abundances of brown trout and cannibalistic Arctic charr may have reduced predation-related risks of zooplankton foraging in Saanajärvi (cf. Eloranta *et al.*, 2010), similar seasonal zooplanktivorous diets in small Arctic charr have been reported elsewhere. For example, small Arctic charr in temperate Loch Ness (Grey *et al.*, 2002), in subarctic Fjellfrøsvatn (Amundsen & Knudsen, 2009) and in high-arctic Linnévatn (Svenning *et al.*, 2007) have been observed to consume zooplankton despite the existence of piscivorous brown trout or cannibalistic Arctic charr. The observed slight increase in liver $\delta^{15}N$ with Arctic charr size probably results from facultative cannibalism of large individuals (McCarthy *et al.*, 2004; Eloranta *et al.*, 2010), but may also result from foraging on profundal chironomids with elevated $\delta^{15}N$ values (Sierszen, Peterson & Scharold, 2006). The concurrent increases in zooplanktivory and EFA percentage suggest that small Arctic charr utilise increased pelagic resources in summer to build FA reserves for overwintering. The seasonal dietary shift of smaller fish may also be due to increased competition for littoral food resources that may reach an annual minimum in late summer (Svenning *et al.*, 2007). Nevertheless, the high individual variation observed in Arctic charr stomach contents and $\delta^{13}C$ and $\delta^{15}N$ values probably reflects a high degree of individual dietary specialisation within the population, which is commonly suggested to result from strong intraspecific resource competition for limited food resources (Amundsen, 1995; Araújo, Bolnick & Layman, 2011).

Do littoral and pelagic food sources show qualitative differences?

Littoral production has been shown to dominate both primary production (Vadeboncoeur *et al.*, 2003) and the energy supply to top consumers in clear-water high-latitude lakes (Sierszen *et al.*, 2003; Karlsson & Byström, 2005; Eloranta *et al.*, 2010), where the lack of dissolved nutrients and organic carbon limits phytoplankton growth and simultaneously facilitates the growth of benthic algae. Besides higher benthic primary production, fish may rely upon littoral carbon sources due to the higher efficiency of littoral energy transfer (Hecky & Hesslein, 1995; Vander Zanden *et al.*, 2006, 2011). Moreover, benthic algae may also subsidise the energy sources for pelagic cladocerans as demonstrated by recent field and laboratory studies (Karlsson & Säwström, 2009; Mariash *et al.*, 2011; Cazzanelli *et al.*, 2012). Correspondingly, the SIA results from Saanajärvi show that Arctic charr can be highly dependent on littoral carbon sources throughout the year, but also from year to year (cf. Eloranta *et al.*, 2010). The present FAA data indicate that zoobenthos generally have similar FAME compositions and concentrations but provide EFA in lower proportions than zooplankton. The lower ω3 : ω6 ratios of zoobenthos matches what has been previously reported for deposit-feeding taxa, which generally have lower ω3 : ω6 ratios than taxa feeding on phytoplankton (Ahlgren *et al.*, 2009). Moreover, the low ω3 : ω6 and DHA : ARA ratios indicate that zoobenthos can also utilise terrestrial food sources (Ahlgren *et al.*, 2009). Of the analysed littoral prey items, *G. lacustris* had the highest concentrations of total FAMEs and EFAs and thus may play a significant role in Arctic charr survival through the harsh winter conditions. Because of the larger size and similar FAME composition to zooplankton, zoobenthos might be more easily captured under the ice and thus constitute a more energetically profitable prey, particularly for large Arctic charr. For smaller fish, small zooplankton (particularly *D. umbra*) are apparently profitable prey, probably due to their high amounts of EFAs. However, the results suggest that regardless of season or fish size, pelagic zooplankton overall can be a nutritious food source and subsidise the predominant benthivorous diet of top consumers in high-latitude lakes.

To conclude, generalist Arctic charr have evidently adapted to seasonal fluctuations in food availability and composition (Klemetsen *et al.*, 2003; Svenning *et al.*, 2007; Amundsen & Knudsen, 2009; Eloranta *et al.*, 2010) and thus have potential to act as important links

between littoral and pelagic habitats and food-web compartments in high-latitude lakes (cf. Schindler & Scheuerell, 2002; Vander Zanden & Vadeboncoeur, 2002). The dietary plasticity of Arctic charr probably alleviates oscillations in littoral and pelagic trophic pathways (Vadeboncoeur *et al.*, 2005; Rooney *et al.*, 2006; Kratina *et al.*, 2012), whereas the high individual variation in foraging behaviour decreases intraspecific competition within the Arctic charr populations (Amundsen, 1995; Araújo *et al.*, 2011). The results demonstrate that littoral and pelagic habitats and food-web compartments are highly integrated and can act as important sources of carbon and physiologically important fatty acids, supporting top consumers in high-latitude lakes.

Acknowledgments

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Figure S1 Map of the study lake Saanajärvi.

Figure S2 Linear regressions between Arctic charr total length and $\delta^{13}C$ and $\delta^{15}N$ values.

Figure S3 Seasonal concentrations of essential fatty acids in zooplankton.

Table S1. Stable isotope values of zooplankton and zoobenthos taxa