Copepod grazing during an iron-induced diatom bloom in the Antarctic Circumpolar Current (EisenEx): I. Feeding patterns and grazing impact on prey populations

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Abstract

Feeding activity, selective grazing and the potential grazing impact of two dominant grazers of the Polar Frontal Zone, Calanus simillimus and Rhincalanus gigas, and of copepods <2 mm were investigated with incubation experiments in the course of an iron fertilized diatom bloom in November 2000. All grazers were already actively feeding in the low chlorophyll waters prior to the onset of the bloom. C. simillimus maintained constant clearance rates and fed predominantly on diatoms. R. gigas and the small copepods strongly increased clearance and ingestion of diatoms in response to their enhanced availability. All grazers preyed on microzooplankton, most steadily on ciliates, confirming the view that pure herbivory appears to be the exception rather than the rule in copepod feeding. The grazers exhibited differences in feeding behavior based on selectivity indices. C. simillimus and R. gigas showed prey switching from dinoflagellates to diatoms in response to the phytoplankton bloom. All grazers most efficiently grazed on large diatoms leading to differences in daily losses for large and small species, e.g. Corethron sp. or Thalassionema nitzschioides. Species-specific diatom mortality rates due to grazing suggest that the high feeding activity of C. simillimus prior to and during the bloom played a role in shaping diatom population dynamics.

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Keywords: Copepods; Diatom mortality; Grazing selectivity; Iron fertilization; Southern Ocean

1. Introduction

The high nutrient–low chlorophyll (HNLC) ecosystem of the Southern Ocean is characterized by a strong temporal and spatial variability of food resources for zooplankton grazers. Primary production is severely limited by the strong seasonal oscillations of daylight in high latitudes, by deep mixed layers and by the scarcity of essential micronutrients. Strongly enhanced phytoplankton growth following artificial infusion of iron has demonstrated the crucial role of iron in limiting primary production in open waters of the Southern Ocean (Boyd et al., 2000). Therefore, zooplankton grazers face a dilute, rapidly regenerating background community dominated by nanophytoplankton (Smetacek et al., 1990) most of the year. To cope with the shortage of phytoplankton, Southern Ocean copepods are generally omnivorous and fulfill a large percentage of their carbon
need by preying on microzooplankton (Froneman et al., 1996) or even resort to carnivorous feeding on other crustaceans during winter (Pasternak and Schnack-Schiell, 2001). Life cycle strategies are adapted to the strong seasonal variability in quality and quantity of food. A true state of diapause, however, has only been shown for Calanoides acutus (Atkinson, 1998). Most copepods undergo seasonal vertical migrations to deeper water layers but several species seem to adopt a dual over-wintering strategy and maintain a part of the population in the surface waters that continues to feed (Atkinson, 1991) which is also reflected in the accumulation of typical short term storage lipids by some species (Ward et al., 1996).

A rather productive area within the Southern Ocean is associated with the Antarctic Polar Front (APF) for which blooms are frequently reported (Laubscher et al., 1993; Bathmann et al., 1997). These are linked to increased iron concentrations following iceberg melting (Smetacek et al., 1997) or local upwelling events of deep water in connection with meandering of the APF producing areas of enhanced food availability for grazers (Strass et al., 2002). The transient bloom events are usually dominated by large microphytoplankton, on which copepod grazers have been shown to feed efficiently (Atkinson, 1994, 1995, 1996). Apparently, these pulses of productivity are sufficient to sustain the increased copepod populations of the area compared to most other sectors of the Southern Ocean (Pakhomov et al., 2000).

Considering the important role of the modern Southern Ocean in the global silicon cycle (Tréguer et al., 1995; Sarmiento et al., 2004) and in modulating atmospheric CO2 concentrations on geological timescales (Martin, 1990), key processes that influence the biogeochemical cycles need to be identified. In situ iron fertilization experiments provide the possibility to study functioning of the pelagic ecosystem and its effects on ocean biogeochemistry in iron-deplete and iron-replete states of the Southern Ocean. The current study was carried out with the goal to shed light on the structuring potential of copepod feeding on the development of an iron-induced diatom bloom at the APF in austral spring 2000. Feeding of dominant copepod grazers on diatoms and microzooplankton was investigated in incubation experiments before and during the artificially stimulated bloom. Special attention was given to feeding selectivity as it influences prey population dynamics and determines the ecological and biogeochemical efficiency of the grazer.

2. Materials and methods

2.1. Study area and iron fertilization

The in situ iron fertilization experiment “EisenEx” was carried out in the Atlantic Sector of the Southern Ocean (∼21°E, 48°S) during the cruise ANT XVIII/2 of R.V. Polarstern in austral spring (6–29 November 2000). A cyclonic eddy (approximately 120 km wide) shed by the Antarctic Polar Front (APF) was chosen as the experimental site and its centre marked with a drifting buoy (Strass et al., 2001). An area of about 40 km² around the buoy was fertilized with 4 tonnes of acidified iron sulphate solution (FeSO4) on three occasions at intervals of 8 days. Sulphurhexafluoride (SF6) was added as an inert tracer to the first iron infusion in order to relocate the iron fertilized “patch” (Watson et al., 2001). The fertilization induced a diatom
bloom with chl a concentrations and diatom biomass inside the fertilized patch increasing about fourfold (Gervais et al., 2002; Assmy et al., submitted for publication). “In-stations” were situated at the highest observed SF6 concentrations close to the centre of the iron-fertilized patch. “Out-stations” were located in waters well outside the Fe patch and contained background SF6 concentrations. Station 9 occupied 2 days prior to fertilization within the centre of the eddy was chosen as the initial reference station. For a detailed description of scientific activities, see Smetacek et al. (2001).

2.2. Mesozooplankton sampling and analyses

Abundance and composition of the copepod community were determined from samples collected with vertical hauls of a multiple opening and closing net (MN; Weikert and John, 1981) equipped with five 100 μm mesh nets. Depth strata down to 1500 m were chosen variably before each haul in order to resolve distinct bands of organisms observed with acoustic methods (Kasatkina et al., 2004). However, in all but one of the hauls (cast 38-09), at least two nets were closed in the upper 150 m of the water column. MN samples are available only for the first 10 days of the fertilization experiment due to logistical reasons. Details on the time, position and depth strata of the MN casts are presented in Table 1. Zooplankton samples were preserved in hexamethylenetramin buffered formalin (final concentration of 4%) and counted following standard protocols. Copepod species >2 mm were identified according to Razouls (1994). Sample collecting volumes were estimated by multiplying the net opening area (0.25 m²) by distance over which each net was towed in the corresponding depth layer. Depths of MN catches were determined by pressure probes. Copepod standing stock (individuals m⁻²) integrated over 150 m is based on the mean abundance estimated for the according depth strata assuming a homogeneous distribution of individuals. Mesozooplankton for feeding experiments were caught with the 300 μm Bongo net towed vertically at 0.3 m s⁻¹ through the upper 200 to 350 m of the water column. Immediately after the catch, the content of the cod-end was diluted with natural seawater. Actively swimming and apparently undamaged individuals were sorted with a large pipette under a stereo-microscope and maintained in 0.2 μm filtered seawater until the start of the experiment 2 to 5 h after the catch. All manipulations were performed in a cooled laboratory container.

