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Spatio-temporal variability in the winter diet of larval and juvenile Antarctic krill, *Euphausia superba*, in ice-covered waters

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ABSTRACT: Antarctic krill *Euphausia superba* is an ecological key species in the Southern Ocean and a major fisheries resource. The winter survival of age class 0 (AC0) krill is susceptible to changes in the sea-ice environment due to their association with sea ice and their need to feed during their first winter. However, our understanding of their overwintering diet and its variability is limited. We studied the spatio-temporal variability of the diet in 4 cohorts of AC0 krill in the Northern Weddell Sea during late winter 2013 using stomach contents, fatty acids (FAs) and bulk stable isotope analysis (BSIA). Stomach contents were dominated by diatoms in numbers and occasionally contained large volumes of copepods. Many of the prey species found in the stomachs were sea ice-associated. Our results show that the diet of overwintering AC0 krill varies significantly in space and time. Variability in stomach content composition was related to environmental factors, including chlorophyll *a* concentration, copepod abundance and sea-ice cover. In contrast, FA composition mainly varied between cohorts, indicating variation in the long-term diet. The condition of the AC0 krill was reflected in FA and BSIA analysis, suggesting that the availability of sea ice-derived food sources over a long period may impact the condition of developing AC0 krill significantly. The spatio-temporal availability of sea-ice resources is a potentially important factor for AC0 krill winter survival.

KEY WORDS: *Euphausia superba* · Winter · Stomach contents · Fatty acids · Sea ice · Weddell Sea · Furcilia · Juveniles

INTRODUCTION

Due to the pronounced seasonality in the polar regions, polar species need to adapt to drastic changes in primary production (Falk-Petersen et al. 1999, Hagen & Auel 2001). In the Southern Ocean, light limitation and water column mixing due to surface water cooling result in a long period of near-zero primary production during wintertime (Arrigo et al. 2008). During the winter months, biota living in sea ice and at its underside can provide an important energy source (Eicken 1992, Quetin & Ross 2003, Flores et al. 2011, 2012a). In spring, primary production increases in the sea ice as well as in the water column. As the ice edge retreats, starting in September, a series of water column phytoplankton blooms occur (Quetin & Ross 1991, Lizotte 2001). In late summer there is another peak in the water column primary production, after which it starts to decrease towards winter (Quetin & Ross 1991, Lizotte 2001).

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Adult Antarctic krill *Euphausia superba* release eggs from mid-December to April (Ross & Quetin 1986). The duration of the spawning season of krill and the number of spawning episodes that occur can be variable (Ross & Quetin 1986, Spiridonov 1995). Multiple spawning episodes increase the chance of producing larvae that reach the first feeding stage at a time when there is enough food available in the environment, since the timing and length of phytoplankton blooms are highly variable and unpredictable (Quetin & Ross 1991).

Adult *E. superba* overwinter by reducing metabolic activity in combination with opportunistic feeding and utilization of body lipids or body shrinkage (Ikeda & Dixon 1982, Meyer et al. 2010, Virtue et al. 2016). In contrast to adult krill, larvae are not able to survive long periods of starvation (Meyer et al. 2009, O'Brien et al. 2011), and the first winter is therefore considered a critical period for krill survival and recruitment (Quetin et al. 2003, Daly 2004, Flores et al. 2012b). Krill larvae are assumed to rely on sea-ice resources (Daly 1990, Meyer et al. 2002, Meyer 2012), but in addition show flexible overwintering behaviour such as a delay of development, an increased inter-moult period, growth reduction and moderate lipid storage (Hagen et al. 2001, Daly 2004).

Krill larvae often reside directly underneath the sea ice in winter (Frazer et al. 2002, Meyer et al. 2009, Flores et al. 2012a, Schaafsma et al. 2016, David et al. 2017). Using a Surface and Under-Ice Trawl (SUIT; van Franeker et al. 2009), Schaafsma et al. (2016) conducted a large-scale investigation of the krill population structure directly underneath the sea ice in the northern Weddell Sea during winter/early spring of 2013 (Fig. 1). The population mostly comprised larvae (furcilia) and juveniles experiencing their first winter, subsequently referred to as age-class 0 (AC0) krill. The AC0 krill population consisted of several spatially separated cohorts, differing in size and developmental stage composition. The differences between these cohorts could have been caused by a dissimilar timing of spawning and/or different growth conditions due to variable environmental conditions encountered on differing advection paths (Quetin & Ross 2003, Schwegmann 2012, Schaafsma et al. 2016). Furthermore, the metazoan community structure in the ice–water interface layer in the northern Weddell Sea showed a distinct spatial structure, consisting of 3 distinct community types which could be attributed to spatially and seasonally varying environmental conditions (David et al. 2017). These observations indicated that the environmental

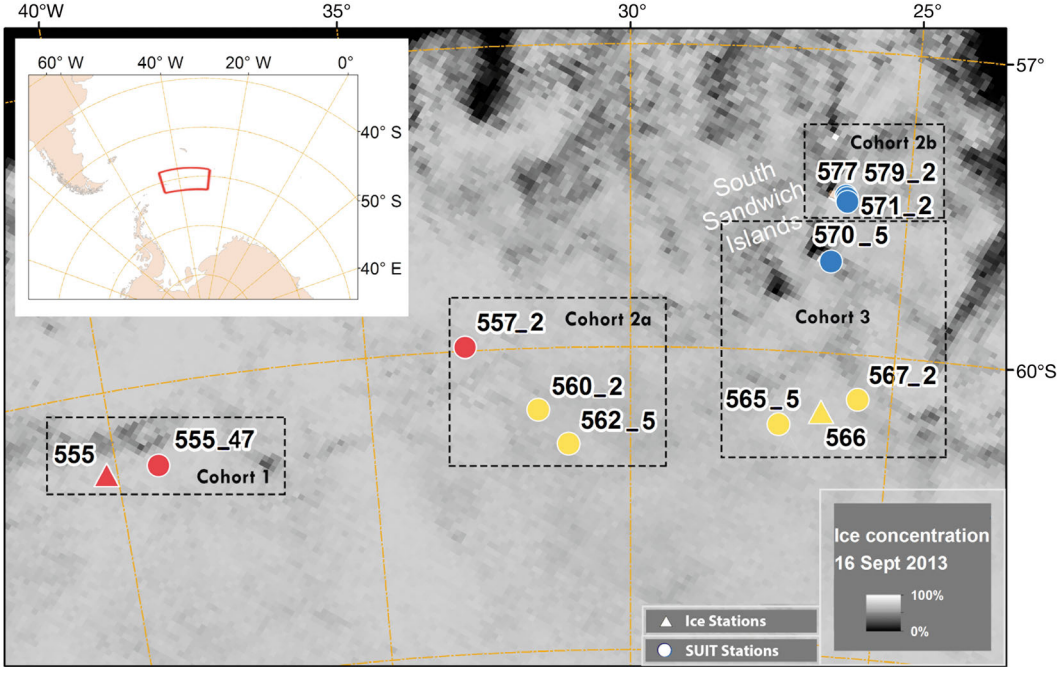

Fig. 1. Sampling locations of the Surface and Under-Ice Trawl (SUIT), indicated with their station numbers. Sea ice concentration data were acquired on 16 September 2013. Round = SUIT hauls, triangles = stationary hauls at ice stations. Black dotted lines indicate the spatial pattern of cohorts of age class 0 krill *Euphausia superba*, as established by Schaafsma et al. (2016). Cohort numbers are indicated within their respective rectangle. Stations are coloured according to the spatial pattern in the under-ice zooplankton community as established by David et al. (2017): red = 'krill-dominated', yellow = 'copepod-dominated', blue = 'low biomass' community

regime in the northern Weddell Sea was influenced by various interacting drivers, such as ocean currents, phytoplankton and ice algae concentrations and sea-ice drift, creating a heterogeneous pattern of food availability and food composition for overwintering krill. This is important because growth and development of overwintering larval krill are strongly influenced by food supply and food type (Ross et al. 1988, Ross & Quetin 1989, Daly 1990).

