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Gut evacuation rate and grazing impact of the krill *Thysanoessa raschii* and *T. inermis*

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Abstract:
Gut evacuation rates and ingestion rates were measured for the krill *Thysanoessa raschii* and *T. inermis* in Godthåbsfjord, SW Greenland. Combined with biomass of the krill community, the grazing potential on phytoplankton along the fjord was estimated. Gut evacuation rates were 3.9 and 2.3 h^{-1} for *T. raschii* and *T. inermis*, respectively. Ingestion rates were 12.2 ± 7.5 µg C mg C^{-1} day^{-1} (n = 4) for *T. inermis* and 4.9 ± 3.2 µg C mg C^{-1} day^{-1} (n = 4) for *T. raschii*, corresponding to daily rations of 1.2 and 0.5 % body carbon day^{-1}. Clearance experiments conducted in parallel to the gut evacuation experiment gave similar results for ingestion rates and daily rations. Krill biomass was highest in the central part of the fjord's length, with *T. raschii* dominating. Community grazing rates from krill and copepods were comparable; however, their combined impact was low, estimated as <1 % of phytoplankton standing stock being removed per day during this late spring study.

1. Introduction

Krill occur in vast numbers in the northern seas. Here, they form an important zooplankton group that serves as a major component of prey for many marine animals (Mauchline & Fisher 1969; Astthorsson & Gislason 1997; Rosing Asvid et al. 2013). Krill perform diurnal vertical migrations throughout the season (Vestheim et al. 2013) and their grazing activity results in the production of carbon rich, fast sinking faecal pellets. Together, this makes krill a significant contributor to the biological pump, exporting carbon and nutrients from surface to deeper waters (Tanoue & Hara 1986). As a result, the large schools of krill greatly influence the transfer of energy and organic matter throughout the marine food web (Mauchline & Fisher 1969). Therefore, quantifying krill grazing in order to evaluate their impact on prey, together with their role in carbon sequestration, is important.

In this context, *in situ* techniques such as the gut fluorescence method (Mackas & Bohrer 1976) have been applied to estimate grazing dynamics of many zooplankton taxa (e.g. Kiørboe & Tiselius 1987; Dam & Peterson 1988; Perissinotto & Pakhomov 1996; Bernard et al. 2012). However, a number of problems related to the approach of estimating the gut evacuation rate, and hereby ingestion rates, have been identified. Studies have suggested that estimations of gut evacuation rates under starvation (which is the most common experimental procedure) will be lower than experiments conducted under continuous feeding conditions (Dam & Peterson 1988; Perissinotto & Pakhomov 1996). To simulate continuous feeding, non-fluorescent charcoal particles have been applied in experiments on the Antarctic krill *Euphausia superba* (Perissinotto & Pakhomov, 1996; Bernard et al. 2011). Perissinotto & Pakhomov (1996) found that gut evacuation rates were strongly correlated to krill feeding activity, showing faster evacuation rates under high feeding activity. Another important factor, which could lead to underestimation of the ingestion rates, is pigment destruction of Chlorophyll *a* (Chl *a*) to non-fluorescent end products during digestion (Båmstedt et al. 2000; Perissinotto & Pakhomov 1996). Furthermore, Dam & Peterson (1988) showed that gut evacuation rate was related to temperature and suggested an equation including the temperature dependency. However, bearing all these different parameters in mind, estimating ingestion rates based on gut fluorescence and gut evacuation rate is a useful method for studying zooplankton grazing on autotrophic organisms (Peterson et al. 1990).

In the Godthåbsfjord system (Nuup Kangerlua) SW Greenland, krill is dominated by *Thysanoessa raschii*. Other resident species include *T. inermis*, *T. longicaudata* and *Meganyctiphanes norvegica* (Agersted & Nielsen 2014). The fjord is draining the Greenland Ice Sheet to the open sea, and the run-off from the Ice Sheet has a strong influence on the fjord (Mortensen et al. 2011) and plankton composition (Calbet et al. 2011; Arendt et al. 2013). In relation to this, much attention has been given to describe the role of micro- and mesozooplankton in the fjord (Arendt et al. 2010; Calbet et al. 2011; Tang et al. 2011). However, knowledge about larger zooplankton organisms in the fjord is limited (e.g. Agersted et al. 2011; Agersted and Nielsen 2014).

The aim of the present study was therefore to estimate krill grazing impact on the phytoplankton biomass during late spring and to compare this with the potential grazing impact by copepods.

2. Materials and methods

All sampling and experiments were conducted on a cruise aboard RV Sanna (part of monitoring program Marine Basis Nuuk, Greenland Institute of Natural Resources), from May 7 to May 15, 2013 in Godthåbsfjord, SW Greenland (Fig. 1).

2.1. In situ measurements

Depth profiles of water temperature, salinity and fluorescence were obtained using a CTD profiler (SBE 19plus, SeaCat) equipped with a Seapoint Chlorophyll *a* Fluorometer and a Biospherical/Licor sensor. Water samples for Chl *a* measurements were taken using a 5 L Niskin water sampler at depths of 1, 5, 10, 20, 30, 40, 50, 100 and 400 m. Water was filtered through GF/F filters, and Chl *a* then extracted from the filters using 96% ethanol in dark and at room temperature for 12-24 h (Jespersen & Christoffersen 1987). The Chl *a* was analysed using a fluorometer (T-700, Turner Designs) before and after acid addition (1M HCl) in order to asses Chl *a* and phaeopigment concentrations in the natural sample. Fluorescence was calibrated to *in situ* Chl *a* measurements using a linear regression for all stations.

Krill were collected using a 2-m ring MIK-net (1500-µm mesh size, black) towed in oblique hauls to 140 m with a speed of 2-2.5 knots at stations FB1.5, GF3, GF7, GF10, GF13 and GF17 (Fig. 1). The net was fitted with a flow meter (G. O. Environmental, General Oceanics) in order to calculate the water volume filtered. Samples were preserved in buffered formalin (4% final concentration). A minimum of 400 krill from each sample were later identified to species and their body length measured (from tip of rostrum to end of telson, mm).

Copepods were collected at the same sites using a 45-µm modified WP2-net towed in vertical hauls from 140 m to the surface. Samples were fixed in buffered formalin (4% final concentration), and identified to either species or genus and developmental stage (by Arctic Agency, Poland). The prosome lengths of a minimum of 10 individuals for each copepod stage were measured. Carbon content of copepods was calculated using length-weight relationships from the literature (see Table 1 in Arendt et al. 2013).