2.3. Feeding experiments

In regular intervals at stations inside and outside the Fe-fertilized patch, mesozooplankton grazing activity was investigated with bottle experiments according to the method of Frost (1972). Natural seawater was used as a prey assemblage and was sampled with a CTD from the chl a maximum depth (18–50 m). Water from two 12-l Niskin bottles of the same cast, closed at the same depth, was mixed in a plastic carboy from which experimental bottles were filled. The incubation medium was not pre-screened to remove other grazers (nauplii and copepodite stages of Oithona sp.; see also Henjes et al., submitted for publication) as this procedure would have taken out large and chain-forming diatoms at the same time. Mesozooplankton were assumed to be already acquainted with the food organisms collected in the Niskin bottles.

The choice of grazer used in the various experiments depended on relative numerical abundance in the Bongo net catches and in the case of Calanus simillimus and Rhincalanus gigas the availability of sufficient numbers of healthy adult females. Three feeding experiments

<table>
<thead>
<tr>
<th>Expt. Date</th>
<th>Days since 1st Fe fertilization</th>
<th>Station Position</th>
<th>Initial chl a (μg l⁻¹)</th>
<th>Grazers (no./stages)</th>
<th>Duration (h)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 7 November</td>
<td>−2</td>
<td>Initial</td>
<td>0.56</td>
<td>Copepods &lt;2 mm (10/n.d.), R. gigas (5/C6♀)</td>
<td>24</td>
</tr>
<tr>
<td>2 10 November</td>
<td>2</td>
<td>9</td>
<td>0.52</td>
<td>C. simillimus (5/C6♀)</td>
<td>27</td>
</tr>
<tr>
<td>3 13 November</td>
<td>5</td>
<td>14</td>
<td>0.55</td>
<td>R. gigas (4/C6♀)</td>
<td>43</td>
</tr>
<tr>
<td>4 15 November</td>
<td>7</td>
<td>42</td>
<td>1.0</td>
<td>Copepods &lt;2 mm (20/n.d.)</td>
<td>41</td>
</tr>
<tr>
<td>5 16 November</td>
<td>8</td>
<td>45</td>
<td>1.15</td>
<td>C. simillimus (5/C6♀)</td>
<td>36</td>
</tr>
<tr>
<td>6 24 November</td>
<td>16</td>
<td>46</td>
<td>1.4</td>
<td>R. gigas (4/C6♀)</td>
<td>30</td>
</tr>
<tr>
<td>7 25 November</td>
<td>17</td>
<td>89:90</td>
<td>0.47</td>
<td>Copepods &lt;2 mm (15/n.d.), C. simillimus (5/C6♀)</td>
<td>34</td>
</tr>
</tbody>
</table>
were carried out for each of the three grazer categories: *C. simillimus*, *R. gigas* and an arbitrarily chosen mixture of copepods <2 mm. A first experiment for each grazer was completed during the initial stage of the iron fertilization (experiments 1 and 2, Table 2). Subsequently, experiments for each grazer were run at stations inside and outside the fertilized patch. These experiments will be referred to as “initial”, “in-patch” and “out-patch”. Grazers were incubated in 1 liter bottles and grazer densities ranged from 4 to 20 individuals l$^{-1}$, depending on grazer size and prior knowledge of their feeding rates. Details on experimental conditions are presented in Table 2. Bottles were kept on a plankton wheel, in the dark, at 4 °C. In every experiment, two control bottles and three replicates for every grazing treatment were incubated. Following the incubation, grazers, except the copepods <2 mm, were recovered and checked for mortality which was negligible. All incubation bottles were well mixed by rotating them end over end before any sub-sampling was performed. On board, initial and final concentrations of chl $a$ were determined from a 500 ml sub-sample following standard JGOFS procedures (Knap et al., 1996). The decrease of the chl $a$ in the grazing bottles compared to the control was 38% at most. The initially chosen duration of 24 h was extended in subsequent experiments up to 43 h as the first experiment failed to yield a clear reduction in chl $a$.

2.4. Diatom and microzooplankton analyses

One 200 ml sub-sample of incubation water from each control and grazing bottle was fixed with acidic Lugol’s solution for a microscopic analysis of diatom community composition. At the institute, cells in a 20–50 ml aliquot from every treatment were allowed to settle for 24 h in an Utermöhl chamber and diatoms counted if possible to the species level under an inverted microscope (Utermöhl, 1958). Abundant organisms were counted on single or several transects across the chamber, for less abundant cells one-half or the whole chamber was counted. This resulted in a mean of 956 cells in total being counted in the control (minimum 326 cells, maximum 2086 cells). For a specific prey organism, the same chamber surface was counted in the control and in the sub-samples from the grazing treatments. A detailed list of the diatom species and genera identified in the phytoplankton community of the fertilization experiment is presented by Assmy et al. (submitted for publication). For the purpose of this grazing study, more than 30 species were routinely identified and counted as such. However, species had to be subsequently regrouped into genera because clearance could not be calculated for each species individually.

### Table 3

Prey categories for the calculation of clearance and ingestion rates and list of species regrouped in these categories when determined.

<table>
<thead>
<tr>
<th>Category (pg C cell$^{-1}$)</th>
<th>Species (mean length, μm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chaetoceros spp. (21–582, mean 246)</td>
<td>Chaetoceros aequatorialis (25)</td>
</tr>
<tr>
<td></td>
<td>Chaetoceros atlanticus (18)</td>
</tr>
<tr>
<td></td>
<td>Chaetoceros bulbosus (15)</td>
</tr>
<tr>
<td></td>
<td>Chaetoceros convolutus (22)</td>
</tr>
<tr>
<td></td>
<td>Chaetoceros debilis (12)</td>
</tr>
<tr>
<td></td>
<td>Chaetoceros dicaeta (16)</td>
</tr>
<tr>
<td></td>
<td>Chaetoceros neglectus (5)</td>
</tr>
<tr>
<td></td>
<td>Chaetoceros peruviensis (20)</td>
</tr>
<tr>
<td></td>
<td>Chaetoceros indet. (n.d.)</td>
</tr>
<tr>
<td>Corethron spp. (231–2001)</td>
<td>Corethron inerme (90)</td>
</tr>
<tr>
<td>Fragilariopsis kerguelensis (35–182)</td>
<td>Corethron pennatum (90)</td>
</tr>
<tr>
<td>Guinardia spp. (202–2318)</td>
<td>Fragilariopsis kerguelensis (40)</td>
</tr>
<tr>
<td>Pseudo-nitzschia lineola (31–77)</td>
<td>Guinardia cylinresta (80)</td>
</tr>
<tr>
<td>Pseudo-nitzschia turgidula (20–61)</td>
<td>Guinardia delicata (40)</td>
</tr>
<tr>
<td>Rhizosolenia chunii (202–1072)</td>
<td>Pseudo-nitzschia lineola (95)</td>
</tr>
<tr>
<td>Thalassionema nitzschioides (8–49)</td>
<td>Pseudo-nitzschia turgidula (45)</td>
</tr>
<tr>
<td>Other diatoms (variable)</td>
<td>Rhizosolenia chunii (80)</td>
</tr>
<tr>
<td></td>
<td>Thalassionema nitzschioides (25)</td>
</tr>
<tr>
<td></td>
<td>Cylindrotheca closterium (80)</td>
</tr>
<tr>
<td></td>
<td>Dactyliosolen antarcticus (250)</td>
</tr>
<tr>
<td></td>
<td>Fragilariopsis rhombica (25)</td>
</tr>
<tr>
<td></td>
<td>Fragilariopsis obliquecostata (70)</td>
</tr>
<tr>
<td></td>
<td>Haslea sp. (90)</td>
</tr>
<tr>
<td></td>
<td>Membranella imposter (90)</td>
</tr>
<tr>
<td></td>
<td>Navicula spp. (35)</td>
</tr>
<tr>
<td></td>
<td>Nitzschia spp. (10)</td>
</tr>
<tr>
<td></td>
<td>Pleurosigma atlantica (120)</td>
</tr>
<tr>
<td></td>
<td>Pseudo-nitzschia heimi (90)</td>
</tr>
<tr>
<td></td>
<td>Pseudo-nitzschia prologanata (60)</td>
</tr>
<tr>
<td></td>
<td>Pseudo-nitzschia turgiduloides (90)</td>
</tr>
<tr>
<td></td>
<td>Proboscia sp. (300)</td>
</tr>
<tr>
<td></td>
<td>Rhizosolenia hebetata (500)</td>
</tr>
<tr>
<td></td>
<td>Thalassionema nitzschioides var. lanceolatum (90)</td>
</tr>
<tr>
<td></td>
<td>Thalassiothrix antarctica (1500) undet. centric diatoms (~20 μm, 20–50 μm, &gt;50 μm)</td>
</tr>
<tr>
<td>Silicoflagellates (626)</td>
<td>Dictyocha speculum (25)</td>
</tr>
<tr>
<td>Phototrophic nanoflagellates (8)</td>
<td>n.d.</td>
</tr>
<tr>
<td>Heterotrophic nanoflagellates (10)</td>
<td>n.d.</td>
</tr>
<tr>
<td>Heterotrophic dinoflagellates (160)</td>
<td>n.d.</td>
</tr>
<tr>
<td>Ciliates (254)</td>
<td>n.d.</td>
</tr>
</tbody>
</table>