Investigating the diet of AC0 krill can give insight in the survival through their first winter (Virtue et al. 2016). Due to the difficulty of sampling during winter, only a limited number of studies have described the stomach contents of larval krill during this season (Daly 1990, Ju & Harvey 2004, Meyer et al. 2009, O'Brien et al. 2011). Because of the small spatial coverage of these studies, determinants and variability of diet composition remain unclear. The analysis of stomach contents can provide essential information on the recent diet composition of a consumer. Combined with lipid and fatty acid (FA) compositions, it is possible to elucidate trophic interactions over larger temporal scales (Falk-Petersen et al. 1999, Dalsgaard et al. 2003, Kohlbach et al. 2016). Zooplankton lack the ability to biosynthesize certain FAs de novo. Hence, these essential FAs produced by primary producers are not metabolically modified and can be used as trophic markers to trace back dietary carbon sources (Lee et al. 1971, Graeve et al. 1994, Virtue et al. 2016). Diatoms (Bacillariophyceae) produce large amounts of the FAs 16:1n-7 and 20:5n-3, while dinoflagellates (Dinophyceae) produce large amounts of the FAs 18:4n-3 and 22:6n-3 (Graeve et al. 1994, Dalsgaard et al. 2003 and references therein). Sea-ice algae communities often contain high proportions of diatoms compared to the underlying water column (Garrison 1991, Lizotte 2001). Conversely, dinoflagellates are typically more abundant in the water column, compared to sea-ice communities (Garrison 1991, Lizotte 2001). The FA composition of krill can therefore give some qualitative insight into the origin of carbon in dietary sources.

The aim of this study was to evaluate temporal and spatial differences in the diet of AC0 krill in late winter/early spring. Microscopic stomach content analysis and FA analysis were combined to gain insight into the diet and carbon sources of *E. superba* during their first winter. Additional information was integrated, including carbon:nitrogen content (C:N mass ratio), as indicators of lipid storage and body condition of the krill. Furthermore, the isotopic fractionation of carbon ($\delta^{13}C$: $^{13}C/^{12}C$) was measured to assess the potential contribution of ice algae-derived carbon to the diet of overwintering krill. This is possible because the $\delta^{13}C$ values of sea ice-derived carbon are often higher compared to pelagic produced carbon (Fry & Sherr 1984, Hecky & Hesslein 1995, Jia et al. 2016, Kohlbach et al. 2016). The isotopic composition of nitrogen ($\delta^{15}N$: $^{15}N/^{14}N$) was used as an indicator of trophic position (DeNiro & Epstein 1981, Minagawa & Wada 1984).

We used this comprehensive methodical approach for a detailed analysis of the spatial variability of the trophic ecology of overwintering krill across a geographically large research area in the northern Weddell Sea, aiming to: (1) assess the importance of sea ice-associated carbon sources in the diet of overwintering krill; (2) investigate the association of the diet composition with spatio-temporal patterns in the environmental properties of the research area; and (3) analyse correlations between the size and stage composition of different krill cohorts and recent and long-term dietary sources.

MATERIALS AND METHODS

Sampling and data collection

Sampling was performed in the northern Weddell Sea during RV 'Polarstern' expedition PS81 (ANT-XXIX/7), between 24 August and 2 October 2013 (Fig. 1). The upper 2 m of the water column directly underneath the sea ice was sampled using a SUIT. Environmental parameters, such as sea-ice concentration and thickness, and under-ice surface water temperature, salinity and chlorophyll *a* (chl *a*) concentration, were measured during trawling using a sensor array attached in the frame of the SUIT, including an acoustic Doppler current profiler (Nortek, Aquadopp®) and a CTD probe (CTD75 M, Sea & Sun Technology) with connected altimeter (PA500/6-E, Tritech). In addition, regional gridded sea ice concentrations during SUIT hauls were calculated from AMSR2 satellite data, which were acquired from the sea ice portal of the Alfred Wegener Institute (AWI, www.meereisportal.de), using the algorithm from Spreen et al. (2008). Ice floe size was estimated visually during SUIT hauls by an observer on deck, and varied between 10 m and <1 km diameter. Detailed information on sampling and data collection can be found in David et al. (2015) and Schaafsma et al. (2016). Additional to trawling stations, krill were collected at sea-ice stations, by deploying the SUIT from the stationary ship in the current. *Euphausia superba* for stomach content analysis were directly preserved

in a 4% hexamine-buffered formaldehyde-seawater solution. *E. superba* for C:N ratio, FA and bulk stable isotope analyses were immediately frozen at –80°C.

Stomach analysis

Prior to stomach content analysis, the preserved krill were weighed and total length (TL) was measured, to the nearest mm, from the anterior margin of the eye to the tip of the telson ('Discovery method'; Marr 1962). The developmental stage of furcilia larvae was determined according to Kirkwood (1982). Juveniles were distinguished from furcilia and other post-larval krill according to Fraser (1936) and Makarov & Denys (1981). A Discovery V8 stereomicroscope (Zeiss) was used for krill dissection. After removing the carapace, the stomach was taken out, transferred into a tube with 2 ml of deionized water and mixed using a vortex to break the stomach wall. For each analytical sample, up to 3 stomachs abstracted from krill of comparable size were pooled together. The tube with the stomach contents was emptied into an Utermöhl sedimentation chamber, where it was left to settle for at least 2 h (Schmidt et al. 2006). Identifiable prey items were counted on an Observer A1 microscope (Zeiss) at 400× magnification in half of the counting receptacle. Rare prey items such as dinoflagellates, tintinnids, foraminiferans, radiolarians and copepod or other zooplankton remains were enumerated in the complete receptacle at 200× magnification. Pieces of broken pennate and centric diatoms were measured in order to reconstruct the number of individual diatoms in the stomach, by dividing the average size of the complete surface area of intact diatoms with the average size of the measured pieces of that species found in the stomachs. For unidentifiable diatom pieces, the average surface area of all intact diatoms was used (Garrison et al. 1987, Kang et al. 2001). The total biovolume of prey species or species group in the stomach was calculated by multiplying the number of individuals by the volume per individual from Archer et al. (1996) for dinoflagellates, Kang et al. (2001) for diatoms, Buck et al. (1992) for tintinnids and Gradinger (1999) for foraminiferans. When copepod remains were found in a sample, the number of copepods was estimated to be 1, unless there was evidence that the remains originated from more than 1 individual, e.g. more than 1 urosome or genital segment. To estimate total biovolume of copepods in krill stomachs, the number of individuals was multiplied by a volume of 13.4×10^6 µm^3 ind.$^{-1}$, based on the prosome length from stomach content analysis in Meyer et al. (2009), which was converted into volume according to Mauchline (1998).

Fatty acid analysis

Up to 10 individuals were pooled into 1 sample in order to obtain sufficient sample material for subsequent analyses. The frozen samples were freeze-dried for 24 h and the dry weights were determined gravimetrically. Lipids were extracted with a method modified after Folch et al. (1957) using dichloromethane:methanol (2:1, v/v). The lipids were converted into fatty acid methyl esters (FAMEs) by transesterification in methanol, containing 3% sulphuric acid, at 50°C for 12 h. The FAMEs were extracted with hexane and analysed via gas chromatography. The FAMEs were identified with known standard mixtures. The total FA content and the percentage of individual FAs were quantified with an internal standard (23:0) added prior to lipid extraction. The proportions of the individual FAs were expressed as mass percentage of the total FAs. Details on the procedure and analytical equipment were reported by Kohlbach et al. (2016).

