Marine Biology (2001) 139: 301–311
DOI 10.1007/s002270100590

A. Atkinson · M.J. Whitehouse

Ammonium regeneration by Antarctic mesozooplankton: an allometric approach

Received: 28 November 2000 / Accepted: 16 March 2001 / Published online: 5 July 2001

Abstract Given the importance of copepods in the Southern Ocean food web, there are few assessments of their N budgets or their role in regenerating N. In this study we measured elemental composition and ammonium-excretion rates of copepods and small euphausiids, and estimated the role of metazoans in recycling ammonium in the South Georgia region. Measurements were made during summer on animals ranging over about two orders of magnitude in body mass. A phytoplankton bloom extended throughout the study area, and high C and dry masses of late-stage copepodites suggested good recent feeding conditions. Excretion rates declined roughly exponentially during the ~1 day incubations in filtered sea water. The patterns observed suggested that the onset of starvation rather than the stress of capture caused this. Allometric relationships between body mass m and excretion rate R were derived using the equation $R = am^b$. Large compilations of literature data produce a value of b (the body-mass scaling coefficient) of 0.7–0.8. However, in this study, b ranged from 0.57 (for C as the unit of body mass) to 0.71 for N as the unit of body mass. Such low values are also common to previous studies of feeding and excretion among Antarctic copepods. We suggest that this reflects peculiarities of polar environments; namely, lipid storage and diapause in the largest copepods. Previous studies have suggested that ammonium is a preferred N source for algae at South Georgia. Based on the monitoring of a region to the north-east of South Georgia and on zooplankton abundance and excretion rates from this study, we estimate that within the upper mixed layer the copepods and small euphausiids excrete at least one third of the ammonium potentially required by phytoplankton. Krill excretion in this area was measured in a previous study, and it appears that mesozooplankton and krill are together significant regenerators of N in parts of the South Georgia pelagic system.

Communicated by J.P. Thorpe, Port Erin

A. Atkinson (✉) · M.J. Whitehouse
British Antarctic Survey,
Natural Environment Research Council,
High Cross, Madingley Road,
Cambridge CB3 OET, UK

E-mail: aat@pcmail.nerc-bas.ac.uk

Introduction

The controls exerted by zooplankton on oceanic primary production rates can be both direct and indirect (Banse 1995). Feeding can alter the abundance and species composition of the algae, whereas excretion of nutrients can promote their growth. Protozoans are often more prominent than metazoans in both of these regards, but even when this is the case, metazoans can have indirect effects through selective grazing (e.g. Gifford 1993) and N regeneration (Glibert et al. 1992; Miller and Glibert 1998).

In the Southern Ocean, metazooplankton generally have a low grazing impact on phytoplankton (e.g. Schnack et al. 1985; Atkinson 1996; Dubischar and Bathmann 1997; Razouls et al. 1998). Likewise, their ammonium excretion contributes little to the N budget (Biggs 1982; Huntley and Nordhausen 1995; Alcaraz et al. 1998; Hernández-León et al. 1999). However, swarms of krill or salps can be an exception, having locally high impact through grazing and nutrient regeneration (e.g. Antezana and Ray 1984; Johnson et al. 1984).

In certain parts of the Southern Ocean however, this generally low direct impact of zooplankton on phytoplankton does not seem to hold. One such area is South Georgia, where primary production rates are locally high and may be modulated by regenerated ammonium (Owens et al. 1991; Priddle et al. 1997; Whitehouse et al. 1999). Outside of bloom periods the biomass of metazooplankton can be high enough to exert a grazing control on phytoplankton (Ward et al. 1995; Pakhomov

et al. 1997; Atkinson et al., in press). Krill (*Euphausia superba*) may also be large-scale regenerators of ammonium at South Georgia (Atkinson and Whitehouse 2000). Because copepod biomass is often similar to that of krill there, and copepod metabolic rates may be higher, assessment of their excretion is required to examine how algal growth may be enhanced by mesozooplankton excretion.

In this study we measured ammonium-excretion rates of copepods and small euphausiids ranging over about two orders of magnitude in body mass. Very few data exist on the excretion rates of Southern Ocean copepods. Our first objective was to help remedy this for a group which may have a higher overall biomass and production in the Southern Ocean than either krill or salps (Conover and Huntley 1991; Voronina 1998). The second objective was to estimate the importance of metazoans in ammonium regeneration at South Georgia. For this we used an allometric approach and investigated how their excretion and feeding rates were related to structural (N) mass, C mass and total dry mass.