Numbers in brackets in the category section indicates the min–max values of picograms carbon per cell. In the species section, the average elongation of the cell in the largest dimension is indicated.
only be calculated for diatoms or groups (see Table 3) of which at least 30 cells had been counted in the control (Atkinson, 1995). Diatoms were also placed into four size classes according to the mean cell length: <20 μm, 20–50 μm, 50–100 μm and >100 μm. However, these categories tend to cut across species or genera and their interpretation is of limited value considering that one aim of the present study was to gain insight in the interactions between key grazers and key diatoms in the Polar Front ecosystem. Diatom carbon was calculated using the geometric formulas according to Edler (1979) and the volume to carbon conversion factors proposed by Menden-Deuer and Lessard (2000).

Ciliates, heterotrophic dinoflagellates (hdinos), phototrophic nanoflagellates (pnanos) and heterotrophic nanoflagellates (hnanos) were enumerated from subsamples preserved in glutaraldehyde (final concentration=0.3%), stained with 3-6-diaminoacridine hemisulfate (proflavin) for 1 min (5 μg ml⁻¹ final concentration) and 4′6-diamidino-2-phenylindole (DAPI) for 4 min (5 μg ml⁻¹ final concentration). Discrete volumes were filtered onto 0.8 μm black Nuclepore filters. To achieve an even distribution for counting and measurement purposes, black filters were placed on top of pre-wetted Whatman GF/F backing filters. After filtration, the damp filter was placed on a slide. A drop of low fluorescence immersion oil was placed on top of the filter, which was covered with a cover slip and frozen at −20 °C. Samples were analyzed upon return to the laboratory. An Olympus BX-60 microscope equipped with 100-watt epifluorescence illumination was employed, with appropriate exciter/barrier filter sets for UV (335–365 nm), blue (435–490 nm) and green (510–560 nm) excitation. Dinoflagellates were distinguished from other flagellates based upon cell morphology and structure of the nucleus, especially the unique condensed chromosomes visible by DAPI staining. Heterotrophic and autotrophic cells were discriminated by the absence and presence of autofluorescent chloroplasts, respectively. The autotrophic category also includes mixotrophic cells. Cell abundance, dimensions and biovolumes were determined via quasi-automated color image analysis (Verity and Sieracki, 1993). A minimum of 200 plankton cells of each type was measured. The average coefficient of variation of triplicate counts of nanoplancton was 11%. Cell biovolume measurements were converted to carbon biomass using conversion factors based on literature values of carbon density of microplankton (Verity et al., 1992 and references therein). This method of enumeration precludes knowledge on the species level. Therefore, results on the clearance and ingestion have to been seen as an average for a large and very variable group of organisms. Nevertheless, they yield important information on trophic pathways and interactions. A detailed description of the microprotozooplankton community is given by Henjes et al. (submitted for publication).

2.5. Calculation of clearance and ingestion rates

All clearance and ingestion rates are presented as means of the three grazing replicates with the calculated standard deviation of the mean in brackets.

Clearance rates were calculated following the equation of Frost (1972) modified to:

\[ F = \ln(C_c/C_g) \times V / (n \times t) \]

(Atkinson, 1996) where \( F \) is the clearance rate (ml ind⁻¹ h⁻¹), \( C_c \) the final concentration in the control, \( C_g \) the final concentration in the grazing treatment, \( V \) the experimental volume (ml), \( n \) the number of grazers per bottle and \( t \) the duration (h) of the experiment. When no more cells of a prey species could be counted in the 50 ml sub-sample of a grazing treatment—which occurred occasionally for larger phytoplankton such as Corethron sp., Chaetoceros sp. or Rhizosolenia sp. but never in all three replicate bottles—the final cell concentration in that bottle was set to 1 cell l⁻¹ (roughly equivalent to 1 cell bottle⁻¹) to allow calculation of the clearance rate. Differences between clearance rates were tested with a homoscedastic, two-tailed \( t \)-test.

Total ingestion \( I_{total} \) (μg C ind⁻¹ day⁻¹) was determined as described below. Individual copepod ingestion rates \( I_i \) (ng C ind⁻¹ h⁻¹) were calculated by multiplying positive single clearance rates \( F_i \) of a given prey organism \( i \) with its final abundance in the control bottle \( C_{c,i} \). This was done for every experimental bottle separately and to the greatest detail possible, i.e. in the case of diatoms based on clearance rates for species and genera plus a category “other diatoms” regrouping left-over diatom counts. Total diatom ingestion for each replicate bottle was achieved by summation of all \( I_i \). This approach assumes that a grazer is not preying exclusively on a single food organism but fulfills its carbon needs from a variety of suitable prey. Furthermore, prey organisms may not be 100% homogeneously distributed in every replicate bottle. Ingestion based on an average clearance rate between replicate bottles may underestimate carbon intake by the grazer in the same way as ingestion based on an average clearance calculated for a large variety of food items. An additional
clearance on diatoms in general (“diatoms average”) was estimated from total diatom ingestion divided by the diatom carbon concentration in the control bottles.

The clearance and ingestion rates presented in this manuscript should be viewed as conservative since they underestimate (i) clearance on large prey items due to the length of the incubations and (ii) clearance on microphytoplankton and microzooplankton in general due to trophic cascading (sensu Nejstgaard et al., 2001).