Carbon and nitrogen analysis

Krill samples for carbon and nitrogen analysis were freeze-dried for 24 h, and were mechanically homogenized prior to analyses. Up to 5 individuals were pooled into 1 sample in order to reach a minimum sample dry weight of 1 mg. Carbon and nitrogen were then analysed using a Carlo Erba CN analyser (HEKAtech).

Bulk stable isotope analysis (BSIA)

Up to 10 individual krill were pooled into 1 sample in order to obtain sufficient sample material for analyses. Bulk stable isotope (BSI) compositions were determined with a continuous flow isotope ratio mass spectrometer Delta V Plus, interfaced with an elemental analyser (Flash EA 2000 Series) and connected via a Conflo IV Interface (Thermo Scientific). The isotopic ratios were expressed as parts per thousand (‰) in the delta notation, as deviation from the Vienna Pee Dee Belemnite standard for carbon measurements ($\delta^{13}C$), and atmospheric nitrogen for nitrogen measurements ($\delta^{15}N$). Verification of accu-

racy and precision of BSIA measurements was done by measuring the secondary reference material USGS41, provided by the International Atomic Energy Agency (IAEA, Vienna), which indicated errors as ±0.8‰ for nitrogen and ±0.5‰ for stable carbon measurements (representing ±1 SD of 17 analyses). Furthermore, the laboratory standards isoleucine and peptone were analysed every 5 samples (Sigma Aldrich), indicating errors of ±0.3‰ for nitrogen and ±0.6‰ for carbon isotope ratios of isoleucine (representing ±1 SD of 16 analyses) and ±0.3‰ for both peptone measurements (representing ±1 SD of 8 analyses). For details on the verification of accuracy and precision of the BSIA measurements, see Kohlbach et al. (2016). Samples of particulate organic matter (POM) from surface water and sea ice were collected to provide a baseline for BSIA. Measurements confirmed that the $\delta^{13}C$ values were significantly higher in sea ice-derived carbon compared to carbon derived from the water column (*t*-test, $t = 5.2$, df = 10.9, $p < 0.01$; Kohlbach et al. 2017). There also was no overlap between the $\delta^{13}C$ values of sea-ice POM and water column phytoplankton POM, confirming that these can be recognized as distinct carbon sources (Kohlbach et al. 2017).

Data analysis

The AC0 krill population was in general dominated by furcilia larvae in stage VI (FVI). The sampled population could be divided in 3 separate cohorts according to their length distribution (Schaafsma et al. 2016; see Table S1 in the Supplement at www.int-res.com/articles/suppl/m580p101_supp.pdf). The first cohort (Station [Stn] 555_47) was dominated by AC0 juveniles and contained a smaller proportion of FVI. The second cohort (Stns 557_2 to 562_5 and 571_2 to 579_2) was dominated by FVI, with negligible amounts of other developmental stages. The third cohort was dominated by FVI, but also contained significant proportions of FV and FIV (Stns 565_5 to 570_5; Fig. 1). In spite of the overlap in developmental stages between cohorts, the average length of the developmental stages differed between cohorts. For example, FVI from cohorts 1 and 2 were significantly larger than FVI from cohort 3 (Schaafsma et al. 2016). Average lengths and proportions of developmental stages in each station can be found in Schaafsma et al. (2016) and Table S1. For this study, cohort 2 was split up into groups 2a and 2b. These krill represented AC0 krill of similar length and developmental stage, but were separated by hundreds of kilometres in space and weeks in time (Fig. 1, Table 1). These 4 groups were used to investigate population-driven patterns in short- and long-term diet inferred from stomach contents, FA composition, carbon and nitrogen contents and BSI composition.

An analysis of the community structure of under-ice fauna in the sampling area suggested the presence of 3 distinctive community types, differing in the numerical and biomass composition of abundant taxa (David et al. 2017; Fig. 1). The first community type ('krill dominated'; Stns 555_47 to 557_2) was dominated by krill in terms of proportional biomass (>65%), but overall species abundances and biomasses were relatively low. The second community type ('copepod dominated'; Stns 560_2 to 567_2) had high species abundances and biomasses, and was largely dominated by copepods (>72% in terms of abundance). The third community type, comprising stations close to the sea-ice edge ('low biomass'; Stns

Table 1. Sampling dates and parameters used for BioEnv analysis per station, including environmental parameters and abundances of dominant copepods from the ice–water interface layer (0–2 m)

Station no.	Sampling date (dd-mm-yyyy)	Sea ice			Temp. (°C)	Sal.	Chl *a* (mg m^{-3})	Copepods (ind. m^{-3})		
		Cover (%)	Thickness (m)	Roughness				*Calanus propinquus*	*Ctenocalanus* sp.	*Stephos longipes*
555_47	09-09-2013	99.5	0.475	3.734	−1.85	34.3	0.104	0.09	0.11	1.04
557_2	10-09-2013	94.0	0.700	0.833	−1.86	33.9	0.134	0.06	0.20	1.25
560_2	11-09-2013	96.0	0.525	1.030	−1.86	33.8	0.108	0.08	0.67	3.28
562_5	12-09-2013	92.5	0.525	0.969	−1.86	33.8	0.097	0.47	1.75	6.63
565_5	16-09-2013	96.5	0.525	2.297	−1.87	34.2	0.103	0.46	1.33	0.79
567_2	28-09-2013	86.5	0.675	1.148	−1.88	33.6	0.204	1.19	4.66	0.07
570_5	29-09-2013	96.0	0.425	0.853	−1.86	33.9	0.223	0.30	0.03	0.02
571_2	30-09-2013	84.0	0.225	0.829	−1.84	34.1	0.165	0.07	0.02	0.07
577_2	02-10-2013	51.5	0.475	1.207	−1.84	33.7	0.164	0.01	0.00	0.01
579_2	02-10-2013	46.0	0.575	1.504	−1.83	34.1	0.275	0.03	0.00	0.06

570_5 to 579_2) was characterized by both low species abundance and low total biomass (David et al. 2017). This grouping of community types was used to investigate community-associated patterns in short- and long-term diet.

To investigate the relationship between the sea-ice environment and the krill diet variability between stations, the effect of all possible combinations of measured environmental variables on the average stomach content per station was analysed using a BioEnv analysis (Clarke & Ainsworth 1993), which evaluates the subset of environmental variables that has the highest correlation with the stomach contents. The BioEnv analysis relates 2 distance matrices, the environmental data based on Euclidean distance and the stomach content data on Bray-Curtis dissimilarity (Clarke & Warwick 2001). Environmental variables used are listed in Table 1. Using the measured environmental data, a sea-ice roughness coefficient was calculated according to David et al. (2017). The density of the most abundant copepod species (*Stephos longipes, Ctenocalanus* sp. and *Calanus propinquus*) in the under-ice surface layer were added as parameters to investigate the effect of copepods as an available food source (David et al. 2017). Stomach contents, expressed as abundance as well as volume, were fourth-root transformed to increase importance of food items that generally occur in low abundances (Clarke & Warwick 2001). After data assessment using a draftsman plot, sea-ice thickness and sea-ice concentration were square-transformed, and all other environmental data except temperature were log-transformed. After data transformation, the environmental data were normalised to obtain a consistent scale for each parameter by subtracting the mean value and dividing by the standard deviation over all samples of that parameter. This ensures equal variances of all used parameters and therefore equal importance in the analysis (Clarke & Warwick 2001). A Mantel test was used to test the significance of the association of the environmental variables selected with BioEnv with the stomach content data using Spearman's correlation. The significance of Mantel test correlations was assessed with a bootstrapping procedure using 999 iterations.

To test whether numerical stomach content composition differed between cohort groups or under-ice community types, a multivariate generalized linear model (GLM) was used. Unlike distance-based methods, this approach does not vary in detection of between-group differences depending on variance, which increases with increasing abundances (Warton et al. 2012). Differences were assessed using 999 bootstrapping iterations. Untransformed abundance data were used, and a negative binomial distribution of data was assumed (Wang et al. 2012, Warton et al. 2012). Assumptions were checked by plotting the residuals versus the fits (Wang et al. 2012).

