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ORIGINAL PAPER

Feeding ecology and energetics of the Antarctic chaetognaths *Eukrohnia hamata*, *E. bathypelagica* and *E. bathyantarctica*

Svenja Kruse · Wilhelm Hagen · Ulrich Bathmann

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Abstract Ecological and physiological studies focused on dietary preferences, lipid biochemistry and energetics within the three Antarctic chaetognaths *Eukrohnia hamata*, *E. bathypelagica* and *E. bathyantarctica* from meso- and bathypelagic depths. *Eukrohnia hamata* and *E. bathypelagica* respired 0.15 μL O_2 mg dry mass $(DM)^{-1}$ h^{-1}, which translates to an average metabolic loss of only <1.1% of body carbon per day. Lipid storage was not substantial in *E. bathypelagica* (mean 11.5 ± 6.5% DM) and *E. bathyantarctica* (mean 15.4 ± 4.1% DM) during summer and winter, suggesting year-round feeding of these predators mainly on copepods. In *E. bathypelagica*, total fatty acids were dominated by the fatty acids 16:0, 20:5(n-3) and 22:6(n-3) and in *E. bathyantarctica* also by 18:1(n-9), a fatty acid usually found in storage lipids. Only the latter species was characterized by significant amounts of wax esters, consisting largely of the common fatty alcohols 16:0, 20:1(n-9) and the unusual fatty alcohol isomer 22:1(n-9).

Introduction

Chaetognatha are a small phylum that consists of about 150 species (Kapp 2004). They are known as important predators in all oceans, including the Southern Ocean (Pakhomov et al. 1999) and may contribute substantially to total zooplankton abundance and biomass (Hosie and Cochran 1994; Pakhomov et al. 1999, 2000). As important predators of copepods (Øresland 1990, 1995) and as a significant food source for a wide variety of larger organisms, they hold a central position in planktonic food webs (Feigenbaum 1991).

Intensive studies especially on the taxonomy, abundance and distribution of Antarctic chaetognaths have been carried out in the past 100 years (e.g. Ritter-Záhony 1911; David 1958; Alvariño et al. 1983a, b; Hagen 1985; Kruse et al. 2009). Other than their diet composition, there was limited information on the feeding behavior of chaetognaths due to the difficulty to keep these delicate animals alive in the laboratory (Feigenbaum and Maris 1984; Feigenbaum 1991). One of the first attempts to systematically analyze the gut content of chaetognaths was made by Wimpenny (1936) on *Sagitta setosa* and *S. elegans*. A first report of controlled laboratory feeding was given by Reeve (1964) on *Sagitta hispida*. Subsequent studies also primarily focused on the species-rich genus *Sagitta*. In contrast, our knowledge on the feeding ecology of the genus *Eukrohnia* is rudimentary, except for the abundant *E. hamata* (e.g. Øresland 1990; Froneman et al. 1998). To date, there are no studies available on the feeding of Antarctic meso- and bathypelagic chaetognaths.

Gut contents and fatty acid trophic markers were analyzed to investigate the feeding preferences of *Eukrohnia hamata* and the two deeper-living species *E. bathypelagica* and *E. bathyantarctica*. The trophic marker concept is based on the largely unmodified incorporation of dietary fatty acids into the body lipid of zooplankton (e.g. Lee et al. 1971b). In our study, this observation was used to estimate the chaetognath dietary composition. It is the first application to the

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S. Kruse (✉) · U. Bathmann
Alfred Wegener Institute for Polar and Marine Research,
Am Handelshafen 12, 27570 Bremerhaven, Germany
e-mail: Svenja.Kruse@awi.de

W. Hagen
Marine Zoology, University of Bremen, P.O. Box 33 04 40,
28334 Bremen, Germany

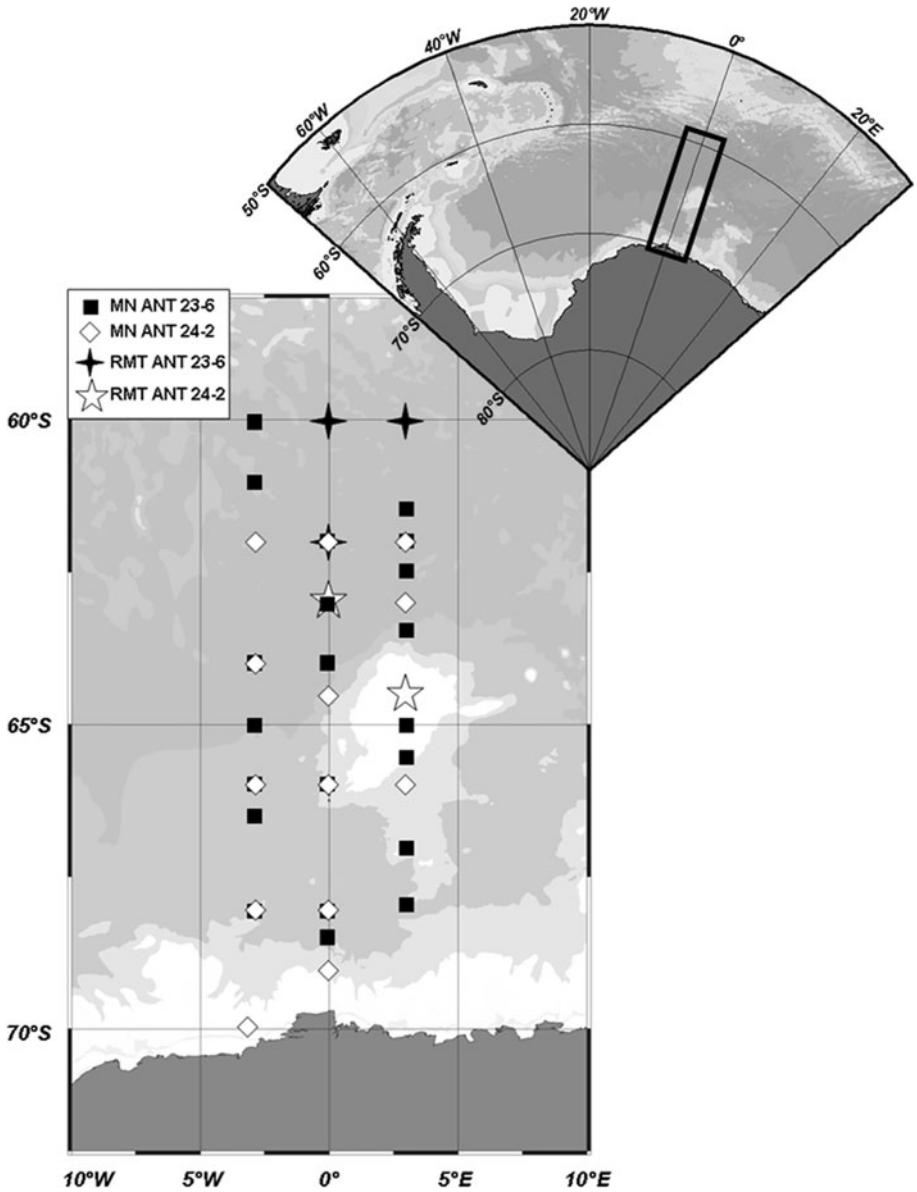

Fig. 1 Map of stations along three transects during the expeditions ANT 23-6 (winter) and ANT 24-2 (summer) in the Weddell Sea, Southern Ocean. *MN* multinet, *RMT* rectangular midwater trawl, *black* winter stations, *white* summer stations

deeper-living *Eukrohnia* species. As information on the metabolism and energy budget in this genus is limited (Båmstedt 1979; Thuesen and Childress 1993), we studied lipids and respiration of the three chaetognath species. Our aim was to elucidate the feeding habits and lipid dynamics of these meso- and bathypelagic predators.