2.6. Estimation of selective feeding behaviour

Selective feeding of copepods was characterized using the chi-square ($\chi^2$) goodness-of-fit test (Cowles, 1979), as described by Sokal and Rohlf (1969). The frequency distribution of food taxa in the copepod diet was compared with that in the environment. Selective feeding was indicated by a significant divergence of the distribution (Kleppel et al., 1996). Food selection on specific food taxa was quantified using the selectivity index (SI) $\alpha$, according to Chesson (1978). The calculation of $\alpha$ is based on the relative contribution of the taxon abundance to total abundance in the control and the relative contribution of the ingested prey organism to total ingestion. According to Chesson (1978), $\alpha = 0.5$ indicates non-selective feeding, $\alpha > 0.5$ a preference for a prey organism and $\alpha < 0.5$ discrimination against a taxon.

3. Results

3.1. Copepod abundance and community composition

The dominant copepod in the >2 mm size class was C. simillimus with a mean relative contribution to total copepod abundance of 57.5±26.0% in-patch and 69.2±17.5% out-patch (Fig. 1). On average, Calanus propinquus, C. acutus, Metridia spp., Pleuromamma robusta and R. gigas each contributed an order of magnitude less to total copepod abundance than C. simillimus. At one station, Metridia spp., dominated by Metridia gerlachei, accounted for 58% of the copepod community in cast 38-09 (in-patch day 3). Lubbockia aculeata, Heterorhabdus spp., Paraechaeta spp. and undetermined Calanoidea (all regrouped in the category “other copepods”) were frequently observed in the samples but always of minor numerical importance. Analysis of concomitant Bongo net catches confirmed the stable composition of the copepod community and the dominance of C. simillimus throughout the fertilization experiment (data not shown). During daytime 64% (±12.4) of the population of C. simillimus resided in the upper 80 m of the water column, during night time the percentage increased to 86% (±8.6; Table 1).

The standing stock of copepods between 1 and 2 mm was on average a factor of 2.6 higher than that of the larger copepods (Table 1). Taxonomic identification

Fig. 1. Composition of copepod standing stock >2 mm estimated from casts with a multiple opening closing net for the first 10 days of the fertilization experiment. “*” indicates casts taken in unfertilized control waters.
of the small copepod fraction is not available. Furthermore, the copepod standing stock estimates have to be heightened by substantial counts of the smallest fraction (<1 mm), mostly copepodite and adult stages of *Oithona* sp., estimated from Niskin bottle samples and presented elsewhere (Henjes et al., submitted for publication).

### 3.2. Temporal development of the prey field

The composition of the microplankton community on which copepod grazers could prey was estimated from the abundance of diatoms, heterotrophic dinoflagellates (hdinos) and ciliates in the control bottles and mirrors well the actual situation inside and outside the fertilized patch. Before the first iron fertilization (day −2), in the initial stage of the bloom (day 2) and outside the fertilized patch (days 5 and 17), diatoms accounted on average for 57%, hdinos for 26% and ciliates for 14% of total abundance. In-patch, the contribution of diatoms gradually increased to 81%, whereas for hdinos and ciliates it decreased to 9% and 8%, respectively (Fig. 2). Contribution of the silicoflagellate *Dictyocha speculum* varied from 0.1% to 6.2% but was usually around 3% and is not included in Fig. 2. Phototrophic and heterotrophic nanoflagellates (pananos, hnanos) theoretically dominated abundance with >90% but are not readily available to the all the larger grazers and therefore not included in the graph either.

Initially, small and delicate species like *Cylin- drotheca closterium*, as well as the heavily silicified pennates *Fragilariopsis kerguelensis* and *Thalassio- nema nitzschioiides* were the most important diatoms in terms of abundance. Biomass was at this stage dominated by *F. kerguelensis* and several large cylindrical diatoms, such as *Dactyliosolen antarcticus*, *Corethron pennatum* and *Guinardia* spp. In response to the iron addition *Pseudo-nitzschia lineola* and *Chaetoceros debilis*, both weakly silicified and chain-forming diatoms, showed the highest accumulation rates and dominated in-patch abundance at the end of the three week study. Large *C. pennatum* still contributed significantly to in-patch biomass on day 21. Out-patch, diatom biomass doubled due to an increase of the large cylindrical species *Rhizosolenia chunii*, *Proboscia alata*, *Guinardia cylindrus*, *C. pennatum* and *D. antarcticus* (Assmy et al., submitted for publication). Concerning ciliates and heterotrophic dinoflagellates, changes due to iron fertilization were not as dramatic as for the diatom community. Dinoflagellate and ciliate abundance showed an initial increase but thereafter declined or remained stable. Biomass of both groups nevertheless doubled, in the case of ciliates due to a shift to larger species, and was dominated by the size class 20 to 60 μm for both taxonomic groups (Henjes et al., submitted for publication).

### 3.3. Mesozooplankton feeding activity in response to the bloom

#### 3.3.1. *C. similimus*

Compared to the initial value of 3.7±0.9 μg C ind⁻¹ day⁻¹, total ingestion of *C. similimus* increased in the in-patch experiment on day 8 by a factor of 1.4 to 5.0±
0.8 μg C ind\(^{-1}\) day\(^{-1}\), whereas in the out-patch experiment on day 17 it decreased to 2.7±0.5 μg C ind\(^{-1}\) day\(^{-1}\). In all experiments, *C. simillimus* predominately fed on diatoms (Fig. 3A). The contribution of diatoms to total carbon ingestion varied between 71% and 88%. Ciliates were the second most important carbon source with a contribution of 9% to 13%. Taken together, carbon drawn from heterotrophic dinoflagellates and the silicoflagellate *D. speculum* supplied another 3% to 15%. Ingestion of diatoms inside the patch increased by a factor of 1.4 and remained unchanged in the out-patch experiment. Ciliate and dinoflagellate ingestion remained unchanged in the in-patch experiment but decreased substantially in the out-patch experiment: for ciliates by a factor of two, for dinoflagellates by one order of magnitude. Fig. 3B presents the contribution of various diatom species and genera to total diatom ingestion by *C. simillimus*. Consistently, *Corethron* spp., *Chaetoceros* spp., *Guinardia* spp. and *R. chunii* supplied the bulk of diatom carbon taken up by the grazer.

### 3.3.2. *R. gigas*

From the initial and out-patch experiments, total ingestion of *R. gigas* was estimated at similar values of 0.9±0.1 μg C ind\(^{-1}\) day\(^{-1}\) and 1.0±0.03 μg C ind\(^{-1}\) day\(^{-1}\). Inside the fertilized patch, ingestion increased by more than a factor of five to 5.4 μg C ind\(^{-1}\) day\(^{-1}\) (Fig. 4A). This increase is reflected in all major food sources. Diatom ingestion increased by more than one order of magnitude and feeding pressure on ciliates and hdinos doubled, although clearance rates for the latter two groups decreased (see below). In the initial and out-patch experiments, carbon ingestion of *R. gigas* was dominated by ciliates that contributed 52% and 54% to total ingestion, respectively. In-patch, diatoms were the dominant food source and accounted for 65% of the daily carbon intake. Hdinos generally supplied between 11% and 27% to the budget. Fig. 4B presents the composition of total diatom ingestion for *R. gigas*. Solely *Corethron* spp. supplied more than 50% of diatom carbon to *R. gigas* in the initial and out-patch experiments. In-patch, *P. lineola* and *Pseudo-nitzschia turgidula* accounted for 1.7 μg C ingested per copepod per day, i.e. 50% of the total diatom ingestion, *Corethron* spp. and *Chaetoceros* spp. combined for another 25%.