2.2. Gut evacuation rate experiments

Two *in situ* gut evacuation experiments were conducted along the fjord at stations GF 7 and GF10 (Fig. 1). Krill were collected at night (2300 hr) from the upper 20 m using a 335 and 500-µm mesh size Bongo net fitted with non-filtering cod-ends (2-L). Immediately after retrieval, the krill was transferred to a 50 L insulated container filled with 0.5µm-filtered seawater. From here, 40-50 undamaged individuals of similar size were collected and carefully transferred to two cylinders (approximately 20-25 individuals per cylinder) hanging in a 50 L thermo-container filled with 0.5µm-filtered seawater. Each cylinder was equipped with a mesh screen bottom to allow sinking faecal pellets to be separated from the active krill, and so surrounding water could easily be exchanged. Prior to each incubation, four freshly collected individuals were processed for the measurement of their initial gut pigment content (total pigments being the summation of Chl *a* and phaeopigments). A concentration of 1.5 mg L^{-1} of non-fluorescent charcoal particles (<100 µm diameter) (Chemviron Carbon, Denmark) was added in order to ensure continuous feeding (Perissinotto & Pakhomov 1996). The amount of charcoal added (S, µg WW L^{-1}) corresponded approximately to the *in situ* concentration of particles, representing available food for the krill (Perissinotto & Pakhomov 1996). This was estimated by converting *in situ* Chl *a* concentrations to carbon by a C:Chl *a* conversion factor of 43.3 (Sejr et al. 2007), then to dry weight (Postel et al. 2000) and finally from dry weight to wet weight (Postel et al. 2000) using the equation:

$$\mathrm{S}\ (\mu\mathrm{g\ WW\ L^{-1}}) = \mathrm{Chl}\ a\ (\mu\mathrm{g\ L^{-1}}) \times 43.3 \times 2 \times 5 \qquad (1)$$

Experiments ran for 48 h at a constant temperature of 1.5°C, corresponding to ambient seawater temperatures. During the first hour of incubation, krill were sacrificed at approximately 5, 10, 15, 20, 40 and 60 min. Then after 2, 4, 8, 16, 24 and 48 h. Filtered seawater and charcoal were changed after 2 h of incubation. At each time point, four sacrificed krill were taken and added individually to vials containing 5 mL 96% ethanol.

These vials were left for approximately 24 h in dark for extraction of pigments, and the gut fluorescence was then measured on a fluorometer (Turner, TD-700), before and after acidification. Each krill was finally measured (body length, mm) and identified to species. Gut evacuation rates (k, h^{-1}) were derived from the slope of the regression of the natural log of total gut pigment vs. time. To avoid possible underestimations of k, the regression was made for measurements within the first 30 min of the experiment.

2.3. Grazing rates – gut fluorescence technique

Grazing rates were estimated using the gut fluorescence technique as described by Båmstedt et al. (2000). Ingestion rate (I, µg Chl *a* ind^{-1} d^{-1}) was calculated as:

$$I = G \times k \qquad (2)$$

Where G = initial gut pigment content (µg Chl *a* ind^{-1}) after removal of background gut fluorescence, and k = the gut evacuation rate (h^{-1}) (Båmstedt et al. 2000). For background gut fluorescence values, krill (n = 8) were incubated in 0.5µm-filtered seawater containing non-fluorescent charcoal particles for 48 h to empty their guts. Afterwards they were processed as described above and the background fluorescence was subtracted from the fluorescence obtained from the experimental animals. Background gut fluorescence (i.e. after 48 h) averaged 3% (± 1.3 standard deviation (SD), n=8) of initial gut fluorescence. No corrections were made for gut pigment destruction, except assessing phaeopigments in the fluorometrical calculation (Durbin & Campbell 2007; Bernard et al. 2012).

Since krill primarily feed during night in the surface where sampling was conducted, the daily ingestion values were calculated assuming that krill only feed in the euphotic zone for 4 h d^{-1} during this time of year. This assumption was made on the basis of sunset and sunrise data from the area (www.asiaq.gl) and acoustical data from the Oslofjord, Norway, showing that krill only stay in the euphotic zone during hours of darkness (Kaartvedt et al. 2002).

Chl *a* values were converted to carbon using a C:Chl *a* ratio of 43.3 (Sejr et al. 2007), and krill weight (W, mg C) was estimated from the length-weight regression in Agersted & Nielsen (2014):

$$\mathrm{W} = 7.25 \times 10^{-5}\mathrm{L}^{3.792}, \mathrm{r}^2 = 0.96 \qquad (3)$$

where L is krill length (mm).

Community grazing rates were calculated for each station as the product of the overall mean daily ingestion rates (µg C mg C^{-1} d^{-1}) for *T. raschii* and *T. inermis* and the total krill biomass of all four species (mg C m^{-3}) at each station. When calculating krill biomass, the estimate was an average for the upper 140 m. This will underestimate the biomass as krill concentrate in layers where the food concentration is high (Hamner et al. 1983; Price 1989). Therefore, we assumed that the krill swarms would be concentrated in a band of 10-15 m width (Simmard et al. 1986; Cox et al. 2009; Tarling et al. 2009). As a consequence, we multiplied the grazing impact by a factor 10 (i.e. assuming a concentrated band of 14 m). Average daily rations, expressed as a percentage of body carbon consumed per day (% body carbon d^{-1}) (Båmstedt et al. 2000) were furthermore calculated at each station.

2.4. Clearance experiments

A different approach to estimate krill grazing dynamics is to conduct grazing experiments, where clearance rates and thereby ingestion rates can be estimated. In order to assess the gut fluorescence method, ingestion rates were therefore also estimated from clearance experiments. Krill (*T. raschii* and *T. inermis*) were collected with the Bongo net at St. GF5 and transferred to cylinders with a false bottom, placed in containers filled with filtered seawater and non-fluorescent charcoal particles. Here they were allowed to empty their gut for 48 h. The low gut fluorescent individuals were then incubated for 2 h in 8 × 2-L polycarbonate bottles, two individuals per bottle, containing *in situ* Chl *a* rich seawater (5.6 µg Chl *a* L^{-1}). Control bottles (with no krill, *n* = 3-5) were incubated simultaneously. After 2 h of incubation, all the water from the control bottles and experimental bottles was filtered onto a GF/F filter, extracted and Chl *a* measured as described above. Prior to the incubation, 200 mL of the water was filtered for initial Chl *a* concentration. Clearance rate (*Cl*, mL mg C^{-1} h^{-1}) was then calculated as:

$$Cl = \left(\frac{V}{(W \times t)}\right) \times ln\left(\frac{C_1^* \times C_2}{C_1 \times C_2^*}\right) \quad (4)$$