Materials and methods

Sampling

Chaetognaths were sampled with RV "Polarstern" in the Weddell Sea during the Antarctic winter 2006 (17 June–21 August 2006, ANT 23-6) and the Antarctic summer 2007/2008 (28 November 2007–04 February 2008, ANT 24-2). These two expeditions were carried out within the scope of the German krill project LAKRIS (Meyer 2005). The study area was located between 60°–70°S and 3°W–3°E (Fig. 1). During winter, samples were taken at 28 stations (with eight repetitive stations at ~66°S 0°E) with a multinet (MN: five nets, 100 µm mesh size, 0.25 m^2 mouth area) and at three stations with a rectangular midwater trawl (RMT 8: 4.5 mm mesh size, 8 m^2 mouth area; RMT 1: 320 µm mesh size, 1 m^2 mouth area). In summer, the MN was deployed at 15 stations (two at 52°S 0°E, not shown on Fig. 1) and a multiple RMT, consisting of three pairs of nets, at four stations (two analyzed in this study). The MN sampled the following standard depth intervals during both seasons: 2,000–1,500–1,000–750–500–0 m (except for two stations to 3,000 m, one to 1,250 m and one to 1,500 m; see Kruse et al. 2009). For this study, only chaetognaths

from below 500 m depth were analyzed. The winter RMT hauls included the depth range from the surface to approximately 3,000 m and back to the surface, whereas the multiple RMT was deployed to an opening depth at 1,900 and 2,500 m, respectively (at 64°30′S 3°E: 1,900–1,500–750–500 m; at 63°S 0°E: 2,500–2,000–1,000–500 m; Fig. 1). The RMT 8 cod-end bucket carried a volume of approximately 26 l of water, which ensured that the animals were suspended. This method yielded high survival rates and healthy specimens.

Prior to the experiments and analyses, all chaetognaths were identified using the relevant literature (Alvariño 1969; Casanova 1999). They were assigned to maturity stages and measured (head to tail, excluding tail fin) to the nearest 0.5 mm under a stereomicroscope (Olympus SZX12) either on board or later in the home laboratory. *Eukrohnia bathypelagica* and *E. bathyantarctica* destined for lipid analysis were classified into maturity stages according to Alvariño (1967, 1969). Specimens carrying brood sacs (only empty brood sacs in the investigated specimens) were assigned to an additional fifth stage.

Chaetognaths of the species *E. hamata*, *E. bathypelagica* and *E. bathyantarctica* were proven to be in good condition in the MN samples. They were either quickly rinsed with Milli-Q water and stored in glass vials at −80°C for biochemical analysis or maintained alive in respiration experiments. A few specimens of *E. bathypelagica* and *E. bathyantarctica* were also taken from the RMT for the lipid and fatty acid analyses. The remaining MN and RMT samples were either frozen or preserved in formaldehyde (4% final concentration, buffered with hexamine) for later investigations.

Gut content analysis

Gut contents of formaldehyde-preserved and frozen chaetognath specimens from MN and RMT samples were analyzed. Two hundred specimens of each species (*Eukrohnia hamata*, *E. bathypelagica*, *E. bathyantarctica*) were investigated from both summer and winter season. Each chaetognath was transferred to a few drops of glycerine on a microscope slide. The presence of lipid droplets in the gut was recorded. The gut fullness was estimated prior to dissection as percentage of total gut length filled with content and divided into four categories (<10, 10–25, 25–50, 50–100%). When ripe ovaries filled the body cavity of *E. bathypelagica*, gut fullness was determined after dissection of the specimen. Jellyfish remains found in the guts were not included for the determination of gut fullness. The gut was dissected with fine needles (0.15 mm) under a stereomicroscope and further analyzed under an inverted microscope (Axiovert 40C; up to 400× magnification). The percentage of *Eukrohnia* specimens containing food (food containing ratio = FCR) and the number of prey per chaetognath (NPC) were recorded for each species. The FCR is given as total FCR when the gut contained food, even if it was an unidentifiable mass. The FCR for identifiable prey was additionally calculated. Prey items found in the foregut were omitted from the analysis to avoid bias from possible cod-end feeding.

Analyses of dry mass, elemental and biochemical composition

For dry mass (DM), carbon (C) and nitrogen (N) analyses the chaetognaths of the three species *Eukrohnia hamata* (summer: $n = 72$, winter: $n = 179$), *E. bathypelagica* (summer: $n = 40$, winter: $n = 43$) and *E. bathyantarctica* (summer: $n = 42$, winter: $n = 35$) from MN and RMT samples were freeze-dried for 24 h and weighed on a Sartorius microbalance (Sartorius Micro and Sartorius Supermicro 4504 MP8). Each specimen was ground and then analyzed (entire animal or subsample) on a Euro Elemental Analyzer for C and N compositions with acetanilide as standard.

The lipid and fatty acid analyses focused on the two meso- and bathypelagic species *E. bathypelagica* and *E. bathyantarctica*. *Eukrohnia bathypelagica* individuals originated from 500 to 2,000 m depth, whereas *E. bathyantarctica* individuals were mostly taken from 1,500 to 2,000 m depth. To ensure sufficient biomass for lipid analysis, some individuals were pooled to obtain more than 1 mg DM per sample. Total lipid was then extracted from single lyophilized individuals or pooled samples with dichloromethane/methanol (2:1 by volume), and the lipid content was determined gravimetrically according to Folch et al. (1957) as modified by Hagen (2000).

Aliquots of the extracted samples were taken for characterization and quantification of the fatty acid and alcohol fractions by gas–liquid chromatographic analysis. Lipids were hydrolyzed, and the fatty acids were converted to their methyl ester derivatives in methanol containing 3% concentrated sulfuric acid at 80°C for 4 h (Kattner and Fricke 1986). The resulting fatty acid methyl esters and free fatty alcohols were separated and quantified using a Hewlett-Packard gas chromatograph (HP 6890A), equipped with a DB-FFAP column (30 m length, 0.25 mm inner diameter). Peaks were identified by comparing retention times with those from fish oil (Marinol) and copepod lipid lab standards of known composition, verified with commercially available standard mixtures (e.g. Matreya). If necessary, additional confirmation was carried out by gas chromatography–mass spectrometry. The proportions of wax esters relative to total lipid were estimated for

E. bathyantarctica on the basis of the fatty alcohol to fatty acid contents.

The trophic ratio [18:1(n-9) + 20:1(n-9) + 22:1(n-9)]/[16:1(n-7) + 18:1(n-7) + 18:4(n-3)] was calculated as an indicator of carnivory, where the first three fatty acids are markers for carnivory and calanoid copepod ingestion, respectively, and the three latter fatty acids for phytoplankton ingestion (diatoms and flagellates, respectively; e.g. Falk-Petersen et al. 1990; Dalsgaard et al. 2003). Fatty acids that occurred with a portion of ≥1% of total fatty acids in at least one case during both seasons are presented in the results. The remaining minor fatty acids were combined to a single group.

Respiration rate measurements

Specimens from the 500 to 1,000 m depth interval (multinet samples only) were chosen for the respiration experiments with *Eukrohnia hamata* and *E. bathypelagica* to ensure good physiological conditions of the chaetognaths and to minimize decompression damage. Additionally, individuals from the 1,000 to 1,500 m depth range were included in eight analyses on *E. hamata* in winter and in four analyses on *E. bathypelagica* in summer. *Eukrohnia bathyantarctica* was not included in the respiration experiments because it was difficult to collect enough specimens with similar properties in good condition.

Before the start of the experiment, the chaetognaths were acclimated for up to a day at constant temperature (0°C) in the dark. For respiration rate measurements, specimens were incubated in sealed glass bottles of about 60 or 500 mL volume filled with filtered (0.7 μm pore size) and oxygen-saturated seawater (saturation above approximately 92%) for 19–26 h at the acclimation temperature and light conditions. During the winter expedition, two specimens were incubated in a 60-mL bottle. In summer, additional experiments were conducted with four specimens per 500-mL bottle to evaluate the influence of bottle volume on respiration. Individuals similar in size and maturity (primarily stage II) were chosen from the same depth range for each measurement. Each respiration experiment consisted of three to eight bottles with chaetognaths and two to four controls without chaetognaths. During winter and summer, 52 and 51 measurements (including 34 in 500-mL bottles) were done with *E. hamata* individuals. Five and nine measurements (including eight in 500-mL bottles) were performed with *E. bathypelagica* individuals in winter and summer. Measurements of dissolved oxygen were performed by the Winkler titration method (Grasshoff 1983) on the entire sample bottle or on subsamples, which were carefully siphoned out of the 500-mL bottles. The decrease in oxygen concentration in the bottles with chaetognaths compared to the controls was always less than 10%. All data were normalized per unit dry mass. Specimens that remained in the 500-mL experimental bottles and specimens separately frozen from the net samples were taken for dry mass and carbon content determination.

To estimate the metabolic loss in terms of the absolute and relative amount of carbon, respired by *E. hamata* and *E. bathypelagica* per day, the measured oxygen consumption rates in μL O_2 individual^{-1} h^{-1} were converted to carbon units using a respiratory quotient (*RQ*) value of 0.97 in the following equation (Ikeda et al. 2000):

$$\text{mg carbon individual}^{-1}\,\text{h}^{-1} = \text{mL O}_2\,\text{individual}^{-1}\,\text{h}^{-1} \times RQ \times 12/22.4$$

where 12/22.4 is the carbon weight (12 g) in 1 mol of CO_2 (22.4 L). For *E. bathyantarctica*, we applied a comparable calculation based on the general DM and carbon measurements using the average respiration rate of the two other *Eukrohnia* species.