### 3.3.3. Copepods <2 mm

Total ingestion of the copepod fraction <2 mm was initially estimated at 0.177±0.085 μg C ind\(^{-1}\) day\(^{-1}\) and remained constant with 0.174±0.065 μg C ind\(^{-1}\) day\(^{-1}\) in the in-patch experiment on day 7 although diatom ingestion increased by a factor of 1.3. Outside the patch on day 17, total ingestion increased to 0.241± 0.076 μg C ind\(^{-1}\) day\(^{-1}\) (Fig. 5A) with diatom ingestion increasing by a factor of 2.5. Thus, ingestion by small copepods was one order of magnitude lower compared to *C. simillimus* and *R. gigas*. Feeding of the small copepods was not clearly dominated by either type of prey; diatoms contributed between 22% and 49%, ciliates 17% to 40%. All four groups of flagellates, i.e. hdinos, silicoflagellates, pnanos and hnanos, supplied carbon to these grazers, the cumulated value ranging from 35% to 61%. Diatom carbon was initially only provided by *Corethron* spp. and *Chaetoceros* spp (Fig. 5B). In-patch, *Fragilariopsis kerguelensis*, *P. turgidula* and other diatoms of the size fraction 20–50 μm were ingested additionally. In the out-patch experiment, the copepods <2 mm fed on all investigated diatom groups and species, with the bulk of carbon originating from *R. chunii*, *Corethron* spp. and *Guinardia* spp.
3.4. Clearance rates and grazing selectivity

3.4.1. C. simillimus

Clearance rates estimated for various prey organisms are presented in Table 4. When compared between diverse types of prey, clearance rates vary over one order of magnitude. No consistent relationship of the clearance rate to cell volume, cell carbon content and cell abundance could be discerned. For diatoms, however, the observed differences were related to cell size. In Fig. 6A, clearance rates for various diatom species or genera estimated in all three experiments are plotted against their respective size. The horizontal lines indicate the average clearance rates calculated for the four diatom size classes (see above). Both the detailed rates and the size class rate showed an increase with increasing size of the diatom cell. The average clearance for cells >100 μm was not higher than the rates determined for R. chunii (80 μm), Corethron spp. (90 μm) and P. lineola (95 μm), indicating that feeding efficiency might have reached a maximum at a cell size of approximately 80 μm. Average clearance for C. simillimus on diatoms remained stable; the slight increase from 12 to 14 ml ind⁻¹ h⁻¹ is not significant ($p>0.05$). Clearance calculated on the species or genus level was more variable when compared between experiments but changes showed no consistent trend. Clearance rates for pnanos and hnanos were consistently negative. Rates for hdinos and ciliates showed a stepwise decrease over the course of the three experiments; however, this variability was not related to the absolute abundance of the prey taxon.

3.4.2. R. gigas

Clearance rates determined in the experiments with R. gigas warrant a more detailed analysis. For diatoms, the average and detailed rates were similar in the initial and out-patch experiment but showed a strong increase in the in-patch experiment on day 16. In the latter incubation, mean clearance rates for different diatoms were again related to diatom size classes (Fig. 6B). Clearance by R. gigas of individual diatom species was

Fig. 4. Daily ingestion rates and prey composition of Rhincalanus gigas estimated from the incubation experiments. (A) Total ingestion. (B) Diatom ingestion. Data are archived in the information system PANGAEA at http://www.pangaea.de/PangaVista?query=@Ref26646.

Fig. 5. Daily ingestion rates and prey composition of copepods <2 mm estimated from the incubation experiments. (A) Total ingestion. (B) Diatom ingestion. Note the different y-axis scaling compared to Figs. 3 and 4. Data are archived in the information system PANGAEA at http://www.pangaea.de/PangaVista?query=@Ref26646.
not as apparent as for *C. simillimus*. However, as was the case for *C. simillimus*, rates for pnanos and hnanos remained below zero and clearance for hdinos and ciliates decreased with time. In Fig. 7, overall clearance rates on diatoms, hdinos and ciliates are plotted against the respective abundance of the prey taxon and for every experiment separately. The decreasing clearance on ciliates and hdinos over the course of all three experiments showed an inverse relationship with abundance. In the initial and out-patch experiments, variation of clearance between the three different taxa also displayed this correlation. However, differences of the clearance rates between diatoms, hdinos and ciliates from the in-patch experiment showed no relationship with absolute abundance of the three groups. These results possibly indicate selective feeding and a change in foraging behaviour and will be dealt with below.

### 3.4.3. Copepods <2 mm

Clearance rates estimated for the small fraction of grazers either remained constant over the course of the three experiments or increased (see Table 4). Clearance rates for ciliates, hdinos, pnanos, hnanos and the silicoflagellate *D. speculum* were always positive and remained constant. This is also true for the diatom genera Chaetoceros spp. and Corethron spp. All other rates determined for diatoms, except diatoms >100 μm, gradually increased from initially negative values to all positive values in the out-patch experiment. Large standard deviations, however, rule out a statistical significance of this increase for 60% of the rates (Student’s *t*-test, *p* < 0.05). Variability in clearance rates for different taxa or between experiments showed no consistent relationship with abundance, size or carbon content of the prey. Only in the out-patch experiment, where grazing on diatoms was strongest, could a tendency be resolved between cell size and clearance rate (Fig. 6C). However, this was only evident in the average rates of the cumulative size classes.

#### 3.5. Selective grazing

In a first step, frequency distribution of diatoms, silicoflagellates, hdinos and ciliates in the environment and the diet were compared. The contribution of pnanos and hnanos was omitted as the small flagellates were by one to two orders of magnitude more abundant than the other prey taxa which would have biased the SI towards exceptionally high positive selectivity of the less abundant taxa. Values calculated for *χ*² and the level of significance are given in Table 5. The *χ*² test indicated selective grazing in all experiments except the initial incubation with *C. simillimus* on day 2. Subsequently, to determine the selectively grazed prey, the SI α was estimated for diatoms, hdinos and ciliates. Results are presented in Fig. 8. Error bars indicate the standard deviation.
deviation of the mean calculated from three replicate grazing bottles.

*C. simillimus* (Fig. 8A) was not grazing selectively in the initial experiment and the SI for the three prey taxa is not significantly different from 0.5. In-patch, a drift could be observed towards a positive selection for diatoms and an avoidance of hidnos. The last experiment, in which copepods caught inadvertently on the edge of the patch were incubated in sample water with low microplankton abundance from outside the patch, shows a further specialization on diatoms and avoidance of hidnos with development of the bloom. In both experiments, in-patch and edge-patch, ciliates continued to be ingested invariantly according to their relative contribution to abundance in the environment.

Grazing of *R. gigas* (Fig. 8B) was selective in all three experiments according to the results of the $\chi^2$ test. The inverse relationship of clearance rate and abundance (see again Fig. 7) is reflected in a positive selection of ciliates and hidnos and overall avoidance of diatoms in the initial and out-patch experiment. In-patch, *R. gigas* displayed a very different grazing behavior. Diatoms were ingested proportionally to their presence in the environment, hidnos were apparently avoided and ciliates selected.

Fig. 8C presents the results for the small copepod fraction. The values estimated for $\chi^2$ indicate selective grazing in all three experiments, albeit with large standard deviations. These incubations included a mixture of copepod species and stages and variability in the type of prey ingested—but not the total amount of carbon ingested—was strong. The overall trend for the initial and in-patch experiment, 1 week after the first Fe infusion, indicated selection for ciliates, invariant ingestion of hidnos and avoidance of diatoms. As was the case for *C. simillimus*, small copepods in the last experiment originated from the edge of the patch. In this incubation, feeding seemed to be proportional to the contribution of the groups in the environment. A slight
avoidance was indicated for hdnos, as was already observed for the larger grazers.