Where *V* = volume of experimental bottle (mL), *W* = weight of krill (mg C), *t* = time of incubation (h), C_1 and C_2 = Chl *a* concentration (µg L^{-1}) in control bottles at start (t_{start}) and end (t_{end}) of experiment, respectively. C^*_1 and C^*_2 = Chl *a* concentration (µg L^{-1}) in experimental bottles at t_{start} and t_{end}, respectively. Ingestion rate (*I,* µg C mg C^{-1} h^{-1}) was consequently calculated as:

$$I = \left(\frac{C_2^* - C_1^*}{\ln\left(\frac{C_2^*}{C_1^*}\right)}\right) \times C \times Cl \quad (5)$$

Where C^*_1 and C^*_2 = Chl *a* concentration (µg L^{-1}) in experimental bottles at t_{start} and t_{end}, respectively, *C* = C:Chl *a* conversion factor (43.3; Sejr et al. 2007) and *Cl* = clearance rate (mL mg C^{-1} h^{-1}) (Frost 1972; Kiørboe et al. 1982). As in the gut evacuation experiment, we assumed that krill only feed on Chl *a* in the surface layers during 4 h at night. Therefore, the estimated ingestion rates (*I,* µg C mg C^{-1} h^{-1}) obtained by Eq. 5 were multiplied with 4 h.

2.5. Grazing by copepods

To compare krill grazing impact with the potential impact by copepods, grazing by the copepod community was estimated by applying the equations of Hirst & Bunker (2003) to estimate growth rates. For this, we assumed a gross growth efficiency of 33% (Hansen et al. 1997). The grazing estimate was based on the biomass of free spawning and egg carrying copepods, respectively (mg C m^{-2}; 0-140 m), *in situ* temperature (°C) and average Chl *a* concentrations (µg Chl *a* L^{-1}; 0-140m), applying the equation:

$$\mathrm{Log_{10}g} = \mathrm{a(T)} + \mathrm{b(log_{10}BW)} + \mathrm{c(log_{10}C_a)} + \mathrm{d} \quad (6)$$

Where g = weight-specific fecundity/growth (d^{-1}), a = 0.0186, T = temperature (°C), b = -0.288, BW = body weight (µg C ind^{-1}), c = 0.417, C_a = total Chl *a* concentration (µg Chl *a* L^{-1}) and d = -1.348 and -1.591 for broadcasters and sac spawners, respectively.

All means are in ± SD, unless other is stated.

3. Results

3.1. Hydrography

The water column structure changed along the transect from Fyllas Bank offshore (FB4) to the inner part of the fjord near the ice edge (G17) (Fig. 1, 2). High salinity water masses were measured at Fyllas Bank, influenced by the West Greenland Current. At the entrance of the fjord where the offshore region fuses with the fjord, vertical mixing occurred (Fig. 2). In the central and inner part of the fjord the water column was stratified with lower saline water masses in the upper layers due to meltwater run-off from land and glaciers. At depth, the water masses became warmer and more saline (Fig. 2A and B). Chl *a* levels generally followed the pycnocline, with subsurface peaks at Fyllas Bank (40-60 m) and in the central part of the fjord (20-60 m) (Fig. 2C). At the fjord inlet, low Chl *a* concentrations were observed due to vertical mixing. Furthermore, low Chl *a* concentrations was observed in the innermost part of the fjord close to the terminating glaciers (Fig. 2C).

3.2. Gut evacuation rate experiment

The decrease in gut pigment over time was measured for *T. raschii* at station GF7 and *T. inermis* at station GF10 (Fig. 3A, B) and was well described by an exponential decline. The gut evacuation rate was calculated as the slope of the regression of the natural logarithm of total gut pigment (Chl *a* and phaeopigments) content vs. time (Fig. 3C, D). To avoid possible underestimations of *k*, the regression was made for measurements within the first 30 min of the experiment (Fig. 3C, D). The highest evacuation rate was found on station GF7 for *T. raschii* (3.9 h^{-1}, r^2 = 0.83) (Fig. 3C, D; Table 1). At station GF10, *T. inermis* had an evacuation rate of 2.3 h^{-1}. Correspondingly, the gut passage time (1/*k*) for *T. raschii* was 0.26 h and 0.44 h for *T. inermis* (Table 1). Average initial gut content was 100 ± 42 ng total pigment ind^{-1} (*n* = 52) at station GF7 and 315 ± 193 ng total pigment ind^{-1} (*n* = 52) at station GF10, with the highest individual gut content of 582 ng total pigment ind^{-1} at GF10. Ambient seawater temperatures did not differ considerably between the two stations and were therefore not considered in the calculations.

Specific ingestion rates (µg C mg C^{-1} d^{-1}) together with daily rations (% body carbon d^{-1}) for *T. raschii* and *T. inermis* are shown in Table 2. Krill from GF10 (*T. inermis*) had the highest ingestion rate, with an individual maximum of 22.5 µg C mg C^{-1} d^{-1} and an average of 12.2 ± 7.5 µg C mg C^{-1} d^{-1}, *n* = 4. The daily ration was on average 1.2 ± 0.8% and 0.5 ± 0.3% body carbon d^{-1} (*n* = 4) at station GF10 (*T. inermis*) and GF7 (*T. raschii*), respectively (Table 2).

3.3. Clearance experiment

Results from the grazing experiment (clearance, ingestion and daily ration) are summarized in Table 3. Ingestion rates ranged from 6.1 to 19.7 µg C mg C^{-1} d^{-1} (11.5 ± 4.6 µg C mg C^{-1} d^{-1}, *n* = 8) and clearance rates from 26.2 to 86.6 mL mg C^{-1} d^{-1} (50.4 ± 20.7 mL mg C^{-1} d^{-1}, *n* = 8). Daily rations averaged 1.2 ± 0.5% body carbon d^{-1}, *n* = 8.

3.4. Krill abundance and biomass

Throughout the fjord, the total abundance and biomass of the four krill species *T. raschii*, *T. inermis*, *T. longicaudata* and *M. norvegica* were measured (Fig. 4A, B). Total abundance was notably higher at station GF7 and GF10 (285 and 170 ind m^{-2}, respectively (Fig. 4A)) than at the other stations (averaging 17 ± 6 ind m^{-2}, *n* = 3). Total abundance and relative

contribution of *T. raschii* and *T. inermis* to the combined abundance and biomass were generally high at all six stations (Fig. 4A, B), with *T. raschii* being the most abundant species, followed by *T. inermis* (75% and 22% of total abundance, respectively). At station GF7, *T. raschii* dominated with a contribution of 95% to both the total abundance and biomass (Fig. 4A, B). Despite a relative low abundance at GF13 (12%), *M. norvegica* contributed 57% of the biomass due to its larger size. It is furthermore noteworthy that *M. norvegica* only appeared in the inner part of the fjord.