Statistical analyses

Differences in carbon (C; μg mg DM^{-1}), nitrogen (N; μg mg DM^{-1}) and lipid content (mg lipid mg DM^{-1}) were analyzed by a full factorial two-way ANOVA for each parameter (C/N/lipid versus species & season & species × season) with a subsequent post hoc Tukey HSD test on differences between means ($\alpha = 0.05$, Sokal and Rohlf 1981). We applied a full factorial two-way ANOVA for each species (lipid content versus maturity stage & season & maturity stage × season) to test for stage-specific differences in the lipid content of *Eukrohnia bathypelagica* and *E. bathyantarctica*. This analysis was done separately because the same maturity stages were not present for both species during both seasons. Stage I and III specimens were excluded from the analyses for *E. bathypelagica*, and stages IV and V were excluded for *E. bathyantarctica* analyses. Individuals of these stages were not in sufficiently good condition during winter.

A full factorial three-way ANOVA was used to examine the influence of depth on respiration (μL O_2 mg DM^{-1} h^{-1}; respiration versus species & season & depth & species × season & species × depth & season × depth & species × season × depth). The 1,000–1,500 m depth range was excluded from the analysis, since no measurements were conducted with *E. hamata* from this depth range in summer and with *E. bathypelagica* in winter. A *t*-test was applied to analyze the dependence of bottle volume on respiration of *E. hamata* in summer. Prior to these parametric tests, the data were Box-Cox transformed (Sokal and Rohlf 1981) when necessary to achieve normality and homogeneity of variances.

We performed a principal component analysis (PCA) on the fatty acid compositions (percentages) to analyze

Table 1 Gut contents of *Eukrohnia hamata* and *E. bathypelagica* in the Weddell Sea during winter 2006 and summer 2007/2008

Species	*E. hamata*		*E. bathypelagica*	
Season	Winter	Summer	Winter	Summer
Number of specimens (*n*)	200	200	200	200
Guts with lipid droplets (%)	92.0	82.5	57.0	34.0
FCR (total, %)	42.0	39.0	48.0	31.0
Degree of gut fullness				
<10%	26.0	25.5	18.5	23.5
10–25%	10.0	12.5	20.0	7.0
25–50%	5.5	1.0	9.0	0.5
50–100%	0.5	–	0.5	–
FCR (identifiable content only)	8.0	25.0	4.0	16.0
FCR (without diatoms, Acantharia and jellyfish remains)	4.0	1.0	1.5	1.0
Diatoms (e.g. *Fragilariopsis*, *Chaetoceros*)	2.0	0.5	–	–
Acantharia	1.0	–	–	–
Radiolaria	0.5	–	1.0	0.5
Tintinnids (e.g. *Cymatocylis*)	1.0	0.5	–	–
Copepoda	3.5	0.5	1.0	0.5
Chaetognatha	0.5	–	–	–
Jellyfish remains	3.0	24.5	3.0	15.0
Fecal pellets	0.5	–	–	–
NPC (without diatoms, Acantharia and jellyfish remains)	0.11 ± 0.95	0.01 ± 0.10	0.03 ± 0.26	0.01 ± 0.10

Data include number of guts containing oil droplets (lipids), the degree of gut fullness, the food containing ratio (FCR; total FCR including unidentifiable mass) and the number of prey per chaetognath (NPC, ± SD). Data represent % of specimens investigated, except for the number of specimens and for NPC

differences between species, maturity stages, depths and seasons. Fourteen common fatty acids found in *E. bathypelagica* and *E. bathyantarctica* were included in this analysis. All statistical analyses were performed with the software Statistica 8 (StatSoft).

Results

Gut content

Less than half of all *Eukrohnia hamata* specimens analyzed had food in their guts (39% during summer and 42% during winter; total FCR, Table 1). In *E. bathypelagica*, 48% contained food during winter and 31% in summer. In both species, the degree of gut fullness was rather low, usually less than 25%. Gut contents were mostly located in the middle and posterior part of the gut. In contrast to the former two species, the gut of *E. bathyantarctica* is characterized by an orange color. Due to this pigmentation, the gut content was more difficult to identify in this species and the gut fullness could not be recorded precisely. Even after careful dissection, no identifiable particles were detected in *E. bathyantarctica*.

In *E. hamata* and *E. bathypelagica* we found identifiable prey items, although most prey were in advanced stages of decomposition. Copepods, protozoans (Acantharia, Radiolaria, tintinnids), jellyfish remains (particularly nematocysts) and diatoms were the main sources of food identified in both species (Table 1). Only one complete copepod and few mandibles (particularly coxal gnathobases) of copepods were found in comparatively good condition. These copepod remains could not be related to the respective species. Excluding diatoms, Acantharia and jellyfish remains, on average ≤0.11 prey items were observed per specimen (NPC, Table 1).

Regardless of the season, oil droplets were found in the guts of most *Eukrohnia hamata* specimens (>82%). In *E. bathypelagica*, 34% of all guts contained oil droplets in summer and 57% in winter (Table 1). Due to the intense gut pigmentation and the oil globules in the intestinal tissue, oil droplets in the guts of *E. bathyantarctica* were difficult to identify even after dissection.

Carbon, nitrogen and total lipid content

The carbon and nitrogen contents of the meso- and bathypelagic chaetognaths depended significantly on

Table 2 Summarized results of body composition and daily metabolic loss (% body carbon; mean ± SD and range) of *Eukrohnia hamata*, *E. bathypelagica* and *E. bathyantarctica*

		Eukrohnia hamata		*E. bathypelagica*		*E. bathyantarctica*	
		Summer	Winter	Summer	Winter	Summer	Winter
Number of specimens	*n*	72 (70)	179	40	43	42 (41)	35
Length (mm)	Mean	21.3 ± 4.9	22.2 ± 3.2	21.1 ± 2.4	21.9 ± 2.2	22.0 ± 5.3	21.7 ± 4.3
	Range	11–29	12–28	15–25	16.5–26	12–30	11–27
Dry mass (mg)	Mean	2.8 ± 1.9	2.4 ± 1.2	2.6 ± 1.6	2.2 ± 1.2	3.4 ± 2.5	2.2 ± 1.0
	Range	0.3–8.8	0.5–6.1	0.5–6.4	0.8–6.7	0.3–9.5	0.4–4.1
Carbon (% DM)	Mean	30.4 ± 7.7	39.4 ± 6.1	24.6 ± 7.2	31.3 ± 8.9	32.4 ± 4.9	42.4 ± 7.1
	Range	12.1–52.8	16.4–65.3	14.3–39.2	19.2–51.5	21.8–42.6	29.2–56.9
Nitrogen (% DM)	Mean	6.8 ± 1.7	7.8 ± 1.2	5.7 ± 1.1	6.9 ± 1.2	6.9 ± 1.0	8.4 ± 1.4
	Range	2.4–13.1	3.7–11.6	3.6–8.0	5.0–9.3	3.9–8.2	5.9–11.8
C:N ratio	Mean	4.6 ± 0.7	5.1 ± 1.0	4.3 ± 0.6	4.5 ± 0.9	4.8 ± 0.8	5.1 ± 0.9
	Range	3.4–7.2	3.6–8.9	3.4–5.6	3.6–7.2	3.6–7.3	3.7–7.1
Body carbon respired per day (% C)	*n*	51	52	9	5	42	35
	Mean	0.67 ± 0.45	0.49 ± 0.17	1.09 ± 0.97	0.45 ± 0.06	0.61 ± 0.10	0.47 ± 0.08
	Range	0.10–1.87	0.22–0.93	0.20–2.64	0.37–0.50	0.45–0.88	0.34–0.66

Fewer nitrogen and C:N data were obtained for *E. hamata* and *E. bathyantarctica* in summer (number of specimens given in brackets)
DM dry mass

season (ANOVA, for C: $F = 117.206$, $P < 0.001$; for N: $F = 72.595$, $P < 0.001$) and species (ANOVA, for C: $F = 40.442$, $P < 0.001$; for N: $F = 23.825$, $P < 0.001$). Carbon, nitrogen and C:N ratios were higher in winter than in summer (Table 2). In *Eukrohnia bathypelagica*, we observed the lowest carbon (winter: 31.3% DM, summer: 24.6% DM) and nitrogen contents (winter: 6.9% DM, summer: 5.7% DM) of all three species. The C and N contents of *E. hamata* and *E. bathyantarctica* were similar but generally higher than those of *E. bathypelagica*. The same was true for the C:N ratio of *E. bathypelagica* (winter: 4.5, summer: 4.3), which was significantly lower than for the two other species (Tukey HSD, $P < 0.001$).