Materials and methods

Experiments

Experimental animals were caught from 20 December 1998 to 7 January 1999 with vertical nets in the top 50 m at 16 sites to the north of South Georgia (Fig. 1). To minimise abrasion of the catch, a motion-compensating spring on our 61-cm-diameter, 200-μm-mesh Bongo net was used at eight of the sites. For the remaining hauls a 75-cm-diameter, 200-μm-mesh net was hauled by hand to compensate for the pitching of the ship. The catches from the solid 5-l cod end were diluted in surface sea water and sorted immediately. Undamaged, actively swimming individuals were transferred to 1.2-l acclimation bottles at ambient mixed-layer temperature and the remainder were preserved in a 5% solution of formaldehyde in filtered sea water. Experiments were performed at each of the 16 sites (Table 1).

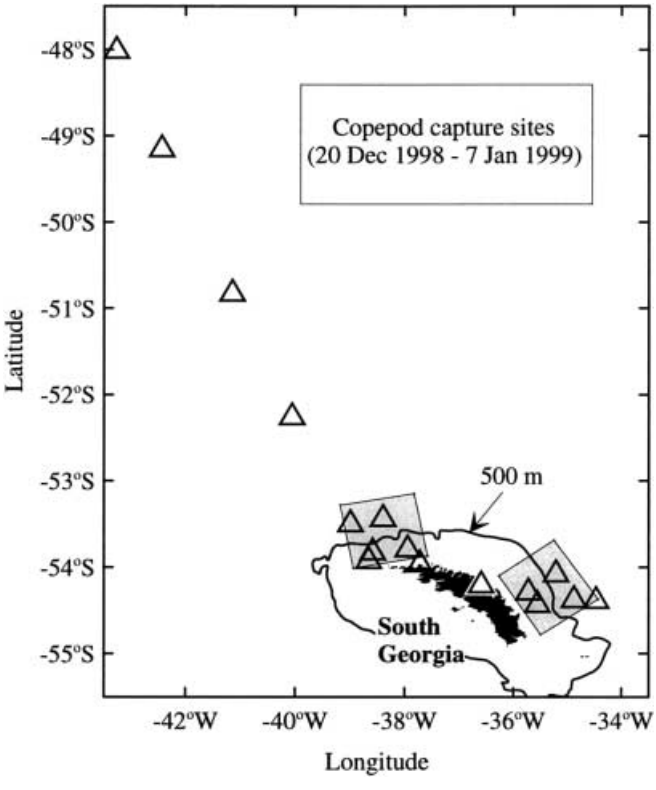

Fig. 1 The study area and the 16 sampling sites in relation to the western and eastern monitoring boxes

Experiments were started after a short (mean 4 h) acclimation period which allowed the animals to recover from the stress of capture and sorting. A bloom was encountered at most sites, and large diatoms were unavoidably picked with the animals during sorting. This meant that the animals were able to feed in high food concentrations during this brief acclimation. Incubations were in filtered sea water prepared freshly before each experiment. To remove the copepods from their acclimation food and set up the experiment, 90% of the bottle contents was siphoned off before topping up with the mixed incubation water, and this process was repeated 3–4 times. This method was successfuly completed for the species for which data are presented here. We also attempted incubations with very small copepods, but it was not possible to separate them adequately from the large diatoms and we abandoned these experiments.

Incubations were in either 1200-ml or 600-ml glass bottles, depending on animal size. Numbers of individuals per bottle ranged from 2 for *Rhincalanus gigas* CVI♀ to 136 for *Calanus simillimus* CIII. Maximum stocking densities (i.e at the end of the experiment after subsamples had been taken) were < 40 mg dry mass (DM) l^{-1} (median of the 73 incubations was 8.7 mg DM l^{-1}). Most incubations were with copepods, but we conducted five with small *Thysanoessa* spp. which made a large contribution to some of the 0- to 50-m catches. Two control bottles of filtered sea water with no zooplankton were used in each experiment. Incubations were in the dark at the ambient temperature of the upper mixed layer at the sampling location: 7°C for the first two experiments, 5°C for the third, and 3°C thereafter. The only mixing was before subsampling.

Mean duration of the experiments was 28 h, during which subsamples were usually taken at 3–5 time points, in addition to an initial one. Subsampling from experimental and from control bottles involved first gently mixing their contents and then obtaining two 20-ml subsamples using a syringe fitted with an integral filter unit containing a mixed-ester membrane (Whatman WME, pore size 0.45 μm). A 100-μm gauze over the intake precluded animals. Subsamples were normally analysed immediately but occasionally were stored in the dark in the experimental cold room for a few hours before analysis. Tests showed that storage for such periods had a negligible effect on ammonium concentrations. Samples were analysed colorimetrically for dissolved nutrients using a Technicon segmented-flow analyser (Whitehouse 1997). Previous calibration tests on the ammonium assay gave a detection limit of 0.01 mmol m^{-3} and precision within 1% (Whitehouse and Woodley 1987).