Abundance of copepods was low at the entrance of the fjord (St. GF3, Fig. 4C). The offshore station (FB1.5) was dominated by *Calanus* spp. and *Metridia longa*, whereas *Microsetella norvegica* dominated at the innermost stations close to the Greenland Ice Sheet (Fig. 4D).

In general, the krill and copepod community biomass were very similar (Fig. 5). However, on station GF3 and GF7, krill biomass was considerably higher (85% and 94% of relative contribution, respectively). The highest copepod biomass (281 mg C m^{-2}) was found on station GF10.

3.5. Community grazing impact

Estimates of krill and copepod community grazing rates are summarized in Table 4. Since *Thysanoessa raschii* and *T. inermis* were the dominating krill species (Fig. 4A, B), we multiplied the mean ingestion rate from these two species to the total biomass of all four krill species at each station, without considering species-specific ingestion rates for the two remaining species. Krill and copepod community grazing rates largely followed the biomass patterns of these two groups, and were generally higher in the central part of the fjord where krill and copepod community biomass were highest. The highest grazing rate for krill was at GF7 (71.5 mg C m^{-2} d^{-1}) and on station GF10 for copepods (75.6 mg C m^{-2} d^{-1}). In general, the copepod community grazing rates were approximately equal to krill community grazing rates (Table 4).

Grazing impacts on phytoplankton by the krill and copepod community were low (in general <1% of standing stock grazed per day) due to high phytoplankton biomass (Table 4). However, at station GF17, low phytoplankton biomass resulted in higher grazing impacts (15.4% and 6.9% of standing stock grazed per day by krill and copepods, respectively). Apart from station GF17, the highest grazing impact was found by copepods on station GF10 (0.9% d^{-1}) and by krill on station GF7 (1.5% d^{-1}).

4. Discussion

The present study has given insight to the ecological role of krill in the Godthåbsfjord system. The grazing potential of krill was comparable to that of the copepod community. However, we did not find krill to be significant grazers on the phytoplankton standing stock during the late spring.

4.1. Gut evacuation rate

The gut evacuation rates *(k)* found for *T. raschii* and *T. inermis* are to our knowledge the first published values for these species and are higher than values found for other krill species (Table 5). Perissinotto & Pakhomov (1996) found estimates for *k* in *Euphausia superba* ranging from 0.10 to 0.42 h^{-1} in adults and from 0.22 to 0.31 h^{-1} in juveniles. Conversely, a recent study by Bernard et al. (2012) found slightly higher values for *k* in *E. superba* with 1 to

1.4 h^{-1} for adults and 1.1 to 1.9 h^{-1} for juveniles, which is comparable to our results. The differences in *k* between the present study and those with *E. superba* could be due to a number of factors. Dam & Peterson (1988) found that *k* was strongly related to temperature and our results are at a slightly higher temperature than those rates for *E. superba*. Furthermore, ambient food concentrations and quality/size structure of food can also have an effect on the gut evacuation rate (Dagg & Walser 1987; Dam & Peterson 1988; Perissinotto & Pakhomov 1996). In Perissinotto & Pakhomov (1996), the surface Chl *a* concentration was between 0.1 and 1.19 µg Chl *a* L^{-1} at stations where gut evacuation experiments were carried out. Similar concentrations were reported in Bernard et al. (2012), Gurney et al. (2002), and Perissinotto et al. (1997) (Table 5). In our study, ambient food concentrations were generally higher and surface Chl *a* concentrations in the upper 20 m at experimental stations averaged 4 µg Chl *a* L^{-1}. The difference in food concentration could most likely be the reason why we witness different *k* values. Furthermore, the fact that *Thysanoessa* spp. both are smaller species (<30 mm in length) than *E. superba* (<60 mm in length), and thus have higher metabolic and growth rates (Fenchel 1974; Banse 1982; Lentz 2000), could strengthen the observed differences. In Gurney et al. (2002), evacuation rates were estimated for a smaller Antarctic krill (*E. vallentini*), which had a maximum *k* value of 1.36 h^{-1}. In that experiment, ambient food concentrations and initial gut content were however considerably lower than in our study, which therefore might result in a lower evacuation rate. Additionally, the time interval to calculate *k* is an important factor. Data for calculation of *k* should be reduced to the exponential phase of the curve, as this will generate the most representative value for *k* under continuous feeding conditions (Dam & Peterson 1988; Peterson et al. 1990; Perissinotto & Pakhomov 1996). In the present study, *k* was only calculated from data points within the first 30 min of the experiment.

4.2. Ingestion and daily ration

Average daily ingestion rates and daily rations were comparable with values available from the literature for *T. raschii* (Agersted et al. 2011) and Antarctic species (e.g. Perissinotto et al. 1997; Gurney et al. 2002; Bernard et al. 2012). However, a recent study by Du & Peterson (2014) found higher ingestion rates and daily rations of *E. pacifica* (~20 mm) in the coastal upwelling zone of Oregon, USA. This study was however conducted in much warmer waters (Table 5). During high food concentration (22 µg Chl *a* L^{-1}), they found a maximum daily ration of 23% body carbon d^{-1}, while the daily ration averaged 4% body carbon d^{-1} under food concentration of 0.5-5 µg Chl *a* L^{-1} (Du & Peterson 2014), the latter Chl *a* concentrations comparable to the present study. Bernard et al. (2012) found mean daily rations of 0.3% for adults and 0.5% for juveniles of *E. superba*, and ingestion rates ranging from 0.4-358 µg (Chl *a* equiv.) ind^{-1} d^{-1}. In Meyer et al. (2010) they found a maximum daily ration of 10% body carbon d^{-1} (*E. superba*) and provide a linear relationship between daily rations of Antarctic krill and ambient food concentration (mg C m^{-3}). We applied the equation for late spring (see Table 7 in Meyer et al. 2010) to our own phytoplankton biomass data from station GF7 and GF10. This resulted in daily rations of approximately 0.6 and 1.5% body carbon d^{-1}, respectively, which is comparable to our estimates based on ingestion rates from the gut evacuation experiment. Nevertheless, due to omnivory (Mauchline & Fisher 1969; Sargent & Falk-Petersen 1981; Agersted et al 2011), *T. raschii* and *T. inermis* gain carbon from other food sources than phytoplankton, which explain this low daily ration when calculations are based on Chl *a* only. In other words, a low gut pigment content may not necessarily mean an empty gut. This is an important limitation when using the gut fluorescence technique, since the parameter measured originates from autotrophic prey only.