The total lipid contents of the two deeper-living species *E. bathypelagica* and *E. bathyantarctica* were significantly different from each other (ANOVA, $F = 9.026$, $P < 0.01$; Tables 3, 4). *Eukrohnia bathyantarctica* had an average lipid content of 15.4% DM (SD ± 4.1) and showed no significant variability related to maturity stage (ANOVA, $F = 1.393$, $P > 0.05$) or season (ANOVA, $F = 0.001$, $P > 0.05$; Table 4). In contrast, the lipid contents of *E. bathypelagica* ranged between 1.4 and 25.4% DM and differed distinctly among maturity stages (ANOVA, $F = 5.499$, $P < 0.05$; Table 3). Stage IV (winter: 12.7% DM, summer: 20.0% DM) contained relatively more lipids than stages II and V. Moreover, a higher lipid level in *E. bathypelagica* was measured in summer than in winter (winter: 8.4% DM, summer: 13.9% DM; ANOVA, $P < 0.05$; all maturity stages included).

Fatty acid and fatty alcohol compositions of *E. bathypelagica* and *E. bathyantarctica*

Total lipids of *Eukrohnia bathypelagica* consisted mainly of fatty acids (mean 95.9% of total fatty acids and alcohols) with only minor amounts of fatty alcohols (mean 4.1% of total fatty acids and alcohols). *Eukrohnia bathyantarctica* had higher amounts of fatty alcohols (Table 4; mean 22.8%) compared to *E. bathypelagica*. The fatty alcohols in *E. bathyantarctica* indicated the presence of wax esters with a contribution of up to 57.3% of total lipids.

The five fatty acids 16:0, 16:1(n-7), 18:1(n-9), 20:5(n-3) and 22:6(n-3) generally dominated within the two chaetognath species *E. bathypelagica* and *E. bathyantarctica* and comprised 60.3 and 69.0% of total fatty acids (TFA), respectively. However, there were distinct differences between the fatty acid patterns of these two species, which are reflected by the clustering in the principal component analysis (PCA, Fig. 2). The PCA based on the fatty acid composition extracted three principal components with eigenvalues >1% of variance. Results were presented for the major two components, together accounting for 73.9% of variance (Fig. 2).

The fatty acid composition of *E. bathypelagica* was characterized by higher amounts of 16:0, 20:5(n-3) and 22:6(n-3). These fatty acids contributed mean values of 9.7, 11.0 and 22.1%, respectively, to TFA (Table 3). Season, depth and maturity stage had a marginal influence on the

Table 3 *Eukrohnia bathypelagica*. Dry mass and total lipid content as well as fatty acid composition for different maturity stages during winter and summer (mean ± SD)

	Winter			Summer				
Maturity stage	II	IV	V	I	II	III	IV	V
Number of samples	4	3	8	3	3	4	7	2
Number of individuals	5	4	10	3	6	10	14	2
Ind. dry mass (mg)	2.7 ± 1.5	4.5 ± 1.1	2.1 ± 0.3	1.4 ± 0.5	2.1 ± 0.4	2.5 ± 1.2	4.2 ± 0.9	2.7 ± 1.0
Lipid (% DM)	8.4 ± 4.1	12.7 ± 3.0	6.8 ± 1.5	9.7 ± 2.2	10.3 ± 11.1	10.7 ± 6.2	20.0 ± 4.6	10.8 ± 9.0
Fatty acids (% TFA)								
16:0	8.0 ± 0.7	9.7 ± 1.2	8.4 ± 2.1	12.5 ± 1.8	9.8 ± 2.4	9.8 ± 2.9	12.2 ± 0.9	5.2 ± 3.1
16:1(n-9)	0.4 ± 0.4	0.3 ± 0.3	0.9 ± 0.5	1.4 ± 0.6	1.0 ± 0.9	0.8 ± 0.6	0.9 ± 0.4	0.8 ± 0.5
16:1(n-7)	6.4 ± 2.4	11.1 ± 1.2	6.4 ± 1.7	6.0 ± 1.5	6.8 ± 1.1	8.2 ± 1.7	9.1 ± 1.9	5.7 ± 0.4
16:1(n-x)[a]	0.1 ± 0.2	1.2 ± 0.4	0.1 ± 0.2	0.0 ± 0.0	0.1 ± 0.2	1.4 ± 1.7	2.6 ± 1.2	0.0 ± 0.0
17:0	0.8 ± 0.6	0.0 ± 0.0	1.0 ± 0.3	1.1 ± 0.3	0.8 ± 0.4	0.5 ± 0.5	0.2 ± 0.2	0.5 ± 0.7
18:0	4.0 ± 1.9	1.6 ± 0.1	4.1 ± 1.2	6.2 ± 1.3	3.8 ± 1.9	3.1 ± 1.6	1.9 ± 0.3	2.7 ± 1.8
18:1(n-9)	8.7 ± 1.8	11.7 ± 4.1	9.6 ± 3.5	11.4 ± 2.0	8.4 ± 1.4	10.3 ± 3.6	11.1 ± 3.8	7.3 ± 1.7
18:1(n-7)	1.7 ± 0.4	2.1 ± 0.5	1.6 ± 0.4	1.7 ± 0.1	1.3 ± 0.4	2.1 ± 0.9	2.5 ± 0.8	1.1 ± 0.5
18:1(n-5)	1.8 ± 0.5	2.3 ± 0.1	1.6 ± 0.4	1.2 ± 0.2	1.4 ± 0.3	1.9 ± 0.4	2.4 ± 0.3	1.3 ± 0.8
18:2(n-6)	2.0 ± 0.4	1.3 ± 0.2	2.1 ± 0.7	3.0 ± 1.1	2.1 ± 1.2	1.5 ± 0.5	1.0 ± 0.5	1.6 ± 0.8
18:4(n-3)	0.3 ± 0.6	0.9 ± 0.2	0.6 ± 1.4	0.0 ± 0.0	0.6 ± 1.0	0.7 ± 0.9	1.4 ± 0.5	3.3 ± 3.8
20:1(n-9)	3.7 ± 5.2	8.0 ± 0.7	2.2 ± 1.2	2.0 ± 0.7	2.8 ± 0.2	6.3 ± 5.4	8.0 ± 4.1	6.6 ± 4.6
20:1(n-x)[a]	2.2 ± 1.2	1.3 ± 0.2	2.4 ± 0.5	1.9 ± 0.4	1.7 ± 0.8	2.1 ± 1.1	1.4 ± 0.3	1.7 ± 1.3
20:4(n-6)	1.4 ± 0.3	1.1 ± 0.1	1.7 ± 0.1	1.5 ± 0.3	1.1 ± 1.0	1.5 ± 0.6	0.9 ± 0.1	1.4 ± 0.3
20:5(n-3)	10.5 ± 0.9	11.4 ± 1.1	11.7 ± 2.0	9.6 ± 3.4	10.4 ± 2.9	10.2 ± 1.0	10.8 ± 2.8	13.3 ± 0.8
22:1(n-11)	3.6 ± 6.5	7.9 ± 5.3	1.4 ± 1.5	0.9 ± 0.9	7.6 ± 11.3	2.5 ± 1.5	6.0 ± 2.6	8.8 ± 10.9
22:1(n-9)	3.7 ± 2.2	5.3 ± 2.4	2.7 ± 1.0	2.1 ± 0.7	4.9 ± 4.6	3.3 ± 0.9	5.1 ± 1.7	6.2 ± 5.1
22:5(n-3)	0.5 ± 0.1	0.6 ± 0.1	0.6 ± 0.3	0.8 ± 0.1	0.6 ± 0.1	0.4 ± 0.3	0.5 ± 0.1	1.2 ± 1.0
22:6(n-3)	27.8 ± 9.6	15.1 ± 3.0	28.3 ± 5.1	24.6 ± 2.0	22.2 ± 11.3	23.5 ± 12.3	13.3 ± 2.1	20.7 ± 17.0
24:1(n-9)	5.0 ± 2.5	1.9 ± 0.2	5.1 ± 0.8	4.3 ± 0.8	4.1 ± 0.7	3.8 ± 1.8	2.2 ± 0.5	3.5 ± 2.8
FA < 1%	7.6 ± 1.3	5.3 ± 1.0	7.3 ± 1.8	7.8 ± 3.5	8.6 ± 4.3	6.0 ± 1.2	6.5 ± 1.0	7.1 ± 4.8
Ratio								
[18:1(n-9) + 20:1(n-9) + 22:1(n-9)]/ [16:1(n-7) + 18:1(n-7) + 18:4(n-3)]	1.9 ± 0.4	1.8 ± 0.1	1.8 ± 0.7	2.0 ± 0.2	1.9 ± 0.5	1.7 ± 0.3	1.9 ± 0.3	2.0 ± 0.1