At the end of each experiment the volume of water remaining in each bottle was measured, the copepods were checked for mortality (which was negligible during these incubations) and the animals were frozen at –80°C for analysis in the United Kingdom.

Dry mass and elemental analysis

The experimental animals were thawed, counted, identified and dried in batches in tin-foil cups at 50°C for 24 h. After cooling in a dessicator, they were weighed immediately to the nearest microgram on a microbalance. Elemental analysis of these samples was done with a Fisher Scientific EA 1108 elemental analyser using acetanilide as a standard. Comparisons of DM, C mass and N mass of copepods from the same cruise obtained with the same analyser (R.S. Shreeve, British Antarctic Survey, personal communication) showed good correspondence between C masses and DMs, but not N masses. The N masses of our copepods caught from the top 50 m

Table 1 Details of the experiments at the 16 sites. The numbers below the species and stages refer to the number of bottles incubated in an experiment. Local times (GMT-3 h) of night-time net hauls are in italics. Darkness lasted from ~2100 hours to 0400 hours local time

Site	Time of sampling	*Metridia lucens* CVI♀	*Metridia gerlachei* CVI♀	*Neocalanus tonsus* CV	*Calanus simillimus* CIII	CIV	CV	CVI♀	*Calanus propinquus* CV	*Calanoides acutus* CV	*Rhincalanus gigas* CV	CVI♀	*Thysanoessa* spp. Larvae plus postlarvae
1	1457	–	–	1	–	1	2	–	–	–	–	–	–
2	1508	–	–	–	1	–	–	1	–	–	1	–	–
3	1917	–	–	–	1	–	1	–	–	–	2	–	–
4	*2238*	–	–	–	–	–	–	–	–	1	–	–	–
5	1208	–	–	–	–	–	–	–	–	1	–	–	–
6	1815	–	–	–	–	–	–	–	–	1	1	2	–
7	1843	–	–	–	–	–	–	1	–	1	–	1	–
8	*0037*	1	–	–	–	–	–	1	–	2	–	1	–
9	*0130*	1	–	–	–	–	1	–	–	2	–	2	–
10	*0056*	–	1	–	–	–	1	–	–	2	–	2	2
11	1807	–	1	–	–	–	1	–	1	2	–	1	–
12	*0100*	1	–	–	–	–	1	–	–	2	–	1	2
13	*0145*	1	–	–	–	–	1	–	–	2	–	2	–
14	*0052*	1	–	–	–	1	1	–	1	2	–	2	1
15	*0203*	–	1	–	–	–	–	1	–	2	–	2	–
16	1910	–	–	–	–	–	–	–	–	2	–	–	–

and stored frozen were consistently less than those caught from the top 200 m and stored dry. Because we could not be sure whether this reflected differences in the copepod populations sampled, or in the experimental method, we have erred on the side of caution and not presented the N mass values of the experimental animals. However, we are confident that our values are at least internally consistent, so they are used in allometric relationships between excretion rate and body mass (see Results and discussion, Excretion rate in relation to body mass).

Enumeration of zooplankton catches

The formaldehyde-preserved catches of the 0- to 50-m net hauls were divided into aliquots using a Folsom splitter and the copepods and small euphausiids were counted under a binocular microscope. The biomass-dominant copepods (see Table 2) were all enumerated, but the smaller copepods (mainly *Oithona similis*, *Ctenocalanus* spp., early copepodite stages) were counted and measured in order to estimate their DM from length-mass regressions (See Results and discusssion, Ammonium regeneration by the zooplankton community).

Calculation of excretion rates

The rate of ammonium excretion in the interval between successive subsampling times was calculated from the equation

$$E = [(K_{t2} - c_{t2}) - (K_{t1} - c_{t1})]V/m(t_2 - t_1)$$

where E is the rate of ammonium excretion (μmol mg^{-1} h^{-1}), t_1 is time 1, t_2 is time 2, K_{t1} and K_{t2} are, respectively, the concentrations of ammonium in zooplankton bottle, c_{t1} and c_{t2} are, respectively, the mean concentrations in the two controls, V is the volume of water in the bottles before subsampling at time t_2 and m is the total mass of zooplankters. Because volumes were depleted by repeated subsampling, the volume of water at each time interval was calculated and used in the above equation.