4.3. Clearance experiment

Estimates of average ingestion and daily rations from the gut evacuation experiment (Table 2) were similar to values obtained from the grazing experiment (Table 3). In Peterson et al. (1990), a comparison between the gut fluorescence technique and clearance experiments resulted in an underestimation of ingestion from the gut fluorescence method, and was attributed to an overestimation of gut passage time. However, the fact that ingestion rates and daily rations from the two present experiments did not differ, confirms that these two methods are comparable, and in addition suggests that the gut fluorescence technique is a useful tool for field investigations on zooplankton grazing impact on phytoplankton, as also suggested by Peterson et al. (1990).

4.4. Zooplankton distribution and grazing potential

Krill distribution throughout the fjord resembled previous studies conducted later in the season, with high krill abundance in the middle and inner part of the fjord (Agersted et al. 2011; Agersted & Nielsen 2014). However, krill biomass in the present study was low compared to previous estimates (Agersted & Nielsen 2014). The highest krill community grazing rate was found at GF7, where krill biomass was correspondingly high. Estimations of krill grazing impact on the phytoplankton standing stock were low, and krill are therefore considered to have a minor impact on the phytoplankton community in the Godthåbsfjord in the late spring. Nonetheless, our results are slightly higher than previous published estimates on grazing impact by krill in the Godthåbsfjord (Agersted et al. 2011). Agersted et al. (2011) found grazing impacts by *T. raschii* on phytoplankton standing stock ranging from 0.002-0.1% grazed per day, based on clearance rates from grazing experiments. In addition, grazing impacts by *E. superba* on the phytoplankton community in the Antarctic region (January, Antarctic summer) have been estimated to be <3% of total integrated Chl *a* d^{-1} (Perissinotto et al. 1997). Furthermore, krill grazing impacts were equivalent to that of the copepods. The copepod grazing rates found in the present study was similar to previous estimates by Arendt et al. (2010) and Tang et al. (2011). In general, the ecological role of krill could seem to be of in particular importance in the central parts of the fjord, where grazing from copepods and krill reached similar high rates.

When calculating krill community grazing, assumptions were made to take into account the behaviour of the krill. Considering that krill perform diel vertical migration (Simmard et al. 1986; Kaartvedt et al. 2002; Vestheim et al. 2013) and accumulate where food concentrations are high (Hamner et al. 1983; Price 1989), community grazing rates and grazing impacts would be much higher than calculated from the average krill concentration in the upper 140 m. This could additionally be supported by Perissinotto et al. (1997) who found much lower grazing impacts with net derived biomass estimates (0.0014–0.42% of total 300 m integrated Chl *a* consumed per day) than those obtained from acoustic data (0.01–2.68% of total 300 m integrated Chl *a* consumed per day). Furthermore, we saw measured mean Chl *a* concentrations to be higher in a band of approximately 10-20 m (Fig. 6), which supports our assumption. On the other hand, the diel migratory behaviour of krill would subsequently mean that the estimated grazing impact on phytoplankton is not exploited 24 h a day as observed by e.g. Simmard et al. (1986). Contrarily, krill could be feeding on other groups of plankton in the deep water during the day (Simmard et al. 1986, Onsrud & Kaartvedt 1998, Cleary et al. 2012).

In conclusion, the ecological role of krill in the Godthåbsfjord system during late spring, is of the same magnitude as the other important zooplankton group in the fjord, the copepods. The gut fluorescence technique showed to be a useful method for field investigation of krill grazing biology on autotrophic organisms. We document that the krill community in

Godthåbsfjord has sufficient food availability during late spring/early summer and that crustacean grazers do not control the phytoplankton community at this time of year.

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Tables

Table 1

Gut evacuation rate experiments with *T. raschii* (GF7, *n* = 52) and *T. inermis* (GF10, *n* = 52)

Station	Species	Length (mm)	Weight (mg C)	Time (GMT, hr)	Gut evacuation rate k (h^{-1})	Gut passage $1/k$ (h)	Average G_0 (ng total pigment ind^{-1})	Chl *a* ($\mu g\ m^{-2}$)
GF7	*T. raschii*	23 (±2.5)	10.4 (±0.002)	2307	3.9 (r^2 = 0.83)	0.26	99.83 (±42.4)	105
GF10	*T. inermis*	22.5 (±2)	9.8 (±0.001)	2312	2.3 (r^2 = 0.71)	0.44	314.6 (±192.8)	184.4

Gut evacuation rate (k, h^{-1}) and gut passage time ($1/k$, h), average initial gut pigment content (total pigments, G_0) (n = 4), in situ temperature (°C) in the upper 140 m, and in situ integrated chlorophyll *a* (Chl *a*) concentration in the upper 50 m. Experiments were conducted at 1.5 °C

Table 2

Average ingestion rates (µg C mg C^{-1} day^{-1}) and daily rations (% body carbon ingested day^{-1}) of krill from stations GF7 (*T. raschii*, *n* = 4) and GF10 (*T. inermis*, *n* = 4)

Station	Krill weight (mg C)	Initial gut content		Evacuation rate (k) (h^{-1})	Average daily ingestion rates		Daily ration %body C day^{-1}
		µg total pigment ind^{-1}	µg C ind^{-1}		(µg C ind^{-1} day^{-1})	(µg C mg C^{-1} day^{-1})	
GF7	18 (±9.9)	0.1 (±0.04)	4.5 (±1.9)	3.9	69.7 (±29.6)	4.92 (±3.2)	0.5 (±0.32)
GF10	10.6 (±0.0)	0.3 (±0.2)	14.2 (±8.7)	2.3	129.1 (±79.1)	12.2 (±7.5)	1.22 (±0.75)
Mean (*n* = 8)						8.55 (±6.6)	0.85 (±0.66)

Ingestion is calculated as the product of gut evacuation rate and initial gut pigment content. Ingestion was calculated only for 4 h in order to compensate for the diurnal migration in and out of the productive surface layers (see Materials and methods section)

Table 3

Grazing experiment

Exp. no.	# Krill	Krill weight (mg C)	Water Chl *a* (µg Chl *a* L^{-1})		Clearance (mL mg C^{-1} day^{-1})	Ingestion (µg C mg C^{-1} day^{-1})	Daily ration % body carbon day^{-1}
			t_{start}	t_{end}			
1	2	21.3	5.59	4.57	29.9	6.8	0.7
2	2	13.7	5.59	4.73	41.5	9.6	1.0
3	2	7.5	5.59	4.54	86.6	19.7	2.0
4	2	21.3	5.59	4.75	26.2	6.1	0.6
5	2	17.5	5.59	4.37	41.5	9.3	0.9
6	2	11.7	5.59	4.63	52.7	12.1	1.2
7	2	12.4	5.59	4.58	51.1	11.7	1.2
8	2	9.3	5.59	4.47	73.3	16.5	1.7
Mean (n = 8)					50.4 (±20.7)	11.5 (±4.6)	1.2 (±0.5)