DM dry mass, *TL* total lipid, *TFA* total fatty acids

[a] Double bond position not identified

fatty acid compositions of *E. bathypelagica*. The PCA revealed a grouping of stage IV *E. bathypelagica* individuals originating from 1,500 to 2,000 m depth that contained lower mean amounts of 22:6(n-3) (winter: 15.1%, summer: 13.3%) compared to the other maturity stages (Fig. 2). Hence, particularly stages I, II and V from 500 to 1500 m depth were similar in their fatty acid composition with the exception of three samples (Fig. 2). Generally, moderate amounts of the monounsaturated fatty acids 16:1(n-7) (mean 7.5%) and 18:1(n-9) (mean 10.0%) were measured in *E. bathypelagica* (Table 3). The fatty acids 20:1(n-9) and 22:1(n-11) represented up to 14.3% (mean 4.9%) and 20.7% TFA (mean 4.3%) in this species. The remaining fatty acids each accounted ≤4% TFA on average.

Eukrohnia bathyantarctica contained high levels of 18:1(n-9), with a maximum of 39.1% of TFA (Table 4; mean 32.7%). The fatty acids 16:0, 20:5(n-3) and 22:6(n-3) were considerably less abundant in *E. bathyantarctica* compared to *E. bathypelagica*. On average 22:6(n-3) accounted for 11.5% TFA (Table 4), which is about half of its contribution to TFA in *E. bathypelagica*. The fatty acids 16:0 and 20:5(n-3) contributed only 6.5 and 5.9% to TFA, respectively. 16:1(n-7) and 18:1(n-7) exhibited higher percentages of 12.3 and 6.4% in this species, whereas similarly low levels of 20:1(n-9) were measured. PCA revealed a species cluster with no apparent seasonal, depth or maturity stage-related pattern identifiable (Fig. 2).

Table 4 *Eukrohnia bathyantarctica*. Dry mass, total lipid and wax ester content as well as fatty acid and fatty alcohol composition for different maturity stages during winter and summer (mean ± SD)

	Winter			Summer				
Maturity stage	I	II	III	I	II	III	IV	V
Number of samples	1	6	3	2	10	3	1	1
Number of individuals	1	6	3	5	15	5	1	1
Ind. dry mass (mg)	2.1	3.4 ± 1.5	5.7 ± 0.3	0.6 ± 0.2	3.4 ± 1.6	5.6 ± 0.7	5.9	4.3
Lipid (% DM)	17.7	16.9 ± 4.7	11.1 ± 4.0	14.2 ± 0.7	16.3 ± 4.1	15.4 ± 3.7	15.3	10.1
Wax esters (% TL)	40.1	47.8 ± 7.6	42.4 ± 6.5	42.5 ± 1.7	44.8 ± 4.6	51.5 ± 3.0	47.5	40.6
Fatty acids (% TFA)								
14:0	1.7	1.0 ± 0.3	1.2 ± 0.2	1.3 ± 0.1	1.3 ± 0.3	1.1 ± 0.4	1.4	1.0
16:0	8.1	6.6 ± 0.6	7.1 ± 0.9	6.6 ± 0.3	6.4 ± 0.7	5.7 ± 0.2	6.3	5.2
16:1(n-7)	10.0	12.0 ± 3.2	11.4 ± 1.1	10.5 ± 0.7	12.9 ± 2.5	14.0 ± 2.4	12.6	12.1
16:2(n-4)	0.8	0.7 ± 0.7	0.2 ± 0.4	0.0 ± 0.0	0.5 ± 0.6	0.9 ± 0.3	0.0	0.0
18:0	1.5	1.4 ± 0.2	1.3 ± 0.3	1.8 ± 0.3	1.2 ± 0.3	1.0 ± 0.3	1.2	1.5
18:1(n-9)	29.0	31.9 ± 3.5	35.1 ± 3.9	29.2 ± 3.5	33.0 ± 1.6	34.9 ± 1.9	31.3	33.5
18:1(n-7)	5.7	6.4 ± 1.0	6.6 ± 1.0	6.5 ± 0.8	6.5 ± 0.6	6.2 ± 0.4	7.3	6.2
18:1(n-5)	1.1	1.0 ± 0.4	1.4 ± 0.6	0.9 ± 0.0	0.9 ± 0.2	1.3 ± 0.1	1.5	1.0
18:2(n-6)	1.6	1.2 ± 0.3	1.1 ± 1.0	1.5 ± 0.2	1.3 ± 0.2	0.6 ± 0.5	1.1	1.3
18:4(n-1)	1.0	0.8 ± 0.4	0.8 ± 0.3	0.8 ± 0.2	1.4 ± 0.5	1.0 ± 0.1	0.7	1.6
20:1(n-11)	0.5	0.6 ± 0.5	0.6 ± 0.6	0.9 ± 0.3	0.7 ± 0.2	0.8 ± 0.1	0.9	1.2
20:1(n-9)	2.7	3.5 ± 1.9	3.2 ± 0.4	6.4 ± 4.5	3.4 ± 1.6	4.4 ± 1.1	3.8	4.1
20:1(n-7)	0.8	0.8 ± 0.2	0.7 ± 0.1	0.7 ± 0.2	0.8 ± 0.2	0.7 ± 0.1	0.7	0.6
20:1(n-x)[a]	0.7	0.8 ± 0.3	1.2 ± 0.4	0.7 ± 0.2	0.8 ± 0.2	0.9 ± 0.2	1.0	1.0
20:5(n-3)	8.3	6.0 ± 0.9	4.5 ± 1.0	5.9 ± 2.3	6.3 ± 1.2	5.3 ± 1.3	6.7	5.7
22:1(n-11)	2.7	1.8 ± 0.8	1.8 ± 1.1	2.4 ± 1.4	1.7 ± 0.6	2.3 ± 0.4	2.3	2.8
22:1(n-9)	2.7	2.7 ± 1.4	1.6 ± 0.4	2.1 ± 0.1	2.1 ± 0.6	2.3 ± 0.5	2.9	1.8
22:6(n-3)	12.1	11.6 ± 2.7	12.3 ± 3.8	13.1 ± 1.3	11.4 ± 2.8	10.2 ± 1.2	11.0	11.6
24:1(n-9)	2.6	2.4 ± 0.7	2.8 ± 1.0	3.2 ± 0.1	2.5 ± 0.9	2.0 ± 0.1	1.9	3.6
FA < 1%	6.5	6.7 ± 2.3	5.1 ± 1.9	5.5 ± 2.4	5.0 ± 0.8	4.7 ± 1.3	5.4	4.1
Ratio								
[18:1(n-9) + 20:1(n-9) + 22:1(n-9)]/ [16:1(n-7) + 18:1(n-7) + 18:4(n-3)]	2.1	2.1 ± 0.3	2.2 ± 0.3	2.2 ± 0.3	1.9 ± 0.2	2.0 ± 0.2	1.9	2.2
Fatty alcohols (% TFAlc)								
14:0	11.6	8.4 ± 3.5	8.8 ± 2.9	9.0 ± 3.0	9.4 ± 3.1	8.2 ± 4.4	4.7	7.0
16:0	28.1	26.3 ± 4.7	21.7 ± 1.4	25.7 ± 1.5	26.1 ± 3.8	25.6 ± 3.4	16.1	24.0
16:1	0.0	0.5 ± 1.1	0.0 ± 0.0	1.3 ± 0.2	2.0 ± 1.4	1.2 ± 1.2	0.0	2.2
18:0	1.8	1.7 ± 0.6	1.6 ± 0.3	2.5 ± 0.3	1.6 ± 0.4	1.3 ± 0.2	1.7	1.9
18:1(n-9)	11.3	12.6 ± 4.7	13.3 ± 1.1	13.4 ± 3.2	14.7 ± 3.9	11.0 ± 0.9	9.3	17.0
20:1(n-9)	13.4	13.6 ± 12.3	25.9 ± 7.2	18.6 ± 9.7	15.6 ± 6.5	23.4 ± 5.0	23.8	17.0
20:1(n-7)	3.9	3.5 ± 2.4	4.4 ± 1.4	2.5 ± 0.5	2.7 ± 1.0	3.6 ± 0.6	4.0	2.7
22:1(n-11)	6.7	8.5 ± 5.1	6.5 ± 2.8	7.2 ± 1.3	7.9 ± 3.4	7.1 ± 1.0	11.9	9.4
22:1(n-9)	20.4	22.1 ± 14.8	15.5 ± 4.3	17.1 ± 4.1	17.4 ± 6.2	16.4 ± 2.2	25.4	15.9
22:1(n-7)	2.7	2.8 ± 1.4	2.3 ± 0.5	2.8 ± 0.1	2.6 ± 1.0	2.1 ± 0.2	3.2	2.7