3.6. Differential mortality of prey populations

Mortality rates for diatoms as % of the in-patch standing stock per day (% s.s. day\(^{-1}\)) were calculated based on available ingestion rates, in situ diatom standing stock and abundance estimates of copepod grazers. For *C. simillimus*, a detailed calculation at three points in time is presented in Table 6. For day 2 and day 8, mortality is based on ingestion rates determined for several diatoms in incubation experiments 2 and 5, and *C. simillimus* standing stock estimates from casts 14-03 and 45-06, respectively. Cast 45-06 on day 7 was chosen as no MN data are available for day 8. For day 21, maximum ingestion rates determined in the three experiments and the maximum abundance of *C. simillimus* determined in during the study are assessed. Copepod standing stock was integrated over 150 m as the larger grazers showed diurnal vertical migration (DVM) behavior. The DVM potentially modulates the grazing impact by a factor of 1.3 during a diel cycle. Considering the lack of net casts to resolve a complete 24 h cycle and conservative estimates of clearance and ingestion, daily mortality calculations do not take into account dial changes of grazer abundance in the mixed layer. Overall mortality of diatoms was highest on day 2, with 15.2% s.s. day\(^{-1}\) and gradually decreased to 9.3% s.s. day\(^{-1}\) on day 8 and 5.5% s.s. day\(^{-1}\) on day 21. Loss rates of species or genera ranged from 0.3% s.s. day\(^{-1}\) for *T. nitzschioides* to 44% s.s. day\(^{-1}\) for *Corethron* spp.

The maximum ingestion rates of diatoms by *R. gigas* and 1–2 mm copepods were compared with their respective standing stock estimated from cast 45-06 (Fig. 1) and diatom standing stock at station 46 (Table 6), resulting in an average diatom mortality of 1.4% s.s. day\(^{-1}\) due to grazing of *R. gigas* and 1.7% s.s. day\(^{-1}\) due to the small copepods.

Standing stock estimates for hdnos and ciliates and the silicoflagellate *D. speculum* are not available. However, based on counts of tintinnids and dinoflagellates at station 46 (Henjes et al., submitted for publication), mortality for these two prey taxa amounted to 12.8% and 1.4% s.s. day\(^{-1}\), respectively, due to grazing activity.
of *C. simillimus*, *R. gigas* and copepods 1–2 mm combined (data not shown).

4. Discussion

4.1. Composition of the copepod grazer community

The grazer community during EisenEx was a typical sub-Antarctic assemblage characteristic for waters north of the PF (e.g. Perissinotto, 1992; Atkinson, 1996; Bernard and Froneman, 2003). In general, abundance and biomass of mesozooplankton are enhanced in the PFZ compared to the Permanently Open Ocean Zone (POOZ) and dominated by copepods (Pakhomov et al., 2000). Euphausids and salps were of minor importance in the study area (data not shown). In the >2 mm size class, late copepodite and adult stages of *C. simillimus* were the key grazers inside and outside the iron fertilized patch with abundances at the higher end of the published range and comparable to concentrations of 273 to 1055 individuals m\(^{-3}\) estimated by Perissinotto (1992) in the vicinity of the Prince Edward Archipelago. Compared to *C. simillimus*, *R. gigas* was of minor numerical importance in the upper 150 m with maximum concentrations reaching 50 individuals m\(^{-3}\). This peculiar giant among copepods with an adult body size of almost 9 mm (Ommanney, 1936) is found throughout the Southern Ocean, with highest abundances near the PF (e.g. Froneman et al., 2000). The duration of its life cycle is still under debate and a regional variation between 1 year in the northern waters including the PFZ and 2 years in the Weddell Sea is discussed (Ward et al., 1997). According to Ward et al. (1997), the northern population of *R. gigas* reaches the surface waters in November so the timing of EisenEx probably coincided with the arrival of this grazer in the upper water column. In comparison, Atkinson (1991, 1998) proposes a 1 year life cycle for *C. simillimus* with mating in the top 250 m in early spring followed by a rapid development of the population and possibly a second generation in the same year.

Community composition and grazing pressure of copepods <2 mm was inadequately addressed in this study. The large-meshed sampling gear used was not fit to retain the small and ubiquitous cyclopoid copepod *Oithona* sp., which was present in the water column with an average of 18,000 individuals m\(^{-3}\) in the upper 150 m (Henjes et al., submitted for publication). Grazing experiments in this study were conducted with a mixture of species with 1–2 mm body length. This fraction is typically dominated by *Ctenocalanus* sp. or *Clausocalanus* sp., both widespread and very abundant in the PFZ in spring (Pakhomov et al., 2000; Dubischar et al., 2002). Although standing stock of this grazer fraction was up to a factor of three higher than the copepods >2 mm (Table 1), its grazing impact appeared to be limited (see below).

4.2. Feeding response to the iron induced diatom bloom

Table 6
Mortality calculation (% of standing stock day\(^{-1}\)) of various diatoms due to the grazing activity of *Calanus simillimus*

<table>
<thead>
<tr>
<th>Day 2—station 14</th>
<th>Day 8—station 45/46</th>
<th>Day 21—station 107</th>
</tr>
</thead>
<tbody>
<tr>
<td>Community I (mg C m(^{-2}) day(^{-1}))</td>
<td>Prey standing stock (80 m) (mg C m(^{-2}))</td>
<td>Mortality (% s.s. day(^{-1}))</td>
</tr>
<tr>
<td>Chaetoceros spp.</td>
<td>13.5 36 37.8 12.6 68 18.6 17.1 218 9.3</td>
<td></td>
</tr>
<tr>
<td>Corethron spp.</td>
<td>45.2 103 44.0 63.4 211 30.0</td>
<td></td>
</tr>
<tr>
<td>F. kerguelensis</td>
<td>0.5 30 1.6 3.9 68 5.7</td>
<td></td>
</tr>
<tr>
<td>Guinardia spp.</td>
<td>18.5 54 34.1 7.7 126 6.1</td>
<td></td>
</tr>
<tr>
<td>P. lineola</td>
<td>3.6 19 19.4 11.9 52 22.8</td>
<td></td>
</tr>
<tr>
<td>P. turgidula</td>
<td>0.9 8 11.0 2.7 22 12.3</td>
<td></td>
</tr>
<tr>
<td>R. chunii</td>
<td>n.d. 60 n.d. 7.7 31 24.9</td>
<td></td>
</tr>
<tr>
<td>T. nitzschioides</td>
<td>0.2 21 0.7 n.d. 28 n.d.</td>
<td></td>
</tr>
<tr>
<td>Total diatoms</td>
<td>135.7 895 15.2 121.7 1304 9.3</td>
<td></td>
</tr>
<tr>
<td><em>C. simillimus</em> (ind m(^{-3}) (150 m))</td>
<td>51,400 32,100 51,400 165.1 3508 165.1 51,400</td>
<td></td>
</tr>
</tbody>
</table>

See text for further explanation.
composition of the grazer changed little in response to the iron induced diatom bloom. A single spring time grazing estimate of 4.2 μg C ind⁻¹ day⁻¹ is available in the literature (Mayzaud et al., 2002)—based on gut fluorescence at environmental chl a concentrations of 0.5 μg l⁻¹—and is comparable to the results from EisenEx. Diatom clearance of *C. similimus* remained constant and ingestion inside the fertilized patch increased with diatom abundance by a factor of 1.4. Similar observations are reported by Atkinson and Shreeve (1995) studying the response of a copepod community to the spring bloom in the Bellingshausen Sea where *C. propinquus*, *M. gerlachei* and *Oithona* sp. were already actively feeding in ice covered waters with ~0.1 μg chl a l⁻¹. Copepods there maintained similar clearance rates over a fourfold increase of chl a from 0.8 to 3.2 μg l⁻¹ and the authors speculated that feeding must at times be severely food limited given that the same feeding effort resulted in approximately half the carbon intake.