Ingestion and daily ration calculated from clearance rates. Krill weight (mg C) is total weight per bottle, i.e., for two krill altogether. See "Materials and methods" section for details

Table 4

Estimates of community biomass and community grazing rates of krill and copepods in the upper 50 m of the water column

Station	Int. SS (mg C m^{-2})	Biomass (mg C m^{-2})		Grazing rates (mg C m^{-2} day^{-1})		Grazing Impact (% day^{-1})	
		Krill	Copepod	Krill	Copepod	Krill	Copepod
GF3	1,420	52.6	9.1	4.5	3.8	0.32	0.27
GF7	4,726.6	835	51.3	71.5	18	1.50	0.38
GF10	8,297.8	435	381.2	37.3	75.6	0.45	0.91
GF13	4,350	132.5	77.6	11.3	19.6	0.26	0.45
GF17	32.2	57.8	58.1	4.9	2.2	15.35	6.85

Krill community grazing rates at GF3, GF13, and GF17 are based on average daily ingestion rates found at stations GF7 and GF10. Integrated phytoplankton standing stock (Int. SS; mg C m^{-2}) is from the upper 50 m of the water column. Total grazing impact is presented as a percentage of phytoplankton standing stock grazed per day (% day^{-1})

Table 5

Results from gut evacuation rate experiments from studies on different krill species

Species	Size (mm)	Seawater temperature (°C)	Ambient food concentration (µg Chl *a* L^{-1})	*k* (h^{-1})	Ingestion rates (µg C ind^{-1} day^{-1})	(µg (Chl *a* equiv) ind^{-1} day^{-1})	References
T. raschii	20–25	1.29 (± 0.026)	4	3.9	69.7 (± 29.6)	–	This study
T. inermis	20–25	1.28 (± 0.3)	4	2.3	129.1 (± 79.1)	–	This study
E. superba (A)	40–50	0–0.5	0.1–1.19	0.10–0.42	–	0.04–3.6	a, b, c
E. superba (J)	15–25	0–0.5	0.1–1.19	0.22–0.5	–	0.5–0.6	b, c
E. superba (A)	–	−0.4–0.8	–	1–1.4	–	2.6–9	Bernard et al. (2012)
E. superba (J)	–	0.2–0.8	–	1.1–1.9	–	1.4–1.9	Bernard et al. (2012)
E. pacifica	20	10.5	0.5–5	–	150.5	–	Du and Peterson (2014)
E. pacifica	20	10.5	21.75	–	1,111.2	–	Du and Peterson (2014)
E. valentini (J)	>15	–	0.2–1.35	0.81	–	0.98–1.92	Gurney et al. (2002)

J juvenile; *A* adult

a: Pakhomov and Froneman (2004)

b: Perissinotto et al. (1997)

c: Perissinotto and Pakhomov (1996)

Figures

Fig. 1. (A) Greenland. The square box indicates the study area. (B) Sampling locations in the Godthåbsfjord and at Fyllas Bank, West Greenland.

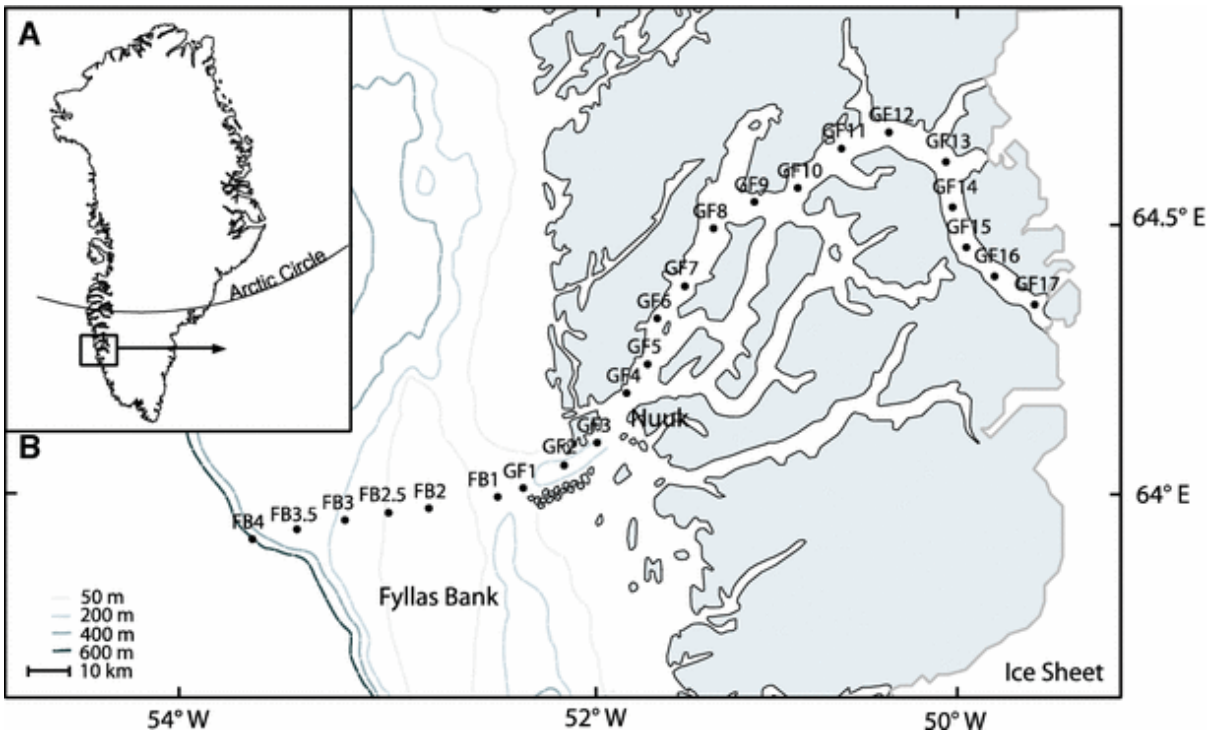

Fig. 2. Hydrography in the upper 140 m along the Godthåbsfjord (see Fig. 1 for stations), (A) temperature (B) salinity and (C) Chlorophyll *a* (µg l^{-1}). Vertical lines represent CTD data points.