The variability in FA compositions was assessed using a principal component analysis (PCA), including all FAs that contributed more than 1% to the total amount of the krill FAs. Proportions of FAs were fourth-root transformed to increase importance of FAs that generally occur in low proportions (Clarke & Warwick 2001). Only a single AC0 krill was sampled for FA analysis from cohort 1. Therefore this cohort is shown in the PCA analysis results but was further excluded from all FA data analyses. Differences in FA composition between cohorts and community types were tested with a distance-based analysis of similarity (ANOSIM), using fourth-root-transformed data and a Euclidean distance matrix (Clarke & Warwick 2001).

Differences in individual marker FAs, C:N ratios and BSI compositions between cohort groups and community types were investigated using 1-way ANOVA, followed by a non-parametric Tukey's HSD post hoc test. Statistical significance was set at $\alpha = 0.05$. All analyses were performed using R version 3.3.1, with packages *vegan*, *ade4*, *ggplot2* and *mvabund* (R Core Team 2015). Details on the properties of krill used for the different analyses can be found in Tables S2 & S3 in the Supplement.

RESULTS

Stomach content analysis

The diet of AC0 krill was dominated, on average, by centric (35%) and pennate (56%) diatoms in abundance, and centric diatoms (58%) and copepods (26%) in estimated volume (Fig. 2). Not all species could be identified to species level. The relative abundance of pennate diatoms in the stomachs was considerably higher in the northernmost stations compared to all other stations (Fig. 2, Table 2). The pennate diatoms were dominated by species of the genus *Fragilariopsis*. Identifiable species were *F. curta*, *F. kerguelensis*, *F. obliquecostata* and *F. ritscheri*. Identifiable species of centric diatoms were *Actinocyclus actinochilus*, *Stellarima microtrias*, *Thalassiosira tumida*, *Thalassiosira* spp. and *Coscinodiscus* spp. *Eucampia antarctica*, *Asteromphalus* spp. and *Rhizosolenia* sp. were encountered occasionally. *A. actinochilus* often represented a large part of the total reconstructed

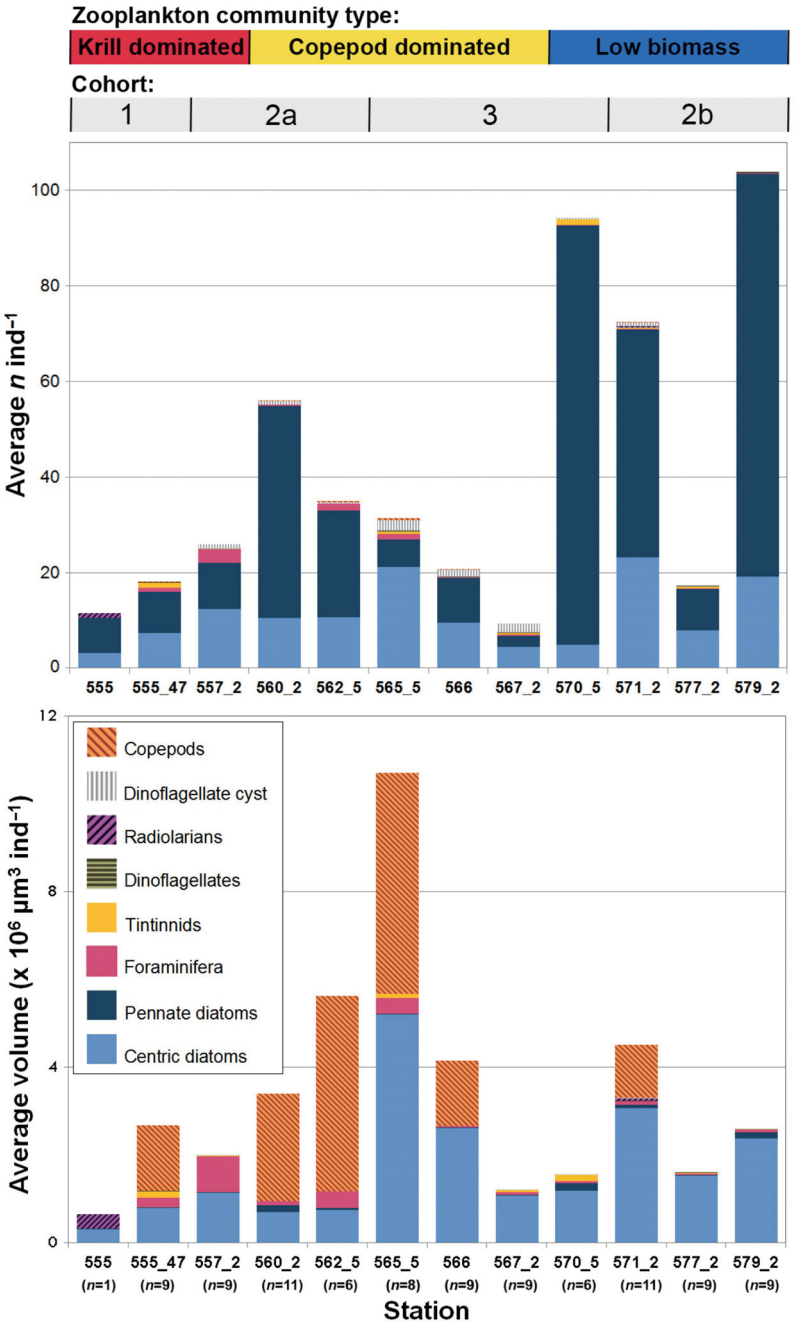

Fig. 2. Average stomach contents of age class 0 krill *Euphausia superba* at each station, shown in (A) numbers per individual krill and (B) estimated volume of food items per individual krill. Bars above the graphs show how the sampled stations were grouped in under-ice surface zooplankton community type or age class 0 krill cohorts, according to David et al. (2017) and Schaafsma et al. (2016), respectively; *n* represents the number of individuals analysed

number of centric diatoms (over 50% at Stns 555_47 – 560_2, and over 30% at Stns 570_5-579_2).

The only copepod appendages that could be identified belonged to *Stephos longipes*. Other prey items regularly found in the stomachs were the foraminifer *Neogloboquadrina pachyderma*, the tintinnids *Laackmanniella naviculaefera*, *Cymatocylis convallaria*, *Cymatocylis vanhoeffeni* and *Codonellopsis glacials*, and dinoflagellate cysts. Dinoflagellates were found in small numbers, some identifiable as *Protoperidinium* spp. and *Dinophysis* spp. Krill setae and radiolarians were found sporadically.

There were no significant differences in stomach contents between cohorts (GLM, Likelihood Ratio (LR) = 32.83, $p > 0.05$). Differences in stomach contents were partially related to under-ice community types and depended on environmental factor levels. Using the 3 community types established by David et al. (2017) as a station grouping factor, a significant difference was found between the stomach contents of krill from the low biomass community at the northern sea-ice edge versus krill from the copepod-dominated community in the centre of the sampling area (GLM, LR = 18.44, $p = 0.038$). At the centre of the sampling area, copepods also dominated the stomach contents of krill in terms of volume (Fig. 2, Table 2).

BioEnv analysis showed that the numerical composition of identifiable prey items was correlated to a combination of under-ice surface chl *a* concentration, sea-ice coverage, under-ice surface salinity and the abundance of *Stephos longipes* in the ice–water interface layer ($r = 0.47$; Mantel test $p = 0.005$). Similarly, the volumetric composition of identifiable prey items in the stomach was best correlated with the under-ice surface abundance of *S. longipes* and under-ice surface chl *a* concentration ($r = 0.58$; Mantel test $p = 0.006$).