Results and discussion

Body mass

Mean values of DM, C mass and C percentage of the prominent zooplankters across the 16 sites are given in Table 2. Both *Calanoides acutus* CV and *Rhincalanus gigas* CVI♀ had large lipid depots which are reflected in their high DMs. For the copepods generally, C percentages in CV tended to be higher than those in earlier copepodite stages, reflecting the buildup of C-rich lipids with age (Hagen 1988; Donnelly et al. 1994; Schnack-Schiel and Hagen 1995; Shreeve and Ward 1998). The high DMs and lipid stores of *C. acutus* and *R. gigas* late-stage copepodites probably reflect the good feeding conditions at South Georgia. A widespread bloom (~5 μg chl *a* l^{-1}) occurred at all of our sampling stations except the most northerly. It appears that blooms can occur throughout a long growing season at South Georgia (Atkinson et al., in press), supporting high rates of feeding (Atkinson et al. 1996a), growth (Shreeve and Ward 1998), and the deposition of large lipid reserves (Ward et al. 1996).

In contrast to *Calanoides acutus* CV and *Rhincalanus gigas* CVI♀, DMs of the colder-water species *Calanus propinquus* CV and *Metridia gerlachei* CVI♀ were low, averaging 600 μg and 183 μg, respectively. These compare with respective values of ~200–1550 μg and ~180–380 μg in the eastern Weddell Sea (Hagen and Schnack-Schiel 1996). In their seasonal study, Hagen and Schnack-Schiel (1996) drew attention to the extreme (over five-fold) variation in DMs of late-stage copepodites of the large copepod species, according to their stage in the life cycle. Likewise, Shreeve and Ward (1998) noted large differences in body mass according to stage in the moult cycle. Possibly the *C. propinquus* CV and *M. gerlachei* CVI♀ were recent moults into these respective stages.

Excretion in relation to experimental duration

Excretion rates for all species and stages declined during the experiments. To examine this for the most commonly incubated species, *Rhincalanus gigas, Calanoides*

Table 2 Mean dry masses and C content of mesozooplankton from the 0–50 m layer at 16 sites north of South Georgia during December 1998–January 1999. Not all species in this list were incubated: see Table 1

Species	Stage	Dry-mass measurement		Carbon content		
		μg ind^{-1}	No. inds measured	C μg ind^{-1}	C as % of dry mass	(No. ind measured)
Oithona frigida	CVI♀	7.7	(25)	–	–	–
Drepanopus forcipatus	CVI♀	55	(38)	–	–	–
Metridia lucens	CVI♀	87	(107)	32	36	(58)
Metridia gerlachei	CVI♀	183	(37)	84	46	(22)
Neocalanus tonsus	CV	257	(16)	121	47	(10)
Calanus simillimus	CIII	20	(38)	8.5	43	(35)
	CIV	36	(52)	14	39	(59)
	CV	134	(52)	62	46	(22)
	CVI♀	210	(73)	78	37	(37)
	CVI♂	166	(5)	–	–	–
Calanus propinquus	CIV	151	(21)	66	44	(11)
	CV	600	(14)	306	51	(2)
Calanoides acutus	CV	596	(190)	331	56	(33)
	CVI	625	(7)	–	–	–
Rhincalanus gigas	CIV	243	(18)	87	36	(7)
	CV	552	(41)	221	40	(11)
	CVI♀	2,106	(90)	948	45	(19)
Thysanoessa spp.	Mixed larvae/postlarvae	486	(38)	156	32	(15)

acutus, Calanus simillimus, Metridia gerlachei and *Metridia lucens*, we have normalised the excretion rate obtained for each time point in each incubation bottle to a percentage of the maximum rate observed in that bottle. These percentages are plotted against time in Fig. 2. An exponential model provides a description of the time course, and this is analagous to the models most frequently used to describe the decrease in gut content during a gut-evacuation experiment (e.g. Kiørboe and Tiselius 1987). The rate of decline in excretion rate varied between species, with that for *Calanoides acutus* being over twice that for *Metridia* spp. The implication is that the observed excretion rate depends heavily on the duration chosen for the experiment.

The apparent simplicity of performing excretion- or respiration rate-experiments, simply incubating animals in filtered sea water, belies the difficulty in applying the results to the real world (Ikeda et al. 2000). For example, there is uncertainty over why excretion rates decline in such experiments. Most workers (e.g. Skjoldal et al. 1984; Båmstedt 1985) attribute it to the onset of starvation, although others suggest that high initial rates reflect the stress of capture or handling (e.g. Gardner and Paffenhöfer 1982; Huntley and Nordhausen 1995). Clearly this distinction is crucial because excretion rates can decrease substantially (e.g. Skjoldal et al. 1984; Båmstedt and Tande 1985) and radically different rates would be obtained from long-term or short-term incubations. For example, in our experiments (Fig. 2), averaging the values over 24 h would yield excretion rates between one-third and two-thirds (depending on species) of those obtained at the start of the experiment. There has been no clear protocol for how long to incubate to arrive at in situ estimates of excretion rate (Ikeda et al. 2000) and in the five previous studies of Southern Ocean copepods, incubation times ranged from 4 h to 24 h.