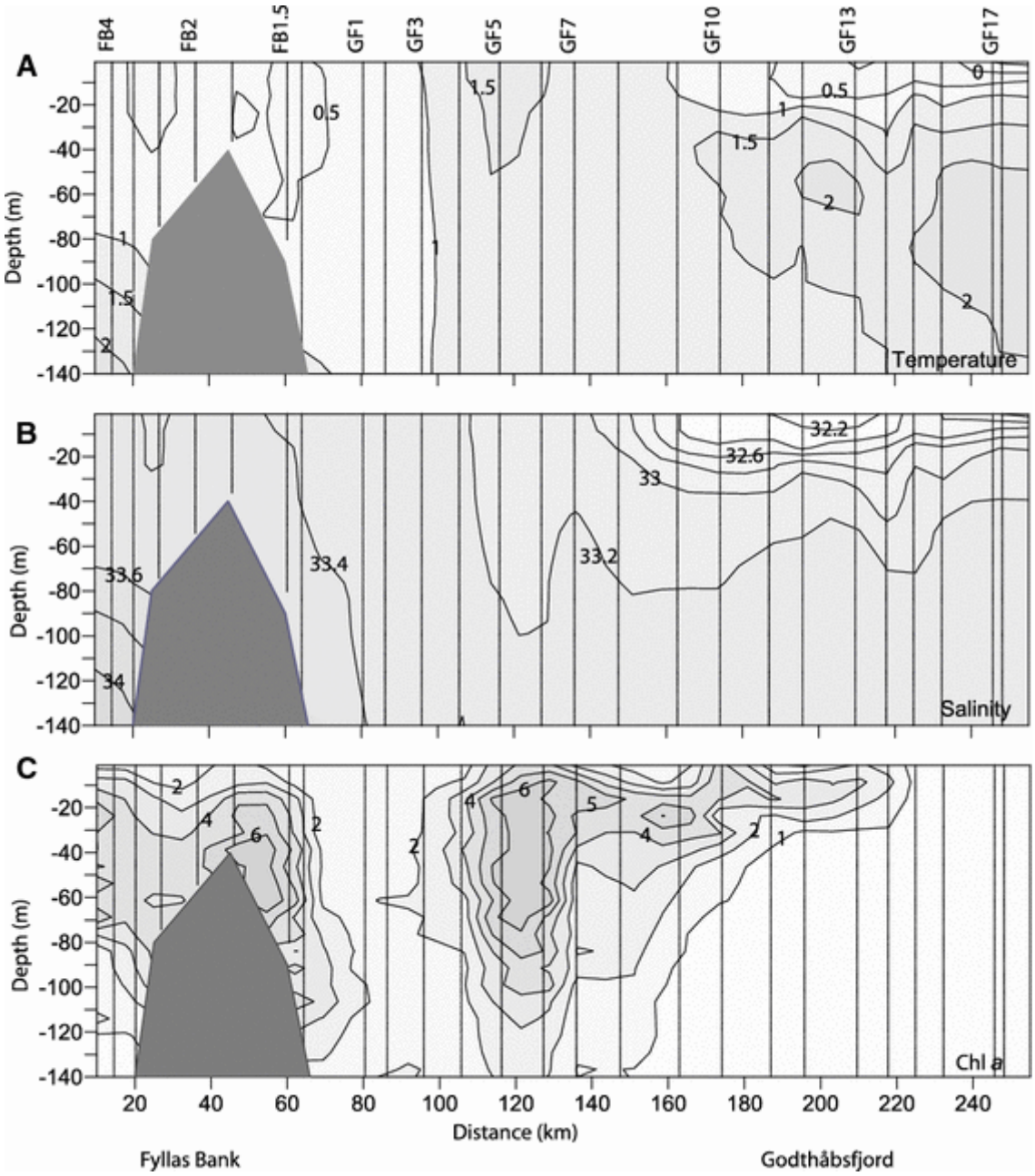

Fig. 3. (A+B) Krill gut evacuation over 48 hours. (C+D) natural log of gut pigment vs. time with standard deviation for *Thysanoessa raschii* (GF7, A+C) and *T. inermis* (GF10, B+D).