DM dry mass, *TL* total lipid, *TFA* total fatty acids, *TFAlc* total fatty alcohols

[a] Double bond position not identified

The fatty acid ratio [18:1(n-9) + 20:1(n-9) + 22:1(n-9)]/[16:1(n-7) + 18:1(n-7) + 18:4(n-3)], used as an indicator of carnivory, was similar in *E. bathypelagica* and *E. bathyantarctica* with mean values of 1.9 and 2.0, respectively (Tables 3, 4). The fatty alcohol content was highly variable in *E. bathypelagica* and ranged between

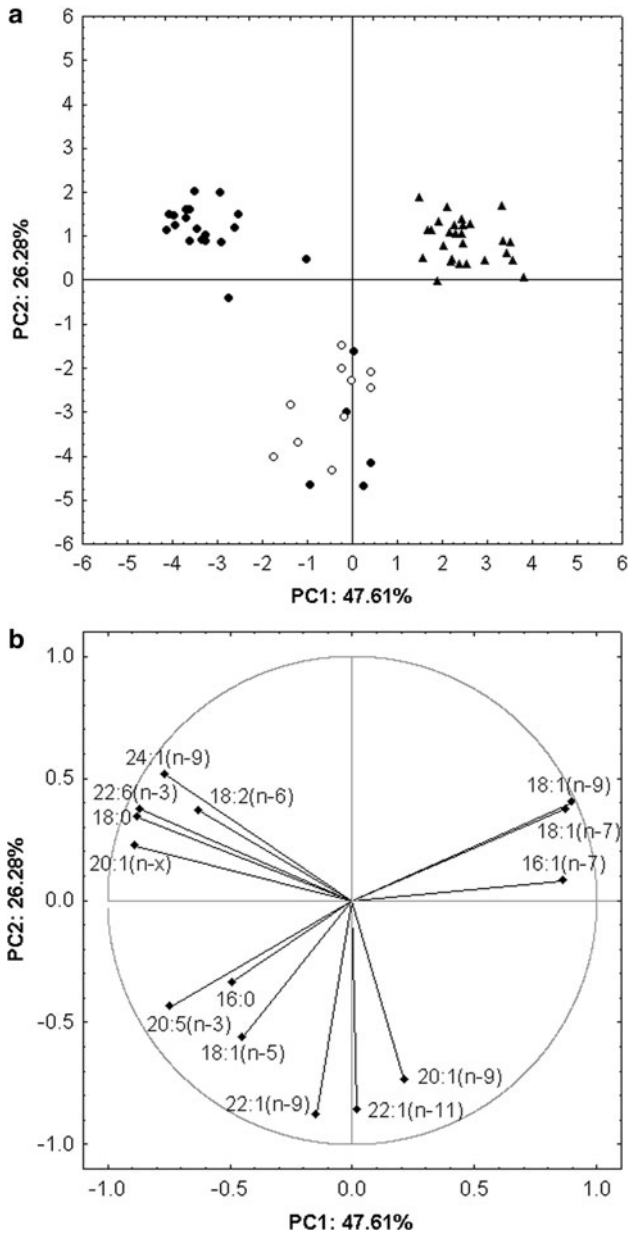

Fig. 2 Principal component analysis (PCA) of *Eukrohnia bathypelagica* and *E. bathyantarctica* according to their fatty acid compositions. Two-dimensional PCA score (**a**) and loading (**b**) plot of the first two principal components (PC1 and PC2) based on the fatty acid profiles of both species (14 common and relevant fatty acids). *Eukrohnia bathypelagica—dots* (*empty dots* stage IV individuals in the 1,500–2,000 m depth range), *E. bathyantarctica—triangles*

0 and 26.6% of TFA and total fatty alcohols (TFAlc; mean 4.1%, not shown in the table). *Eukrohnia bathyantarctica* contained a higher portion of fatty alcohols (mean 22.8% of TFA and TFAlc) compared to *E. bathypelagica*. These fatty alcohols mainly consisted of 16:0 (mean 25.2% TFAlc), 18:1(n-9) (13.3%), 20:1(n-9) (17.7%) and 22:1(n-9) (18.5%; Table 4).

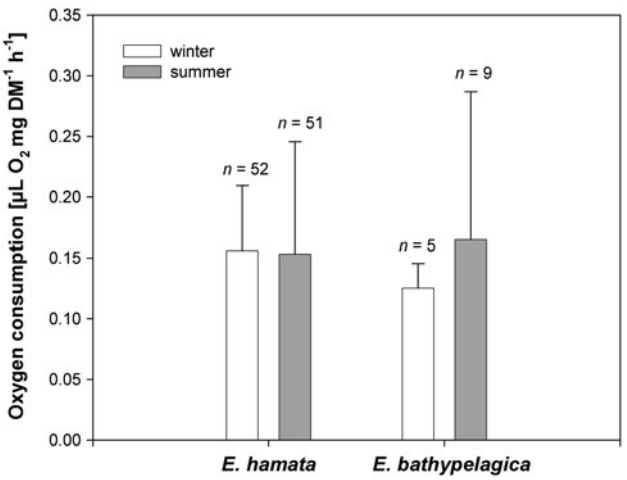

Fig. 3 *Eukrohnia hamata* and *E. bathypelagica*. Oxygen consumption during winter and summer at 0°C. *n* Number of measurements

Oxygen consumption

The respiration experiments showed that the bottle volume (1 ind. per approximately 140 or 30 mL) had no significant influence on oxygen consumption of *Eukrohnia hamata* (*t*-test, $t = 1.34$, $P > 0.05$; 0.16 ± 0.09 µL O_2 mg DM^{-1} h^{-1} for 140 mL and 0.14 ± 0.09 µL O_2 mg DM^{-1} h^{-1} for 30 mL). Mean respiration rates of 0.15 µL O_2 mg DM^{-1} h^{-1} (SD $\pm$ 0.08) were measured in *E. hamata* and *E. bathypelagica* (Fig. 3). No significant differences were observed between species (ANOVA, $F = 0.941$, $P > 0.05$) and seasons (ANOVA, $F = 0.637$, $P > 0.05$). Mass-specific oxygen consumption did not differ between specimens originating from different depths (ANOVA, $F = 2.201$, $P > 0.05$; 500–750, 750–1,000 m).

The mean metabolic loss for all three *Eukrohnia* species during both seasons was very low, with <1.1% of body carbon respired per day (Table 2). The metabolic loss tended to be lower in winter (<0.5%) due to the higher body carbon contents but constant respiration rates.

Discussion

Feeding ecology

Our results on chaetognath feeding suggest a copepod-based diet, although protozoans and chaetognaths may also be found in the chaetognaths' guts. The fatty acid analyses of *Eukrohnia bathypelagica* and *E. bathyantarctica* revealed species-specific results, which further indicate differences in the diet between these two species. The feeding ecology of the three investigated chaetognath species will be discussed in detail in the following section.