Although we cannot rule out the effects of experimental stress on the results, our method was designed to minimise it: a short (~4 h) acclimation allowed copepods the chance to recover from the stress of capture and sorting and to resume feeding. Also the sensitivity of the ammonium analysis allowed larger incubation volumes with lower stocking densities than in other Southern Ocean studies. We believe, in common with most other studies, that the decline in excretion rate was induced by starvation. However, the evidence is only circumstantial. First, the decline was gradual over hours rather than abrupt over minutes, and showed the same slowly decreasing rate found for lab-acclimated marine species switched from a feeding regime to starvation (e.g. Chapelle et al. 1994; Boyce and Clarke 1997). Second, the decline was slowest in *Metridia lucens* and *M. gerlachei*, species with a slower gut-passage time than copepods of similar size (Morales et al. 1993; Lopez and Huntley 1995; Atkinson et al. 1996b).

To allow comparison with previous Antarctic studies, in the following sections the rates have been averaged over the first two subsampling time points after the initial one (i.e. over the first ~15 h of incubation). However, these may be underestimates of the in situ or initial rate, which may be better calculated from the interception of the regression lines in Fig. 2 with the vertical axis (i.e. at time zero). This would give a result ranging from 1.3 times (for *Metridia lucens* and *M. gerlachei*) to 2.0 times (for *Calanoides acutus*) those of our values averaged over the first 15 h.

Excretion in relation to time of day

Half of the experiments were with night-time catches, which allowed a determination of whether excretion