*C. similimus* has been termed an omnivorous copepod (Atkinson, 1998) but detailed information on the composition of its diet with changing environmental conditions is scarce. At a sub-Antarctic site near South Georgia, in summer with concentrations of 0.8 μg chl a l⁻¹, algal carbon and heterotrophic food roughly contributed equal amounts to the grazer’s diet (Atkinson, 1996). During a study at the PF at the Southern Ocean contribution of phytoplankton to daily dietary carbon intake was reported to be as low as 3% in late summer with chl a concentrations in the environment of 0.6 μg l⁻¹ (Urban-Rich et al., 2001). Further evidence for omnivorous feeding and variable diet composition in response to availability is presented by Ward et al. (1996) who analyzed lipid content, hydrocarbons, lipid class and fatty acid composition in PFZ copepods from an intense diatom bloom (up to 20 μg chl a l⁻¹) and a post-bloom situation (<1 μg chl a l⁻¹) 4 weeks later. High amounts of pristane were found in *C. similimus* from both samplings, indicating that it was actively feeding. However, a lack of the polynye diatom marker C_{21:6} indicated that diatoms were not the major food source in the post-bloom situation, but that microheterotrophs were important carbon suppliers instead (Ward et al., 1996). Results from this study solidify the perception of *C. similimus* as an omnivorous grazer and high feeding activity even at low food availability. The clear dominance of siliceous organisms in the grazer’s food spectrum observed during EisenEx has not been reported so far.

In contrast, *R. gigas* responded to the iron induced bloom with apparently increased feeding activity and switch in diet composition from mainly heterotrophic prey to diatoms. Incubation experiments before the bloom indicate daily carbon rations below respiratory demand (Schultes et al., in press). Correction of grazing rates for trophic cascading (Klaas et al., in preparation) and comparison with gut fluorescence measurements (Schultes et al., in press) lead to the conclusion that *R. gigas* refers to carnivorous and detritivorous feeding before the bloom, which cannot be estimated from incubation experiments. In-patch, clearance of diatoms increased threefold thereby increasing diatom ingestion by an order of magnitude. These results confirm previous reports on seasonal changes in the feeding activity of *R. gigas*: in summer/bloom situations phytoplankton is the major food source; in spring and autumn, or pre- and post-bloom situations, protozoans, crustaceans and amorphous debris are additionally ingested (Atkinson, 1998).

Evidence for a response of the fraction of copepods <2 mm to the iron fertilized bloom is ambiguous. In the in-patch experiment, diatom ingestion appeared to be slightly enhanced but the difference is not significant due to large standard deviations. A strong increase in diatom clearance and ingestion was also noted in the out-patch incubation experiment. Furthermore, ingestion rates estimated from gut fluorescence increased inside the patch by a factor of two (Schultes et al., in press). Although not exclusively observed for grazers inside the fertilized patch, feeding on diatoms by the small copepods appeared to increase throughout the cruise.

Diet composition of the small grazers encompasses all prey organisms available in the environment including pnanos and hnanos. Trophic cascades in bottle incubations make it difficult to estimate feeding rates on nanoflagellates (Nejstgaard et al., 2001) and corrections with a simple ecosystem model indicate that ingestion rates of small copepods for nanophytoplankton and nanoozooplankton are underestimated by 20% to 50% (Klaas et al., in preparation). Model results also indicate that ingestion of nanoflagellates is low, but detectable, for *C. similimus* and nil for *R. gigas*. Younger copepodite stages of *R. gigas* actually could feed on the nanoplankton-sized fraction: the diet composition of C V was not different from the adult stages, C III/IV, however, also grazed on hnanos that supplied 3% to 8% of total carbon ingested (data not shown). These dietary differences between developmental stages and species lead to an effective partitioning of food among copepods (Atkinson, 1994) in an environment with low microplankton standing stock.
4.3. Selective feeding and feeding behavior

Copepods show two basic types of foraging behavior: suspension feeding with continuous low amplitude flapping of the second maxillae to accumulate mostly smaller cells, and ambush feeding, in which motile prey and large cells are detected individually with an active, oriented capture response. Copepods switch back and forth between both types of feeding behavior in a mixture of particles with different quality and quantity in order to maximize carbon intake with minimum feeding activity (Price and Paffenhöfer, 1986 and references therein; see also the review by Price, 1988). The iron induced diatom bloom had a major impact on the particle composition of the prey field. In response copepods showed variable changes to their feeding behavior.

Results for R. gigas strongly suggest that it resorted predominantly to ambush feeding in times of scarce supply of phytoplankton and changed foraging behavior in response to the diatom bloom. In the initial and out-patch experiments, motile cells are selected over diatoms. Exceptions make large diatoms, especially Chaetoceros and Corethron that both possess spines. Ambush feeding implicates detection of motile prey via mechanoreception, by using the receptors on the first antennae to sense hydrodynamic disturbances generated by prey movement (e.g. Landry, 1980; Price, 1988; DeMott and Watson, 1991). Suspended particles create deformations in the flow streamlines of the feeding current (Legier-Visser et al., 1986). Similar to the generation of a pressure wave these deformations extend in front of the particle (Price, 1988), and spines will generate a stronger deformation in the streamlines than a more homogeneous particle of spherical shape. A Chaetoceros colony could therefore be more easily detected than other cells and hence included in the prey spectrum of an ambush feeding copepod. Within the bloom, R. gigas preferentially selected ciliates but cleared diatoms according to their contribution to abundance. Probably, the copepod switched between the feeding modes at this stage, still taking advantage of the highly nutritious ciliates via ambush feeding but maximizing ingestion of diatoms through rather passive accumulation. Similar behavior is described for Acartia tonsa feeding in a mixture of diatoms and ciliates at varying concentrations (Kiørboe et al., 1996). Atkinson and Shreeve (1995) termed R. gigas an “indiscriminate feeder” at the bloom stations of their study in the Bellingshausen Sea. The flexible feeding behavior of R. gigas is possibly one reason why this copepod is so widespread throughout the Southern Ocean, a fact that is known since the Discovery Cruises (Ommanney, 1936).