Copepods create a distinct flow field in the water (Bundy and Paffenhöfer 1996; Jiang and Osborn 2004), which is readily detected by chaetognaths via sensory hairs (Newbury 1972; Feigenbaum and Reeve 1977). Based on this fact and due to the dominance of copepods, chaetognaths feed largely on copepods (e.g. Øresland 1990; Froneman and Pakhomov 1998). The Antarctic *E. hamata* preys on the copepodite stages of *Calanus* spp., *Euchaeta* spp., *Metridia gerlachei* and *Rhincalanus gigas* as well as on smaller copepods like *Microcalanus pygmaeus*, *Oithona* spp. and *Oncaea* spp. (Hopkins 1985, 1987; Hopkins and Torres 1989; Øresland 1990, 1995; Froneman et al. 1998). Apart from diatoms and jellyfish remains, copepods were the most frequent food items found in the guts of *E. hamata* and *E. bathypelagica* in our study. Due to the high degree of digestion, mainly parts of copepods were found. Even the few mandible remains and the one complete copepod in the chaetognaths' guts could not be identified to species level. They could belong to deep-sea copepods because their characteristics do not seem to match species frequently encountered in the upper hundred meters of the Southern Ocean. Diatoms were assumed to be ingested inadvertently or via herbivorous prey (Feigenbaum 1991). Medusae were also assumed to be artifacts of collecting and preserving methods rather than being true prey items (Feigenbaum 1991 and references therein). These were found particularly in the guts of chaetognaths from RMT samples, which often also contained jellyfish. Some degree of cod-end feeding or better indiscriminately grasping within the cod-end cannot be excluded. Appendicularians, ostracods and other chaetognaths were documented in the chaetognath's diet (Øresland 1990; Froneman et al. 1998; Froneman and Pakhomov 1998). Few chaetognath hooks or tintinnids were observed in *E. hamata* guts, although the latter prey is more common in young chaetognaths (Pearre 1981). The two acantharians found in the guts of *E. hamata* were in very good condition and probably accidentally ingested by the chaetognaths, either in the cod-end or eaten via their copepod prey. To our knowledge, no data exist on gut contents of the two deeper-living species *E. bathypelagica* and *E. bathyantarctica*. Due to several methodological constraints of the gut content analysis, which will be discussed below, these analyses provide only rough estimates of the feeding preferences.

Generally, the frequency of chaetognaths with gut content is low (e.g. David 1955; Hopkins 1985; Lancraft et al. 1991; Øresland 1995). In our investigation, the number of chaetognaths containing food in their guts ranged between 31 and 48% (FCR total) for *E. hamata* and *E. bathypelagica*, irrespective of the state of digestion. In several previous studies, only the identifiable contents were counted, or as in the case of Froneman and Pakhomov (1998), oil droplets were also considered as gut content, which we treated separately. In the Australian sector of the Southern Ocean, Johnson and Terazaki (2004) found 15.4–48.9% (station means) of *E. hamata* specimens with prey in their guts. Usually, more than 84% of the guts of *E. hamata* or *Sagitta gazellae* contain no food items (Øresland 1995; Froneman et al. 1998). Up to 4.0% of our chaetognaths' guts contained identifiable material when we exclude diatoms, Acantharia and jellyfish remains from our data. However, the amount of unidentifiable food was high in our study. In summer, 0.02–0.06 (Froneman et al. 1998) and 0.21 prey items (0.10–0.26 during 24 h sampling, Øresland 1990) were found in *E. hamata*. For the winter season, 0.08 and 0.12 prey items per *E. hamata* were counted at two different stations in Gerlache Strait (Øresland 1995), which is in the same range as our data (0.01–0.11, diatoms, Acantharia and jellyfish remains excluded). The mean number of prey per chaetognath (NPC) seems to be slightly higher in *Sagitta* compared to *Eukrohnia* species (Øresland 1990: *S. marri*: 0.23, *S. gazellae*: 0.26, *S. maxima*: 0.20).

The gut content analysis has various drawbacks that are associated with the sampling procedure. In our investigation, chaetognaths were taken from great depths, which greatly increases the retrieval time. Two uncertainties, progressed digestion and cod-end feeding, may be associated with such long retrieval times. A digestion time of 9.3 h can be assumed for large copepods, 4.9 h for small copepods and unidentified prey (for *Sagitta elegans* at 6°C in Gullmarsfjorden, Sweden; Øresland 1987). In the Southern Ocean, Giesecke et al. (2010) estimated a digestion time between 9 and 15.8 h (mean 11.5 h, at 0 ± 1°C) for *S. gazellae*, also tending to increase with larger prey. Thus, lower temperatures in the Southern Ocean may result in longer digestion times, but yet this process has to be taken into consideration. Cod-end feeding in plankton net hauls is another problem when applying gut content analysis (Baier and Purcell 1997). Therefore, prey items found in the foreguts were excluded from our analyses. We can also not exclude regurgitation in chaetognaths. Regurgitation and defecation might occur as a stress reaction on capturing or on preservation. Baier and Purcell (1997) presumed that the prey loss in chaetognath guts during sampling was due to stress-induced gut evacuation rather than to continued digestion. This could explain the generally low NPC. Prey loss seems rather to occur at the beginning of the catch, whereas cod-end feeding may take place throughout the tow (Baier and Purcell 1997). We tried to reduce the stress during the catch by using large cod-ends, thus avoiding crowded samples. Nevertheless, the sampling method seems to have a strong effect on results of chaetognath feeding, and hence it is difficult to draw conclusions on seasonal and species-specific differences in diet composition and feeding activity only on the basis of gut content analyses.

To elucidate diet preferences in the two deeper-living species *E. bathypelagica* and *E. bathyantarctica*, we therefore conducted additional fatty acid trophic marker analyses. The latter species was of special interest, as no identifiable prey items were found in its guts. The fatty acid 18:1(n-9) distinguishes these two chaetognaths. In *E. bathyantarctica*, it was a dominant component (32.7%), whereas *E. bathypelagica* contained only moderate amounts of this fatty acid (10.0%). As a trophic marker for carnivory (Falk-Petersen et al. 1990), 18:1(n-9) indicates a high level of carnivorous feeding in *E. bathyantarctica*. Omnivorous and carnivorous zooplankton species are known to have wax esters with high amounts of the 18:1(n-9) fatty acid (Lee et al. 2006). However, the ratio [18:1(n-9) + 20:1(n-9) + 22:1(n-9)]/[16:1(n-7) + 18:1(n-7) + 18:4(n-3)] was similar in both species. The higher this value is, i.e. the larger the amount of the three biomarkers for carnivorous feeding are relative to the three phytoplankton biomarkers, the more pronounced the degree of carnivory will be. The difference in fatty acid compositions between *E. bathypelagica* and *E. bathyantarctica* seems to reflect different prey including copepods. The high amounts of the short-chain fatty alcohols 14:0 and 16:0 again support the high degree of carnivory (Lee et al. 2006) in *E. bathyantarctica*. These two fatty alcohols are also major components in the fatty alcohol compositions of copepods like *Rhincalanus gigas* (Kattner et al. 1994) and *Paraeuchaeta antarctica* (Hagen et al. 1995). The fatty acids 20:1(n-9) and 22:1(n-11) and the respective alcohols are biomarkers for calanid copepods such as *Calanoides acutus* (Graeve et al. 1994; Kattner and Hagen 1995). The mean amounts of these long-chain monounsaturated fatty acids were low in both chaetognath species, but the relative portion of the fatty alcohol 20:1(n-9) was high in *E. bathyantarctica*. This suggests that both chaetognath species feed on calanid copepods, but to a different extent. However, *Calanus propinquus*, which produces large amounts of the unusual 22:1(n-9) fatty acid (Hagen et al. 1993), does not seem to be a preferred prey, as indicated by the low level of this fatty acid in both species. The fatty alcohol 22:1(n-9), however, made up a considerable percentage of the TFAlc composition in *E. bathyantarctica*. This is surprising, because this fatty alcohol is found only in trace amounts in Antarctic copepods and may have been synthesized *de novo* by *E. bathyantarctica*. We cannot prove, however, whether *E. bathyantarctica* is able to synthesize wax esters. The moderate mean percentages of 16:1(n-7) in *E. bathypelagica* and *E. bathyantarctica*, 7.5 and 12.3%, respectively, could suggest the ingestion of diatoms (Dalsgaard et al. 2003). However, this marker fatty acid was probably ingested via herbivorous copepods, which fed on diatoms in epipelagic layers, or via carnivorous copepods. *Paraeuchaeta antarctica* could be a potential prey species. This carnivorous copepod is known to accumulate considerable amounts of the monounsaturated fatty acid 16:1(n-7) (Hagen et al. 1995).

The two long-chained polyunsaturated fatty acids 20:5(n-3) and 22:6(n-3) that represented a significant percentage of the TFA in *E. bathypelagica* were most likely obtained from their prey. A biosynthesis of these fatty acids seems to be improbable. The higher relative portion of these fatty acids compared to *E. bathyantarctica* may be attributed to the lower lipid content and thus less storage lipids of *E. bathypelagica*.