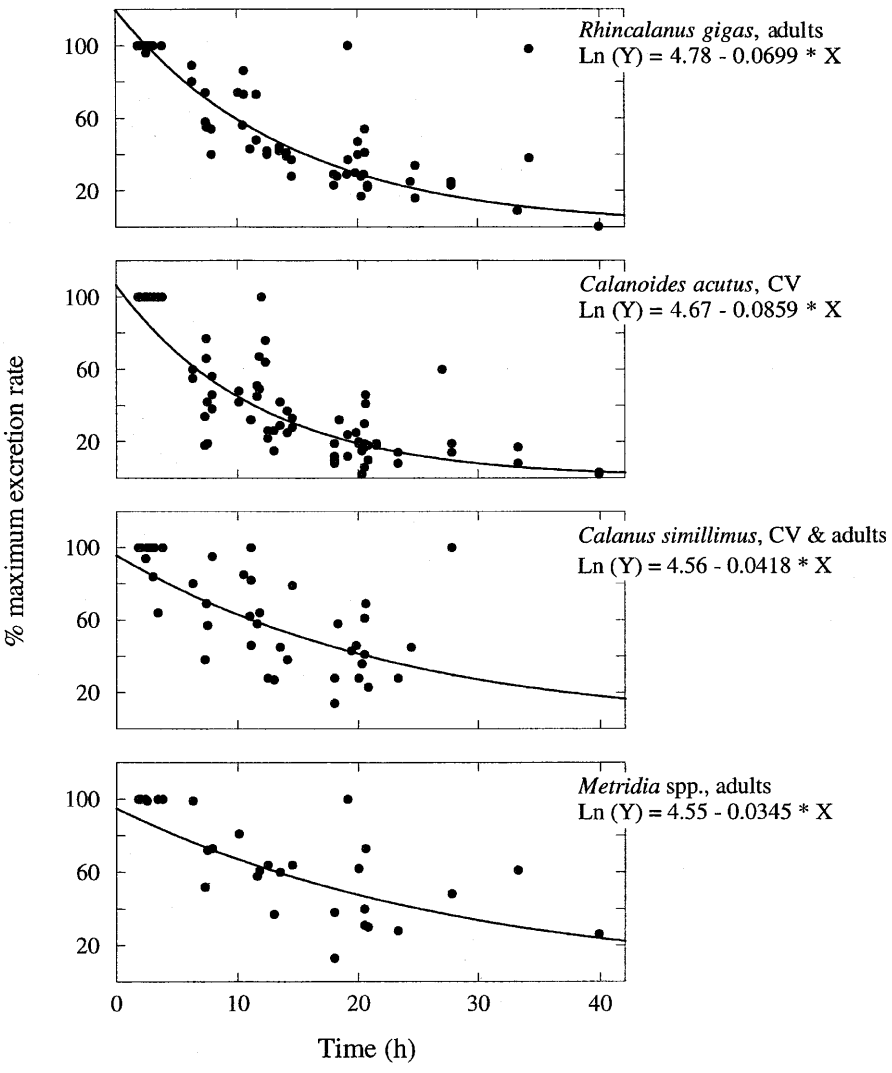

Fig. 2 *Rhincalanus gigas* CVI♀, *Calanoides acutus* CV, *Calanus simillimus* CV and CVI, *Metridia lucens* CVI♀ and *M. gerlachei* CVI♀. Decline in ammonium-excretion rate in relation to experiment duration. Each point represents an excretion rate measured for an experimental bottle at the mid-point of the subsampling time interval. Each excretion rate is plotted as a percentage of the maximum value observed for the bottle during that experiment. Equations relate percentage of maximum excretion rate (Y) to duration of experiment in hours (X)

rates of animals in the upper mixed layer were higher at night (e.g. Harris and Malej 1986) or during the day (e.g. Checkley et al. 1992) in association with a diel feeding cycle. No clear pattern was evident (Table 3). Simple day–night differences may be obscured by a lag between the start of the diel feeding cycle and the increase in excretion. However the daytime hauls tended to be in the afternoon or evening (i.e. generally after >12 h daylight; see Table 1) and the night-time hauls were generally after midnight (i.e. after >3 h of darkness). Therefore, if a pronounced diel rhythm in excretion had either accompanied nocturnal feeding or lagged by less than $\sim$4 h behind it, we should have observed it. Previous observations during a dense summer bloom (Atkinson et al. 1996a) suggested that feeding occurred during day and night, and there was no synchronised diel vertical migration. This might explain the lack of clear day–night differences in the excretion rate we observed.

Although no diel differences in excretion were clear, mean mass-specific excretion rates per individual varied over three-fold between sites (Table 3). Variation in the measurement and between replicate bottles (i.e. within-site variability) was much less than that between sites. Of the 16 replicate incubations with *Calanoides acutus* and *Rhincalanus gigas*, over two-thirds produced rates within 25% of each other. Instead, this variability probably reflects the wide spread of sample coverage, from distinct oceanographic regimes (Brandon et al. 1999; Whitehouse et al. 2000), although no clear regional trends were apparent.

Comparison with previous studies

The variability in size, body mass and lipid storage between copepods of the same species and moult stage complicates comparisons of rate processes between studies (Table 4). Although comparisons based on structural (N) mass may be the most realistic (Vidal and Whitledge 1982; Paffenhöfer and Gardner 1984), we are forced to use DM as the only common currency between studies. Our results have been made as comparable as possible using a Q_{10} of 1.67 (a DM-based value from

Table 3 Mean excretion rates in daytime, night-time and overall values. (*n*) number of experimental sites

Species	Ammonium-excretion rate (μmol mg^{-1} DM h^{-1})		
	Day (*n*)	Night (*n*)	Overall (range)
Rhincalanus gigas CVI♀	6.1 (4)	4.4 (6)	5.1 (3.2–8.9)
Rhincalanus gigas CV	13 (2)	4.8 (1)	10 (4.8–13)
Calanus propinquus CV	13 (1)	5.8 (1)	9.6 (5.8–13)
Calanoides acutus CV	5.9 (4)	5.3 (8)	5.5 (3.2–8.0)
Thysanoessa spp. larvae + postlarvae	–	5.0 (3)	5.0 (2.2–8.0)
Neocalanus tonsus CV	16 (1)	–	16
Calanus simillimus CV + CVI♀	12 (5)	11 (7)	11.1 (6.8–14)
Metridia lucens + *M. gerlachei* CVI♀	10 (1)	16 (7)	15 (9.3–26)
Calanus simillimus CIII + CIV	19 (3)	–	19 (14–23)

Table 4 Excretion rates, Q_{10} adjusted to –0.5°C (see text) to allow comparison with previous data. All studies were conducted soon after capture of animals except for that by Ikeda and Hing Fay (1981) where they were kept in the lab for 2–6 weeks before use. Species are listed in order of decreasing DM. Results of Biggs (1982) have been converted to DM from the original wet-mass data using a wet mass:DM ratio of 4:1