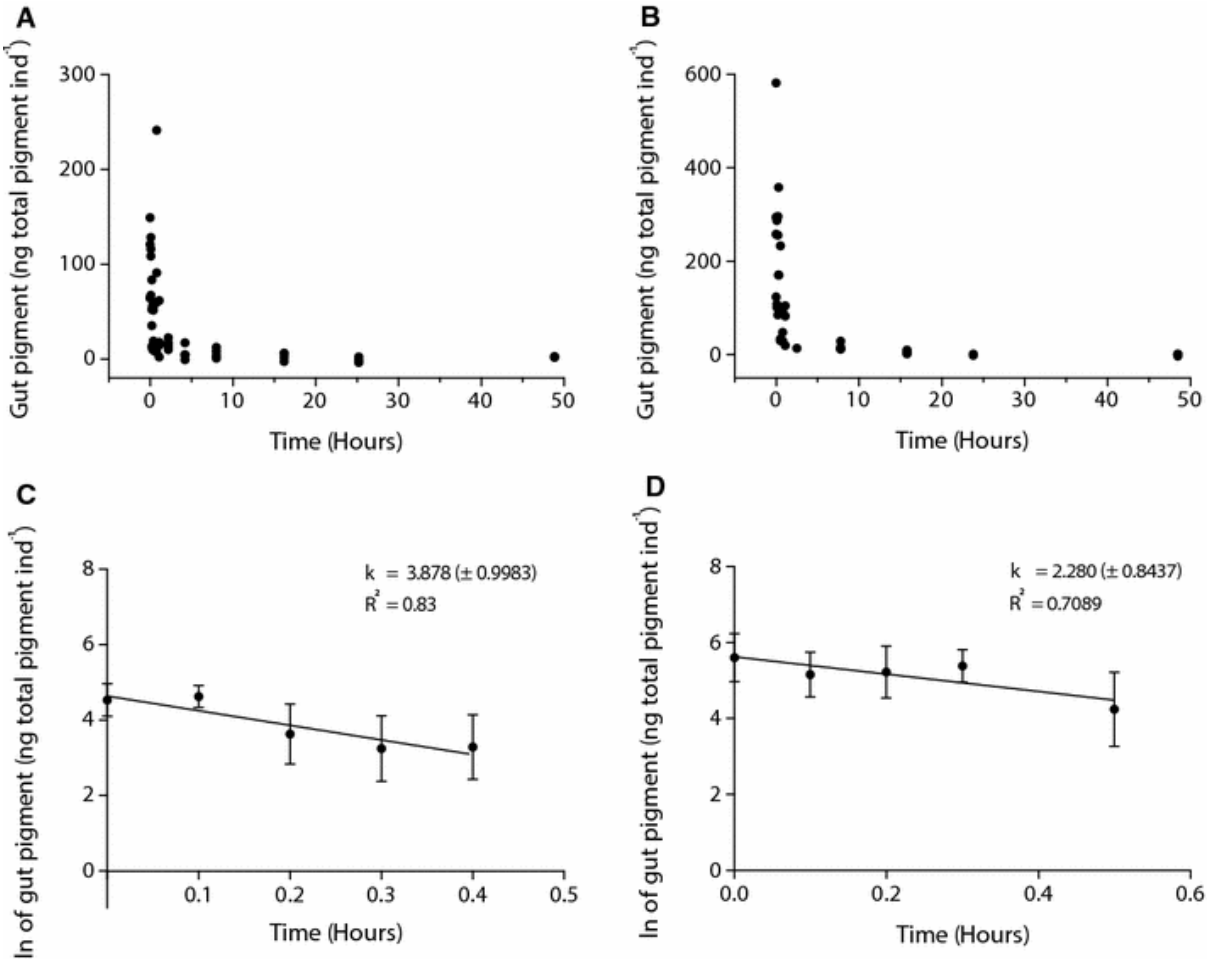

Fig. 4. Abundance (ind. m^{-2}) (A, C) and biomass (mg C m^{-3}) (B, D) for krill (upper 140 m) and copepod (upper 140 m) species, respectively. Relative contribution of the different krill and copepod species to (A, C) abundance (%), and (B, D) biomass (%) throughout the Godthåbsfjord.

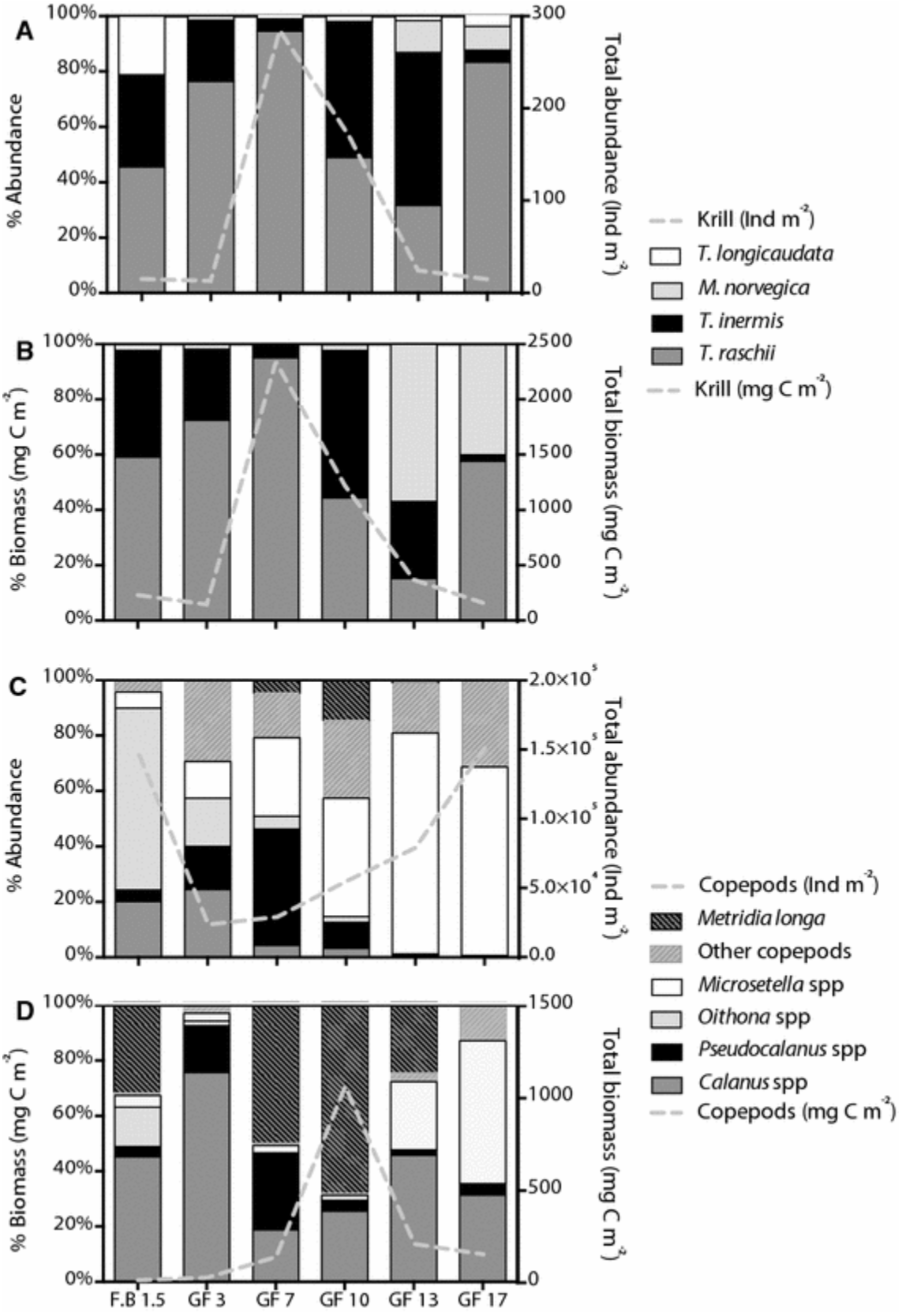

Fig. 5. Relative contribution of copepod and krill biomass (140 m) to the total combined biomass (%) throughout the Godthåbsfjord. Also illustrated is copepod (dashed line) and krill (full line) biomass (mg C m^{-2}).

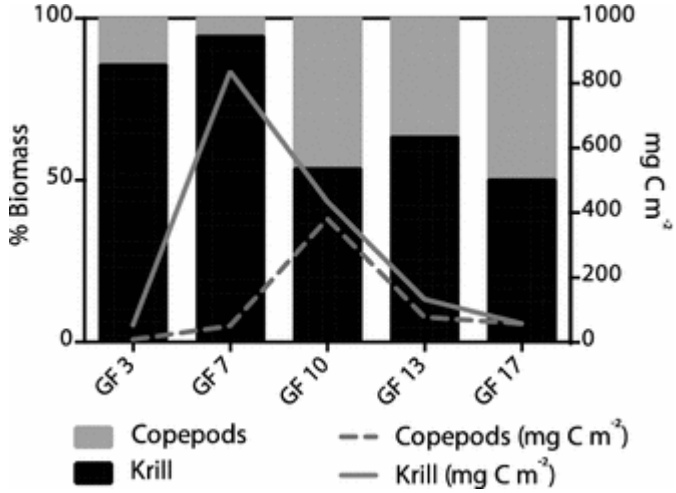

Fig. 6. Mean Chlorophyll *a* concentrations throughout the water column (140 m) from 4 stations in the Godthåbsfjord (GF3, GF7, GF10 and GF13). Standard deviation is shown as grey dotted lines.