C. simillimus is restricted to the comparably productive waters of the PFZ (Froneman et al., 2000) and appeared to be a specialized harvester of large, carbon rich diatoms during EisenEx. The average clearance rate on diatoms is consistently higher compared to ciliates and hdnios. Furthermore, clearance on diatoms rises rapidly with increasing size of the cell, similar to results found by Atkinson (1994, 1995, 1996) and then is maintained stable for a whole range of biomass important species (see Fig. 6A). Feeding on the biomass dominant particle type is referred to as “peak tracking” (Richman et al., 1977) and is not necessarily tied to large sized cells, as demonstrated in studies by Schnack (1983, 1985) and Perissinotto (1992). The latter author reported preferential feeding of C. simillimus on the particle fraction <20 μm, which represented the biomass peak during that investigation. No sharp line can be drawn whether size or biomass determine the efficiency with which a diatom is preyed upon. This seems reasonable considering that a grazer has to survive in an environment with highly variable food supply in terms of quality and quantity as was impressively demonstrated by the floristic shift following the fertilization. This variability is also observed in situ. In November 1992, Polarstern expedition ANT X/6 encountered three distinct blooms in the PFZ, each dominated by a different diatom species (Smetacek et al., 1997).

Both C. simillimus and R. gigas apparently “avoided” hdnios with progression of the bloom. Such “prey switching” is a common strategy among copepods to maximize energy gain (e.g. Kiørboe et al., 1996; Meyer-Harms et al., 1999) and is often related to a change in feeding behavior from ambush feeding to suspension feeding. Furthermore, suspension feeding copepods increase the rate of flapping of the cephalothoracic appendages with increasing cell concentrations (Price and Paffenhöfer, 1986). Especially dinoflagellates are able to escape the feeding current that is created during suspension feeding. Some ciliate species, however, can be entrained in the feeding current (Kiørboe et al., 1996). In line with above appraisal of the grazers’ feeding behavior, it is proposed that both copepods reduced their grazing pressure on hdnios due to modifications in their feeding mode: for R. gigas from ambush feeding to suspension feeding, and for C. simillimus due to increased activity in the suspension feeding mode with development of the bloom.

4.4. Grazing impact on diatom populations

Overall diatom mortality due to the grazing activity of C. simillimus alone reaches 15.2% of the standing stock
per day (% s.s. day$^{-1}$) and decreases to 5.6% with the development of the bloom. Studying the grazing impact of this copepod in a bloom and non-bloom situation near the Prince Edward Archipelago, Perissinotto (1992) found C. simillimus ingesting 13% to 16% s.s. day$^{-1}$ in the non-bloom situation and 1.2% to 2.1% s.s. day$^{-1}$ during the bloom. As previously mentioned, abundance of C. simillimus during EisenEx was comparable to the study of Perissinotto (1992) and the same holds true for the grazing impact estimates. C. simillimus frequently dominates the copepod communities of the PFZ and within these often shows highest daily ingestion rates among grazers (Perissinotto, 1992; Atkinson et al., 1996; Pakhomov et al., 1997; Froneman et al., 2000). Overall grazing impact of copepods in these studies reached 25% of phytoplankton standing stock removed per day with the major fraction being attributable to C. simillimus. The combined grazing impact of R. gigas and the copepods <2 mm estimated from the incubations did not exceed 1.5% s.s. day$^{-1}$ due to low diatom ingestion rates or low grazer standing stock in the mixed layer.

Large diatoms were cleared with highest rates by all copepods. Assuming that changes in handling effort are minor, this strategy maximizes carbon ration per feeding operation (sensu Frost, 1972). The fact that few diatom species, C. pennatum and R. chunii for example (see Fig. 3B), contribute a major fraction of ingested carbon to the copepods reflects this principle very well. Consequently, the mortality that these species suffer from copepod grazing is proportionally higher than for species cleared with lower efficiency. Average mortality for Corethron sp. for example is almost an order of magnitude higher than the grazing loss inflicted on F. kerguelensis. Assmy et al. (submitted for publication) determined six response types for the accumulation of diatom species following the iron fertilization. Type I is characterized by fast growing, weakly silicified diatoms following the iron fertilization. Type I is determined six response types for the accumulation of diatom species following the iron fertilization. Type I is determined six response types for the accumulation of diatom species following the iron fertilization. Type I is determined six response types for the accumulation of diatom species following the iron fertilization. Type I is determined six response types for the accumulation of diatom species following the iron fertilization. Type I is determined six response types for the accumulation of diatom species following the iron fertilization. Type I is determined six response types for the accumulation of diatom species following the iron fertilization. Type I is characterized by fast growing, weakly silicified diatoms like P. lineola and C. debilis, which dominated the bloom and accumulated exponentially inside the patch. Assmy et al. (submitted for publication) derived a gross growth rate of 0.41 day$^{-1}$ for P. lineola. Gross growth of C. debilis could potentially be even higher, up to 1.8 day$^{-1}$ measured during the iron fertilization experiment SEEDS (Tsuda et al., 2005). The initially high grazing mortality for P. lineola and Chaetoceros spp. is substantially reduced in the fully developed bloom. Both bloom formers, P. lineola and C. debilis, clearly outgrew their predators. For several large diatoms, Assmy et al. (submitted for publication) report a linear accumulation pattern which indicates a growth limitation via bottom-up or top-down factors. Representatives of this group are C. pennatum and R. chunii and G. cylindrus. Daily grazing loss of the population of Corethron spp, R. chunii and Guinardia spp. is still high on day 21. A strikingly low ratio of empty/broken diatom frustules observed for C. pennatum (Assmy et al., submitted for publication) confirms intense mesozooplankton grazing on this species. The author’s give a gross growth rate of 0.3 day$^{-1}$ for C. pennatum which is in the same range as grazing mortality calculated in this study. The population dynamics of C. pennatum are apparently a direct function of copepod grazing activity in this area.

The initial phytoplankton community was numerically dominated by F. kerguelensis and T. nitzschioides that both suffer lowest grazing mortality from large grazers. Calculation of selectivity indices for various diatoms shows unselective feeding of C. simillimus on F. kerguelensis and avoidance of T. nitzschioides (data not shown) which we believe reflects the influence of their smaller size on grazing efficiency (sensu Boyd, 1976). Alternatively, the high degree of silicification of F. kerguelensis is argued to protect it from grazing (Verity and Smetacek, 1996; Hamm et al., 2003). The continued presence of a part of the copepod community in the surface layer during austral winter—preferably feeding on larger diatoms or microzooplankton—may partly explain the dominance of F. kerguelensis and other small species in the initial population before the iron infusion, and in spring populations of the Southern Ocean in general (Hart, 1934).

Henjes et al. (submitted for publication) explain the stagnant development of microzooplankton populations during EisenEx with an increasing grazing pressure by copepods. However, the mortality of microzooplankton attributable to the here investigated grazers appeared to be limited and in addition, the larger copepods diverted their grazing pressure to diatoms with progression of the bloom. Although grazing on microzooplankton by the copepods <2 mm might be underestimated by 15% (Klaas et al., in preparation) the bulk of the grazing mortality is most probably inflicted by small copepod species like Oithona sp. that feed preferentially on motile prey (Atkinson, 1996) and of which populations indeed increased by a factor of three during the study (Henjes et al., submitted for publication).

5. Conclusion

Froneman et al. (2000) proposed that C. simillimus should be considered a key organism of the PFZ and the results of the present study support this conclusion. This grazer dominated the mesozooplankton community, showed highest ingestion rates, consistently
preyed on diatoms most effectively and likely structured the phytoplankton composition of communities thriving in this area of enhanced primary production. Furthermore, importance of heterotrophic prey for copepod grazers in the Southern Ocean has been confirmed and thus future investigations on feeding should include methods to estimate both autotrophic and heterotrophic carbon sources. Differences in foraging behavior in response to the changing supply of microplankton standing stock as observed for *R. gigas* may be critical for grazers to adapt to a wider range of productivity regimes.

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