Fatty acid analysis

Using ANOSIM, a significant difference between FA profiles was found when the AC0 krill were grouped according to cohorts ($R = 0.42$, $p = 0.001$), but

Table 2. Average stomach content composition of age class 0 krill *Euphausia superba* per under-ice zooplankton community type as established by David et al. (2017). K: krill-dominated community, C: copepod-dominated community, L: low biomass community. n = number of individuals analysed, + represents a volume < 0.01 × 10^6 µm^3 ind.$^{-1}$

Community type	K (n = 19)	C (n = 43)	L (n = 35)	Total (n = 97)
Average number (ind.$^{-1}$)				
Centric diatoms	9.45	10.97	15.02	12.14
Pennate diatoms	9.04	17.98	53.88	29.18
Foraminifera	1.74	0.58	0.2	0.67
Tintinnids	0.53	0.14	0.31	0.28
Dinoflagellates	0.16	0.12	0.2	0.15
Radiolarians	0.05	0	0.06	0.03
Dinoflagellate cysts	0.42	1.33	0.29	0.77
Unidentified round body < 20 µm	6.53	4.28	2.71	4.15
Copepods	0.05	1.05	0.34	0.6
Krill setae	1.15	0.67	1.26	0.98
Average volume (×10^6 µm^3 ind.$^{-1}$)				
Centric diatoms	2.91	3.38	4.63	3.74
Pennate diatoms	0.02	0.04	0.11	0.06
Foraminifera	0.49	0.16	0.06	0.19
Tintinnids	0.11	0.03	0	0.06
Dinoflagellates	+	+	+	+
Radiolarians	0.02	0	0.02	0.01
Dinoflagellate cysts	+	+	+	+
Unidentified round body < 20 µm	+	+	+	+
Copepods	0.71	2.49	0.38	1.38
Total volume (excluding krill setae)	5.01	8.78	5.68	6.92

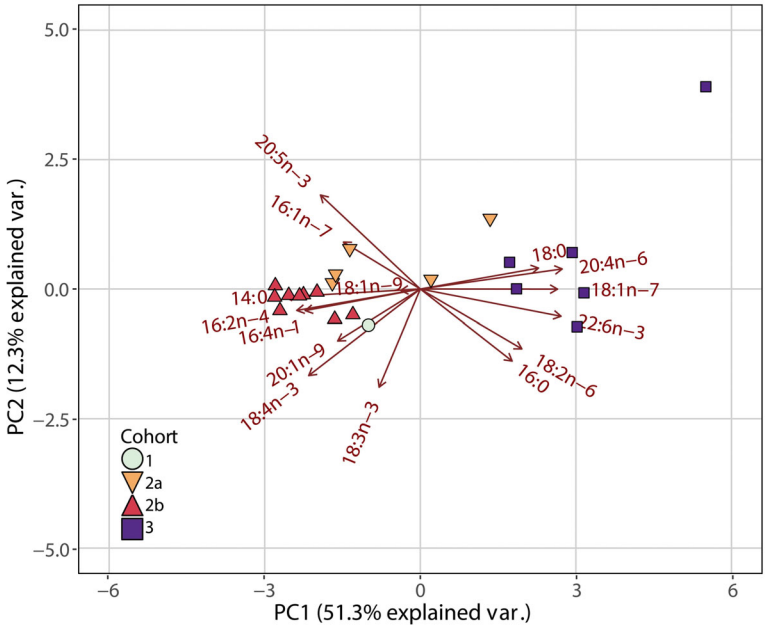

Fig. 3. Results of a principal component analysis (PCA) using fatty acid profiles of different cohorts of age class 0 krill *Euphausia superba*, showing the first and second principal components. Each point represents a replicate sample

none when they were grouped according to community types. This was confirmed by the PCA (Fig. 3). The first 3 principal components (PC) of the PCA accounted for 74.8% of the variance between the cohorts. The first PC explained 51.3% of the variability, separating cohort 3 from the other cohorts. The FAs 14:0, 16:2n-4, 16:4n-1, 20:4n-6, 20:5n-3 and 18:1n-7 contributed most to the variability in the data. The FA composition per cohort is given in Table S4 in the Supplement.

Following the results of the previous analysis, 4 biomarker FAs were compared between cohorts (Fig. 4). In cohort 3, the relative contribution of the diatom-associated marker FA 16:1n-7 was significantly lower compared to cohort group 2a (ANOVA, $F_{2,18}$ = 5.18, p = 0.02; Tukey's HSD, p = 0.01), and the diatom-associated marker FA 20:5n-3 was significantly lower compared to cohort group 2b (ANOVA, $F_{2,18}$ = 5.19, p = 0.02; Tukey's HSD, p = 0.01). Conversely, the dinoflagellate-associated marker FA 22:6n-3 was significantly higher in cohort 3 compared to the other cohorts (ANOVA, $F_{2,18}$ = 41.57, p < 0.0001, Tukey's HSD, p < 0.0001). The dinoflagellate-associated marker FA 18:4n-3 was significantly higher in cohort group 2b compared to cohorts 2a and 3 (ANOVA $F_{2,18}$ = 32.28, p < 0.0001, Tukey's HSD, p < 0.0001).

Body condition

The C:N ratios of AC0 krill ranged between 3.38 and 4.10 (Table 3). There was a significant difference between the C:N ratio of the krill from the 'copepod-dominated' community type versus krill from the other community types (ANOVA, $F_{2,24}$ = 10.81, p < 0.0001; Tukey HSD, p < 0.004). However, testing for differences between cohort groups within community types indicated that these differences could be explained by differences between cohort groups. C:N ratios of AC0 krill differed significantly between all 4 cohort groups (ANOVA, $F_{3,23}$ = 26.6, p < 0.0001; Tukey HSD, p > 0.04), decreasing from cohort group 1 to cohort group 3. The C:N ratio of

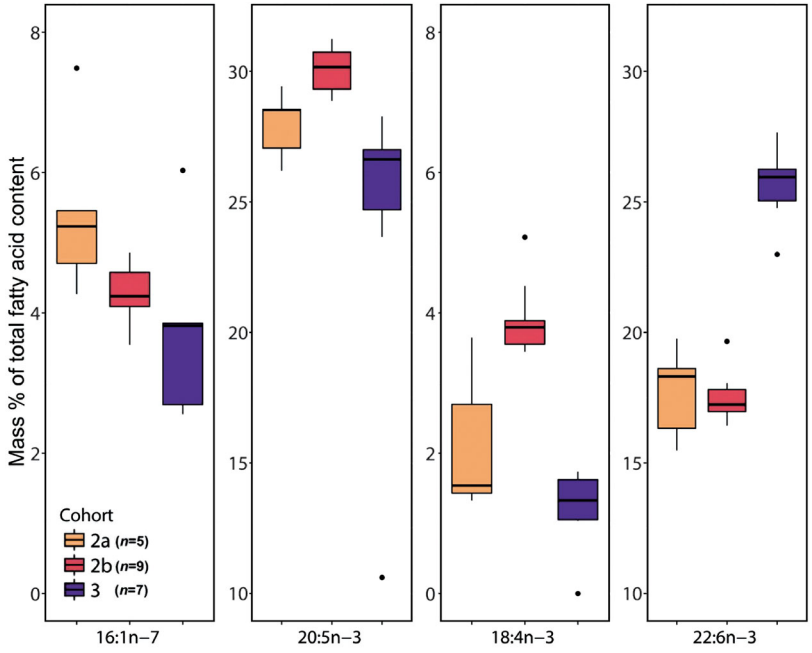

Fig. 4. Proportion of biomarker fatty acids (mass % of total FA) of age class 0 krill *Euphausia superba*. Cohorts are defined as in Fig. 1. FAs 16:1n-7 and 20:5n-3 are considered diatom-associated markers, 18:4n-3 and 22:6n-3 are dinoflagellate-associated markers. The horizontal black lines show the median FA proportion in a cohort. The upper and lower limits of the coloured squares indicate the 25th and 75th percentiles. The upper and lower limits of the vertical line indicate the minimum and maximum FA proportions in a cohort excluding the outliers (represented by dots), which are numbers that are 1.5 times less than or greater than the lower or upper percentiles, respectively

Table 3. Average carbon content, nitrogen content, C:N ratio and total fatty acid (FA) content (SD within brackets) of age class 0 krill *Euphausia superba* per cohort; na: not analysed

Cohort	Carbon content (% of dry mass)	Nitrogen content (% of dry mass)	C:N ratio	Total FA content (% of dry mass)
1	40.60 (0.64)	10.12 (0.27)	4.01 (0.07)	na
2a	39.06 (1.13)	10.59 (0.40)	3.69 (0.07)	12.48 (11.81)
2b	39.32 (1.06)	10.20 (0.21)	3.86 (0.09)	19.50 (14.55)
3	34.61 (1.00)	9.81 (0.19)	3.53 (0.10)	2.63 (2.09)

cohort group 2b was significantly higher than that of cohort group 2a (Tukey HSD, p = 0.005). A similar pattern was found in the total FA content of the AC0 krill (Table 3). There were, however, no significant differences in total FA content between cohort groups.