Energetics

The C:N ratios of the three chaetognath species were above 4 on average, which is comparable to the ratios and C and N values previously reported for chaetognaths (Omori 1969: 4.2; Schneider 1990: ~4.4). Terazaki (1993) found a C:N ratio of 4.7 and 3.5 for *Sagitta elegans* in the Japan Sea and in the Pacific, respectively. He assumed that this high ratio in the Japan Sea is caused by body lipids, as oil droplets were observed in the intestinal tissue as in meso- and bathypelagic species (like in *Eukrohnia bathyantarctica* in our study). It was observed that *E. hamata* and *E. bathypelagica* had oil droplets in their guts, independent of maturity stage and season. These oil droplets were already reported by several authors for *E. hamata* (Sameoto 1987; Øresland 1990; Froneman et al. 1998; Froneman and Pakhomov 1998). Their function is still unknown, although they are assumed to act as energy deposit (Kapp 1991), as buoyancy aid or both (Øresland 1990). The role of these lipid droplets with regard to seasonal and breeding migrations and reproduction is not understood.

The dominance of the fatty acids 16:0, 20:5(n-3) and 22:6(n-3) in *E. bathypelagica*, which are typical components of biomembranes, indicates a limited dependence on lipid reserves (Graeve et al. 1997). However, lipids seem to gain in importance with increasing sexual maturity of this species. The higher lipid values in summer primarily reflect the higher number of mature specimens investigated during this season, as ripe individuals with large ovaries filling the body cavity (maturity stage IV) had maximum lipid contents. The stage V individuals carried only empty brood sacs, from which their offspring had been released. Lower lipid levels in these specimens are therefore not surprising. The ovaries in this species appear to accumulate fatty tissue (Alvariño 1983), as most brooding species do. Lipid droplets are often observed in zooplankton ovaries, which may partially be transferred to developing oocytes (Lee et al. 2006). In chaetognaths, this is probably an important strategy to support the development of the offspring from oocytes to young chaetognaths that leave their parental brood sacs. The increase in chaetognath density

associated with gonad maturation may be a reason for the movement of adult individuals to deeper layers (Alvariño 1964). This is in agreement with findings that mature individuals usually occur deeper in the water column (e.g. David 1955; Hagen 1985; Kruse 2009). A higher lipid content in adult specimens would partially compensate the increased body density and also act as buoyancy aid reducing the sinking speed of adults to deeper waters.

Eukrohnia bathyantarctica showed no stage-specific differences in its lipid content. However, the lipid level was usually higher (mean 15.4% DM) than in *E. bathypelagica* (mean 11.5% DM). The lack of differences between maturity stages may partially be explained by their less voluminous ovaries and their smaller number of ova, as compared to *E. bathypelagica* (Kruse 2009). In addition, the orange gut of *E. bathyantarctica* probably contains a carotenoid pigment with many small oil droplets similar to the gut of *E. fowleri* (Terazaki 1991). This might be responsible for the relatively higher lipid content in all maturity stages of *E. bathyantarctica*. It remains unsolved, whether the carotenoids are derived from the prey, e.g. lipid-rich copepods, or whether the chaetognaths synthesize these pigments themselves. Based on their investigations on *Eukrohnia fowleri* and *Sagitta macrocephala*, Terazaki et al. (1977) assume that the carotenoids of the intestinal tissue are produced by the chaetognaths because these carotenoids have a unique character differing from that of phytoplankton and copepods.

Wax esters were a major lipid component in *E. bathyantarctica* (45.5% TL). These are efficient long-term energy reserves and also serve as buoyancy aid due to their very low density (Hagen 2000). A high wax ester content of 34 and 71% TL was found in unidentified deep-living subtropical chaetognaths with an orange colored gut (at 31°N 119°W, probably of the genus *Eukrohnia*, Lee et al. 1971a). In contrast, a deep-water chaetognath with orange pigment from the central South Pacific contained distinctly lower wax ester content (26% TL, Lee and Hirota 1973). The intestinal oil globules observed in *E. bathyantarctica* might contain wax esters that are missing in species like *E. bathypelagica*. We conclude that the truly bathypelagic species *E. bathyantarctica* apparently contains more wax esters, relies more on long-term energy reserves, and/or utilizes wax esters as buoyancy aids. These characteristics are in contrast to species that also inhabit upper mesopelagic layers like *E. bathypelagica*.

Eukrohnia hamata and *E. bathypelagica* respired 0.15 µL O_2 mg DM^{-1} h^{-1} on average. They were usually inactive during the experiments. As chaetognaths have periods of activity and of inactivity in their natural environment (Thuesen and Childress 1993), our data may be a good estimate of inactive periods, i.e. basic metabolism. Seasonal differences are not expected due to an assumed year-round activity and feeding at meso- and bathypelagic depths. Båmstedt (1979) measured respiration rates between 0.10 and 0.21 µL O_2 mg DM^{-1} h^{-1} for *E. hamata* in Kosterfjorden, western Sweden ("standard animals" of 4 mg DM, at 5–6°C, 0–200 m depth) with highest rates in spring (May) and lowest rates in winter (February). However, we found no seasonal differences. Båmstedt (1979) additionally reported a mean respiration rate of 0.11 and 0.86 µL O_2 h^{-1} per individual *E. bathypelagica* and *E. hamata*, respectively. Our experiments at 0°C revealed consumption rates of 0.36 and 0.44 µL O_2 $ind.^{-1}$ h^{-1}. The difference between the two chaetognath species was more pronounced in Sweden. However, respiration rates were still in the same range. Oxygen consumption rates between approximately 0.29 and 0.48 µL O_2 $ind.^{-1}$ h^{-1} (converted from a mean value of 0.805 µmol O_2 g^{-1} h^{-1} and a wet mass range of 0.0161–0.0268 g) measured for *E. hamata* at 5°C (Thuesen and Childress 1993) support our data. Differences between the data sets may be attributable to the smaller number of measurements by Thuesen and Childress (1993) compared to our study. Additionally, differences in temperature and chaetognath body mass could contribute to the different results (Kruse et al. submitted). Previous investigations showed that hydrostatic pressure seems to have little effect on the metabolic activity of chaetognaths (Childress and Thuesen 1993). These authors observed no significant variation in metabolic rates at 0.101 MPa (surface) and at 10.1 MPa (~1,000 m). During our study, oxygen consumption rates were measured at normal surface pressure on chaetognaths mostly originating from 500 to 1,000 m depth, with no significant differences between the sampled strata.

As our respiration rates may be underestimations, considering higher animal activities in situ, the daily body carbon respired is probably underestimated as well. Our calculated data suggest between 0.45 and 1.09% of body carbon respired per day, which is low compared to other Antarctic taxa. Other Antarctic zooplankton species, e.g. amphipods, copepods and euphausiids, usually have higher daily metabolic losses of carbon, varying from 0.44 to 2.75% (at −0.8 to −1.4°C, Ikeda and Mitchell 1982). However, our mean values for the three *Eukrohnia* species are similar to data presented for the Antarctic species *Sagitta gazellae* with an average metabolic loss of 0.5% (at −0.9°C, Ikeda and Kirkwood 1989). Hence, Antarctic chaetognaths may differ from other zooplankton species in this respect.

Conclusions

The chaetognaths *Eukrohnia hamata* and *E. bathypelagica* from meso- and bathypelagic depths had generally low oxygen consumption rates and hence low body carbon respiration,

which may indicate low food requirements in turn. *Eukrohnia bathypelagica* and *E. bathyantarctica* exhibit different modes of feeding and lipid storage. *Eukrohnia bathypelagica* was characterized by a lower but stage-dependent lipid level, whereas *E. bathyantarctica* contained a higher relative amount of total lipids and significant amounts of wax esters. The lipid reserves were not substantial with little seasonal differences during summer and winter in both species indicating the presence of year-round feeding. Based on our analyses of gut contents and trophic marker fatty acids, we assume that all three *Eukrohnia* species, including *E. hamata*, rely largely on copepods as prey (e.g. calanid copepods and *Paraeuchaeta antarctica*). Consequently, chaetognaths may exert a continuous predation impact on the copepod community. Feeding conditions for chaetognaths might be better in winter, when several copepod species overwinter in deeper layers (Schnack-Schiel and Hagen 1994; Hagen 1999; Auel and Hagen 2005). Similar fatty acid compositions during summer and winter point to a homogenous diet composition in meso- and bathypelagic waters, although the zooplankton community is known to vary due to seasonal vertical migration of e.g. copepods. The role of lipids in chaetognaths, also in terms of oil droplets, requires further clarification.

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