Species	Mean ammonium-excretion rate μmol mg^{-1} DM h^{-1}	Austral time of year	Duration of incubation	Reference
Rhincalanus gigas CV, CVI♀	2.1	Summer/autumn	24 h	Pasternak et al. (1994)
	4.2	Summer	15 h	This study
	6.4	Summer	4 h	Biggs (1982)
Calanus propinquus CV	3.1	Autumn	24 h	Pasternak et al. (1994)
	3.4	Summer	24 h	Ikeda and Hing-Fay (1981)
	6.3	Summer	4 h	Biggs (1982)
	7.4	Summer	24 h	Ikeda and Mitchell (1982)
	8.0	Summer	15 h	This study
Calanoides acutus	0.34	Summer	24 h	Ikeda and Hing Fay (1981)
CV, CVI♀	1.4	Winter	12 h	Huntley and Nordhausen (1995)
	1.7	Summer/autumn	24 h	Pasternak et al. (1994)
	4.6	Summer	15 h	This study
Metridia gerlachei	3.0	Winter	12 h	Huntley and Nordhausen (1995)
CVI♀	8.4	Summer	24 h	Ikeda and Mitchell (1982)
	8.6	Summer	15 h	This study

Ikeda 1985) to adjust them to –0.5°C, a similar temperature to that of previous studies. We have also used excretion rates calculated over the first ~15 h of our experiments to make them broadly comparable with those of previous studies. On this basis, excretion rates in this study are generally at the top end of the range observed previously. This might reflect the high food concentrations throughout the study area, but seasonal diapause or methodological differences (e.g. stocking density, capture stress, experimental duration) probably contribute to the variability.

Excretion in relation to body mass

Figure 3 shows the relationship between excretion rate and body mass, expressed either as DM, C mass or N mass. This relationship is described by the general allometric equation relating a metabolic rate R, to body mass, m:

$$R = am^b$$

where a and b are constants; b is known as the body-mass scaling coefficient. Since b is a power function and m can vary by five or more orders of magnitude in a zooplankton community, small changes in b can reflect large differences in the relative metabolic rates across the community. Thus a small decrease in the value of b would mean a large increase in the importance of small-zooplankton metabolism relative to that of larger species. The log-log relationships in Fig. 3 are all significant, but their slopes (i.e. b, the body-mass scaling coefficients) are different: 0.57, 0.64 and 0.71 for body mass expressed as C mass, DM and N mass, respectively.

Literature compilations of zooplankton excretion and feeding rates have arrived at fairly similar values for b of ~0.75 (Peters and Downing 1984; Ikeda 1985; Wen and Peters 1994). This has led to the application of such values for examining community rate processes (e.g. Moloney and Field 1989, 1991), since temperature and body mass explain a large amount of the variance in such data compilations. However, studies within actual communities have found a much wider variation in b;

for example, from 0.52 (Ikeda et al. 1982) to 1.2 (Paffenhöfer and Gardner 1984). In Table 5 allometric regressions for zooplankton excretion and feeding are compiled for Antarctic zooplankton assemblages. Although some of the values of b are within the theoretical range, many, including the results of this study, are lower. We believe that some of the basic features of high-latitude zooplankton, namely lipid storage and diapause, are the reasons for this.

None of the studies in Table 5 encompasses the enormous range of size and life style of Antarctic metazooplankton, from tiny nauplii to krill and salps. The regressions thus reflect the particular size spectrum incubated. For the relationships determined for copepods, the values of b are consistently low. The largest animals in this range are the late-stage copepodites of large copepods, for which $>50\%$ of their DM may be C-rich, non-metabolically active, lipid (Hagen and Schnack-Schiel 1996). Conversely the early copepodites and smaller species store less lipid, as reflected by the increase in C:N ratios of copepods with maturity stage (Shreeve and Ward 1998). Ikeda (1985) highlighted the fact that choice of body-mass unit dictates the allometric relationships found (as does the regression model used; see Ikeda et al. 2000). Vidal and Whitledge (1982) suggested that for species storing non-metabolically active tissue, structural (N) mass is a better predictor of metabolic rate than DM or C mass. Our results (Fig. 3) support this contention because, when C mass is replaced by N mass as a predictor of copepod excretion rate, the value of b rises from 0.57 to 0.71, closer to the most commonly derived empirical values (Moloney and Field 1989; Wen and Peters 1994).

Copepod diapause is another factor which could result in widely varying values of b. For example, in the winter study by Huntley and Nordhausen (1995), the low value of b (see our Table 5) probably reflected this. These authors suggested that the larger species incubated, *Calanoides acutus,* was in diapause while the smaller species, *Metridia gerlachei*, was active.

Therefore, in some circumstances, feeding or excretion of high-latitude zooplankton assemblages may scale with body mass very differently to that predicted from large compilations of literature data. This may reflect the species and size composition of the assemblage, the time of year or the unit chosen to express body mass.Therefore, allometric models of zooplankton activity in high-latitude environments must be chosen with care.