BSI composition

The $\delta^{15}N$ value of AC0 krill differed significantly between both community types and cohorts (ANOVA, $F_{2,27} = 16.86$, $p < 0.001$ and $F_{3,26} = 29.47$, $p < 0.001$, respectively). Again, further analysis indicated that the cohort grouping explained the differences more robustly. Apart from cohort 1 vs. cohort 2b, $\delta^{15}N$ values differed significantly between cohort groups (Fig. 5; ANOVA, $F_{3,26} = 29.47$, $p < 0.0001$; Tukey HSD, $p < 0.02$). The average $\delta^{15}N$ value in cohort 3 (2.41‰) was lowest. In this cohort, $\delta^{15}N$ values did not exceed 3‰. The average $\delta^{15}N$ values of cohort 1 (3.72‰) and 2b (4.05‰) were significantly higher than in cohort 2a (3.24‰; Fig. 5; Tukey HSD < 0.02). The $\delta^{13}C$ values of cohort 3 (average −26.8‰) were significantly lower than all values of cohort groups 1, 2a and 2b (average −25.1, −24.5 and −24.5‰, respectively; ANOVA, $F_{3,26} = 17.92$, $p < 0.001$, Tukey HSD, $p < 0.003$). The $\delta^{13}C$ values did not show significant differences when the krill were grouped according to community type.

DISCUSSION

Stomach contents and FAs of AC0 krill in winter/early spring

The stomach contents of AC0 krill showed a variable diet in terms of taxonomic composition. In general, the diet of larvae was numerically dominated by diatoms, in particular the pennate species *Fragilariopsis* spp., and had a heterotrophic component consisting of foraminiferans, tintinnids, dinoflagellates, dinoflagellate cysts and copepod appendages. This is consistent with findings of winter studies conducted in the Weddell–Scotia Confluence (Daly 1990) and in the Lazarev Sea (Meyer et al. 2009, Schmidt et al. 2014), although the scale of our study enables us to show that the degree of utilization of these food sources varies within a region and correlates with environmental factors.

The importance of heterotrophic taxa in the diet may be under-estimated by stomach content analysis, because soft-bodied organisms such as flagellates, ciliates and turbellarians are easily digested and therefore unlikely to be found in the stomachs of the AC0 krill (Meyer et al. 2009). Studies suggest that

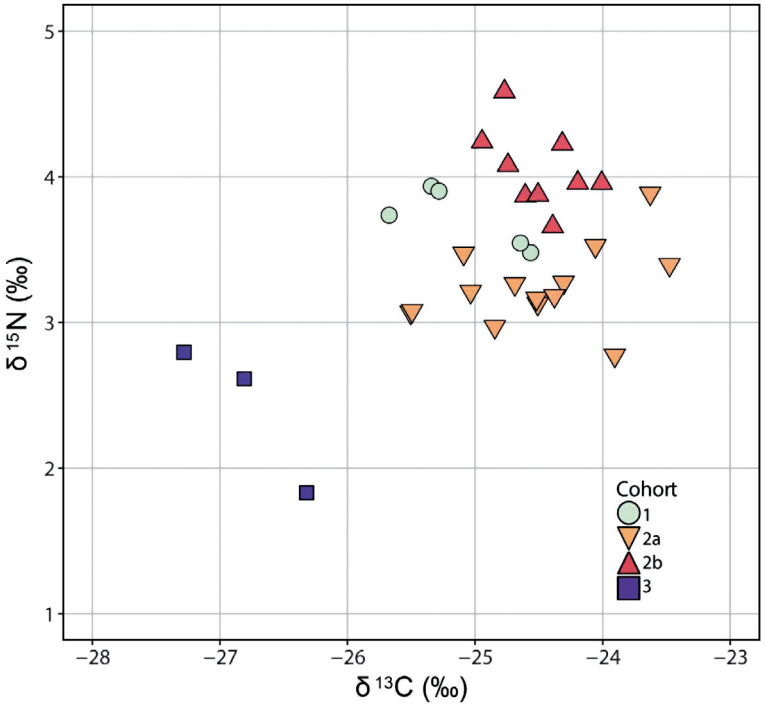

Fig. 5. Bulk stable isotope values of age class 0 krill *Euphausia superba* per cohort. Cohorts are defined as in Fig. 1. Every point represents a replicate sample

detritus may provide an additional food source for furcilia (Daly 1990, Ju & Harvey 2004), but no further analysis was done on unidentifiable stomach items during this study. The lack of copepod mandibles in the AC0 krill stomachs could be an indication that the copepod appendages found originated from moults. The reconstructed volume of the copepods in the stomachs could therefore represent an overestimate, although feeding on copepods during winter is common (Meyer et al. 2009, Töbe et al. 2010). Moreover, moults were sparse in the SUIT samples compared to live copepods (David et al. 2017), indicating a low encounter probability in the under-ice layer. This is probably due to the fact that moults tend to sink quickly and therefore only stay in the under-ice habitat for a very short time (Frangoulis et al. 2005).

In general, the FAs of all AC0 krill were dominated by 16:0, 20:5n-3 and 22:6n-3, similar to larval krill from East Antarctica (O'Brien et al. 2011, Virtue et al. 2016) and the Lazarev Sea (Hagen et al. 2001) in winter/early spring, and the western Antarctic Peninsula in winter (Ju & Harvey 2004). FAs are typically components of different classes of lipids. The marker FAs 20:5n-3 and 22:6n-3 are mainly incorporated into phospholipids (Hagen et al. 2001). The phospholipid phosphatidylcholine was the most dominant lipid class found in the AC0 krill from our study (Kohlbach et al. 2017), explaining the high proportions of these FAs. While phospholipids usually represent biomembrane components, phosphatidylcholine also serves as a storage lipid for *Euphausia superba* (Hagen et al. 1996). The marker FAs 16:1n-7 and 18:4n-3 are mainly incorporated into other storage lipids (Stübing et al. 2003).

Sea-ice association of prey

Many of the identified species in the krill stomachs of our study were sea ice-associated species. *Actinocyclys actinochilus* has been found in higher abundances within sea ice compared to the underlying water column (Armand et al. 2005). *Fragilariopsis* spp. such as *F. curta* and *F. cylindrus* often dominate the sea-ice algal assemblage (Nöthig et al. 1991, Garrison & Close 1993, Ugalde et al. 2016). Dinoflagellate cysts can be abundant in sea ice, and it has been proposed that sea ice is an overwintering site for resting or dormant stages (Garrison & Buck 1989). The copepod *Stephos longipes* migrates actively between the water column and sea-ice habitats, and the presence of this copepod in the water column is concomitant with their presence in the sea ice above (Wallis et al. 2016). Abundances of juvenile and adult *S. longipes* were highest in the sea ice during winter/early spring (Schnack-Schiel et al. 1995, Mauchline 1998).

The high proportional contribution of sea ice-associated species found in the stomachs of AC0 krill suggests that they were largely relying on sea ice-associated prey during winter. This was confirmed by the $\delta^{13}C$ values, which suggested that the AC0 krill had continuous access to sea ice-associated food sources (Kohlbach et al. 2017).

Relationship between diet and the environment

The stomach contents, reflecting the recent diet, were related to environmental factors, such as under-ice surface chl *a* concentration and the abundance of copepods present, sea-ice concentration, and under-ice surface water salinity. The results on the propor-

tions of carbon, nitrogen, BSI and FAs of individual krill at any point in time is a reflection of integrated conditions over a period of days to months prior to collection (Daly 2004, Graeve et al. 2005, Töbe et al. 2010). FA and BSI thus reflect the diet over a longer term which explains why their variability could not be attributed to environmental factors measured during sampling.

Stomach contents of AC0 krill in the central part of the study area contained considerable volumes of copepods. At these stations, the highest abundances of copepods were found in the ice–water interface layer, dominated by *S. longipes* and *Ctenocalanus* spp. (David et al. 2017). The central part of the study area was further characterized by a high biomass zooplankton community in the ice–water interface, consisting of amphipods, pteropods, chaetognaths and ctenophores, indicating a diverse heterotrophic food web (David et al. 2017). Exceptionally, the stomach content of AC0 krill from Stn 567_2 had a small total volume, and no copepods were found in the krill stomachs, despite their abundance in the water column (David et al. 2017). This could be due the extreme dominance of *Ctenocalanus* spp. at this station (Table 1), which are not a food source of larval krill (Töbe et al. 2010).

The under-ice zooplankton community structure at the 4 northernmost stations was characterized by low abundances and biomass of species compared to the rest of the sampling area (David et al. 2017). These stations were further characterized by relatively higher under-ice surface chl *a* concentrations and limited ice floe size (David et al. 2017). This suggests that the sea ice had started to melt. The increase in pennate diatoms in the stomach contents of AC0 krill is therefore likely a result of residing closer to the sea-ice edge where the sea ice started to release its contents (Ackley et al. 1979), and/or a phytoplankton bloom started (Quetin & Ross 1991, Bianchi et al. 1992). Alternatively, it is possible that sea-ice algae became more easily accessible as the sea ice began to soften and became more porous due to melt (Quetin et al. 2003).

Our findings suggest that the diet of AC0 krill is a reflection of the food available and accessible in the environment. Therefore, seasonal and biogeographical patterns in food availability govern the diet of AC0 krill over the short term. Food availability, in turn, is dependent on environmental factors driven by the sea ice, which can be the properties of the sea ice itself, but also other effects, such as the increase in chl *a* concentration in the water column due to melting of the sea ice.

Relationship between diet and size/developmental stage

The FA composition of furcilia is markedly influenced by their food composition (Stübing et al. 2003). FA and lipid signatures may reflect different food sources and, in omnivorous species, ingestion of both phytoplankton and zooplankton, which can complicate the interpretation of trophic relationships (Mayzaud et al. 1999, Auel et al. 2002, Dalsgaard et al. 2003). It must be considered that the relative FA composition can depend on total lipid content (Stübing et al. 2003). The total FA content of larvae and AC0 juveniles from our study was highly variable between individuals, which Virtue et al. (2016) previously attributed to the patchiness of the available food. Nevertheless, the AC0 krill in this study showed distinct FA profiles, BSI values and C:N ratios, indicating that the dietary history of the various cohort groups was different and is related to differences in size and development. The lack of relationship between stomach contents and cohort groups suggests that there was no size restriction in the utilization of various prey.

The time scales for the incorporation of carbon, nitrogen and different FAs into tissues as well as their turnover rates are often not well defined (Dalsgaard et al. 2003). However, FAs incorporated in storage lipids are assumed to reflect a more recent carbon source composition compared to FAs incorporated in membrane components such as phospholipids (Stübing et al. 2003). Therefore, the differences found between cohort groups during this study using a variety of analyses suggest that the availability and/or utilization of food sources changed over time.

Larger juvenile krill from cohort 1 were in good condition despite low stomach content volume, indicating that rapid development to the juvenile stage may be advantageous for survival (Feinberg et al. 2006). These findings also support the idea that the ability to withstand poor food conditions increases with age (Daly 2004).

Despite their similar size and developmental stage, cohort groups 2a and 2b showed some differences in several analyses. This suggests that they encountered distinct environmental conditions during advection from their spawning area or areas (Schaafsma et al. 2016). The krill from cohort group 2b had a higher C:N ratio than those from cohort group 2a, suggesting that they were in better condition, likely due to ice edge feeding, as would be expected at the beginning of a spring bloom of ice algae and phytoplankton.

The relatively high proportion of the dinoflagellate-associated marker FA 18:4n-3 in cohort 2b suggests a relative increase in feeding on dinoflagellates at the end of the sampling season. A similar enhanced feeding on dinoflagellates during the winter/spring transition was also found in East Antarctica, based on FA analysis (Virtue et al. 2016). While the aforementioned study suggests that diatoms were not a major food source during this time of year (Virtue et al. 2016), our cohort 2b had a relatively high proportion of the diatom-associated marker FA 16:1n-7 and on average the highest number of diatoms in their stomachs. The proportion of FA 16:1n-7 was also similar to that of AC0 krill from cohort 2a, caught earlier in the season. Possible explanations for contradictions between FA and stomach content analyses of cohort 2b are increased feeding on athecate (naked), easily digested dinoflagellates, and/or that the increased feeding on diatoms had occurred only recently.

Cohort 3 had relatively low proportions of the diatom-associated marker FAs 16:1n-7 and 20:5n-3, indicating that diatoms had a consistently lower contribution to the diet of this cohort compared to the other cohorts (Reiss et al. 2015, Virtue et al. 2016). Additionally, the krill from cohort 3 also had smaller amounts of the FA 16:4n-1 which has also been found to be an important FA for diatoms (Dalsgaard et al. 2003). The relatively low proportion of the dinoflagellate-associated marker FA 18:4n-3 in cohort 3 either indicates that dinoflagellates were less important in the more recent period before the sampling, or that the krill from cohort 3 have recently been starving. This FA metabolizes rapidly, and is found to decrease when not replaced by new dietary input (Stübing et al. 2003). The relatively large amount of the dinoflagellate-associated marker FA 22:6n-3 in the krill of cohort 3 could be a result of their relative high proportion of the phosphatidylcholine compared to other cohorts (Kohlbach et al. 2017). However, FA 20:5n-3, also usually incorporated in PC, was lowest in the krill of cohort 3, strongly indicating that AC0 krill from cohort 3 had fed more extensively on dinoflagellates in the more distant past compared to the other cohort groups. Based on the larger proportion of dinoflagellates often residing in the water column as opposed to the sea ice (Garrison 1991, Lizotte 2001), this suggests that feeding in the more distant past occurred to a larger extent on pelagic resources, which are scarce during winter. The relatively low $\delta^{15}N$ value suggests that AC0 krill from cohort 3 were feeding predominantly herbivorously in the past, while the other cohorts were feeding more omnivorously. This was based on the mostly low $\delta^{15}N$ values in sea-ice and pelagic POM (Kohlbach et al. 2017). Results show that compensating a lack of sea-ice resources with heterotrophic pelagic food sources, as seemed to be the case during 1 year in East Antarctica (Jia et al. 2016), is not a general pattern in the Southern Ocean during winter. The combined results, including the relatively small size and lower C:N ratio of cohort 3, strongly suggest advection through regions with poor food availability, probably related to regional properties of the sea-ice habitat as supported by different $\delta^{13}C$ values.

Conclusion

During winter in the northern Weddell Sea, sea ice-associated prey were crucially important in the diet of AC0 *E. superba.* Data mirrored patterns of local food availability, influenced by the sea-ice environment. Differences in size and development of AC0 krill are a result of differences in the earlier food availability.

This study shows that there is considerable temporal and spatial variation in the diet of AC0 krill within a season, and adds insight on how this can relate to the environment and the condition of the krill. Dietary differences found between groups in variable physiological states indicate that the long-term availability of sea-ice resources during advection over winter could have a significant influence on the condition of AC0 krill. The potential of the sea-ice habitat to sustain sufficiently productive sea-ice algae communities may be an important factor for AC0 krill to survive their first winter. Further investigation of the relationship between diet, environmental factors and food availability can improve our understanding of AC0 krill overwintering. A better understanding of within-season and annual variations will help to predict the consequences of environmental change.

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The following supplement accompanies the article

Spatio-temporal variability in the winter diet of larval and juvenile Antarctic krill, *Euphausia superba,* in ice-covered waters

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Additional information on the sampled population of AC0 Antarctic krill (*Euphausia superba*), the krill used for various analyses and the fatty acid composition.

Table S1: Average length (mm) of different stages of *Euphausia superba* furcilia larvae (F) and age class 0 juveniles (JUV) per station. Additionally the proportion (%) of the developmental stages in the catch per station is presented. The remainder of the proportion per station consists of sub-adult and adult krill (not shown). Station numbers are shaded to indicate which stations belong to the same cohort.

	Stage							
	FIV		FV		FVI		JUV	
Station	mm	%	mm	%	mm	%	mm	%
555_47					13.64	24.3	16.35	75.7
557_2			11.23	0.9	12.10	68.7	16.36	29.8
560_2					11.14	88.0	17.85	8.7
562_5			8.69	3.0	10.13	88.0	18.19	2.8
565_5	5.79	19.3	6.76	38.9	8.23	41.8		
567_2	6.36	8.9	7.03	29.6	8.78	60.7	18.00	0.7
570_5	6.49	5.2	7.02	25.8	9.54	59.6	16.87	5.9
571_2	7.32	2.4	8.06	10.0	10.46	84.9	15.30	2.6
577_2					11.18	88.6	15.90	9.2
579_2	7.95	1.7	8.36	8.4	10.82	81.1	15.05	7.4

Table S2: Number of individuals (*n*), developmental stages and average length of AC0 *Euphausia superba* used for stomach content analysis. FVI indicate furcilia larvae in stage six, Juv are juveniles in their first winter. The standard deviation is given within brackets.

Station	*n*	Stages	Average length (mm)	Station	n	Stages	Average length (mm)
555	1	Juv	16	566	9	FVI	8.78 (1.0)
555_47	9	FVI, Juv	14.11 (1.9)	567_2	9	FVI	9.33 (0.8)
557_2	9	FVI, Juv	15 (1.7)	570_5	6	FVI, Juv	13.5 (1.8)
560_2	11	FVI, Juv	15.62 (4.8)	571_2	11	FVI, Juv	14 (2.3)
562_5	6	FVI	11.17 (0.90)	577_2	9	FVI, Juv	13 (2.3)
565_5	8	FVI	9.71 (1.0)	579_2	9	FVI	13.08 (1.1)

Table S3: number of measured replicates (*n*), total number of individuals used, developmental stages, average length and average dry weight of AC0 *Euphausia superba* used for carbon/nitrogen, fatty acid and bulk stable isotope analysis. FV and FVI indicate furcilia larvae in stage five and six, Juv are juveniles in their first winter. The standard deviations of length and dry weight are given within brackets.

Cohort	*n*	Total number of individuals	Stages	Average length mm	Average DW mg
Carbon and nitrogen content					
1	5	5	FVI, Juv	17.98 (2.39)	6.74 (2.96)
2a	11	26	FVI	13.09 (2.56)	3.13 (1.58)
2b	7	20	FVI	11.36 (1.14)	1.69 (0.50)
3	4	16	FV, FVI	8.91 (0.30)	1.29 (1.06)
Fatty acids and total fatty acid content					
2a	5	31	FVI, Juv	12.37 (3.69)	2.94 (2.0)
2b	9	75	FVI, Juv	10.16 (3.57)	1.78 (1.8)
3	7	50	FV, FVI, Juv	10.35 (3.49)	1.80 (1.5)
Bulk stable isotopes					
1	5	5	FVI, Juv	17.98 (2.4)	6.74 (3.0)
2a	14	51	FVI, Juv	13.09 (2.6)	3.13 (1.6)
2b	9	36	FVI, Juv	11.36 (1.1)	2.49 (1.6)
3	3	17	FVI	9.07 (0.03)	2.22 (1.1)

Table S4: Average fatty acid composition of age class 0 *Euphausia superba* per cohort, expressed as average % of total fatty acids. The standard deviation is given in brackets. *n* represents the number of replicates measured.

	Cohort			
	1	2a	2b	3
Fatty acid	*n* = 1	*n* = 5	*n* = 9	*n* = 7
14:0	5.86	4.38 (1.0)	5.08 (0.3)	2.13 (0.2)
i-15:0	0	0	0	0
a-15:0	0	0.04 (0.1)	0	0
15:0	0	0.11 (0.1)	0.06 (0.1)	0
16:0	19.16	17.21 (0.5)	16.54 (0.4)	17.84 (1.9)
16:1(n-7)	6.31	5.43 (1.1)	4.28 (0.4)	3.64 (1.1)
16:1(n-5)	0	0.08 (0.1)	0.17 (0.3)	0.06 (0.1)
16:2(n-4)	2.82	1.71 (1.1)	1.42 (0.4)	0.19 (0.3)
16:3(n-4)	0	0.16 (0.2)	0.29 (0.6)	0
16:4(n-1)	0.80	0.97 (0.6)	1.61 (0.6)	0.13 (0.3)
18:0	1.4	1.08 (0.1)	1.08 (0.3)	1.81 (0.5)
18:1(n-9)	8.42	6.46 (1.5)	5.83 (0.7)	6.78 (3.4)
18:1(n-7)	6.44	7.00 (0.6)	5.64 (0.1)	7.86 (0.8)
18:1(n-5)	0	0.06 (0.1)	0.02 (0.1)	1.17 (2.7)
18:2(n-6)	2.30	2.25 (0.1)	2.10 (0.2)	2.50 (0.4)
18:3(n-6)	0	0.166 (0.2)	0.35 (0.2)	0
18:3(n-3)	0.82	0.39 (0.3)	0.93 (0.1)	0.71 (0.3)
18:4(n-3)	2.10	2.13 (0.9)	3.91 (0.5)	1.20 (0.6)
20:0	0	0	0	0
20:1(n-9)	0.60	1.06 (0.2)	1.23 (0.5)	0.61 (0.4)
20:1(n-7)	0	0.10 (0.1)	0	0
20:2(n-6)	0	0.10 (0.1)	0.04 (0.1)	0
20:3(n-6)	0	0.10 (0.1)	0	0
20:4(n-6)	1.02	1.31 (0.3)	0.80 (0.0)	2.09 (0.2)
20:3(n-3)	0	0.06 (0.1)	0.07 (0.2)	0
20:4(n-3)	0	0.38 (0.3)	0.53 (0.2)	0.16 (0.3)
20:5(n-3)	24.90	27.95 (1.2)	30.02 (0.8)	24.13 (5.7)
22:1(n-11)	0	0.63 (0.7)	0.03 (0.1)	0.05 (0.1)
22:1(n-9)	0	0.38 (0.6)	0.03 (0.1)	0
22:5(n-3)	0	0.60 (0.5)	0.44 (0.2)	0.22 (0.3)
22:6(n-3)	17.04	17.70 (1.6)	17.49 (0.9)	26.72 (3.0)
24:1(n-9)	0	0	0	0