Ammonium regeneration by the zooplankton community

South Georgia typifies an environment where it is difficult to measure rate processes across the full size spectrum of metazoans. In particular, small copepods are often numerous, diverse, and hard to separate physically from the large diatoms which characterise Southern Ocean blooms. Since the small fraction can be major contributors to community rate processes in the Southern Ocean (Schnack et al. 1985; Fransz and Gonzalez 1995; Atkinson 1996) and elsewhere (Paffenhöfer 1971; Morales et al. 1993), they need to be assessed. Here we use an allometric approach to this problem. We selected for incubation the prominent mesozooplankters which covered as wide a range in body mass as possible. This

Table 5 Literature values of Southern Ocean studies expressed by the relationship $R = am^b$. Listed are values of the body-mass scaling coefficient, b, for excretion or feeding rate, R using body mass, m expressed as dry mass (DM), C mass and N mass. Dashes denote that these mass units were not measured

Rate process, R	Body-mass scaling coefficient, b			Zooplankton size incubated	Mean size range (μg DM)	Reference
	Mass, m, as DM	Mass, m, as C mass	Mass, m, as N mass			
Excretion rate	0.76	0.78	0.8	Large copepods to euphausiids	265–354,000	Ikeda and Mitchell (1982)
	0.73	0.72	0.71	Macroplankton up to *Euphausia superba*	280–67,930	Huntley and Nordhausen (1995)
	0.24	0.22	0.24	Small to large copepods (two species only)	30–569	Huntley and Nordhausen (1995)
	0.64	0.57	0.71	Small to large copepods	28–2,106	This study
Feeding rate	0.61, 0.67[a]	–	–	Small to large copepods	2.2–1,507	Atkinson and Shreeve (1995)
	0.42–0.59[b]	–	–	Small to large copepods	1.5–543	Atkinson (1996)
	0.49–0.65[b]	–	–	Small to large copepods	36–2,960	Atkinson et al. (1996a)

[a] Feeding rate expressed as mean and maximum clearance rate, respectively
[b] Range, reflecting different (autotrophic and mainly heterotrophic) food sources

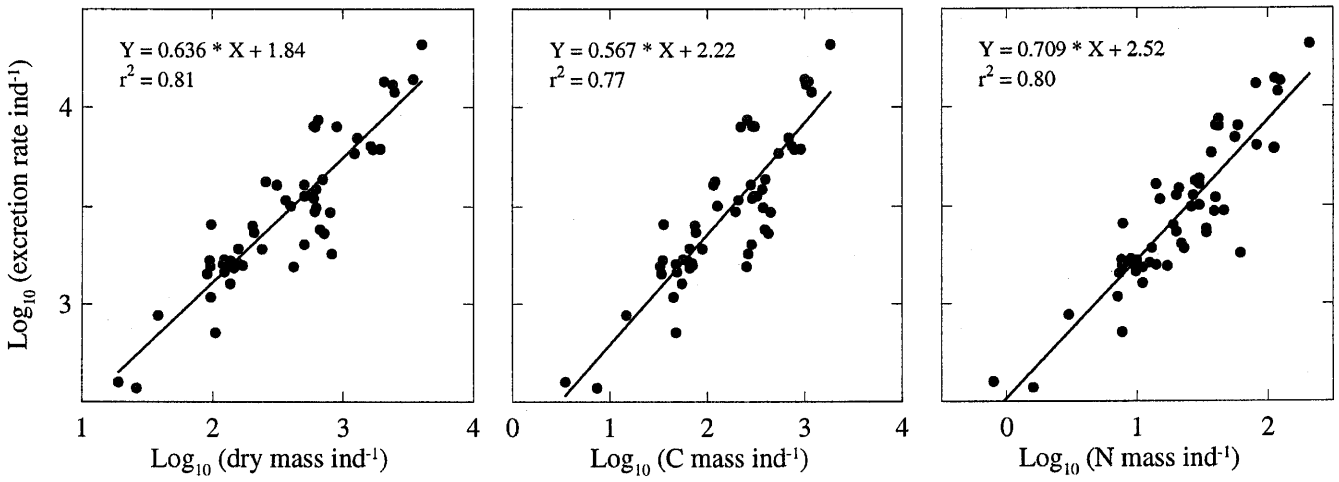

Fig. 3 Relationship between $\log_{10}$ ammonium-excretion rate (nmol ind^{-1} h^{-1}) and $\log_{10}$ body mass (expressed as micrograms drymass, micrograms C mass and micrograms N mass) for copepods and small euphausiids. The least-squares linear regressions are all significant ($P < 0.01$)

allowed the construction of allometric relationships between excretion rate and body mass (Fig. 3). The net catches were then enumerated and, for those species and stages not incubated, excretion rates were estimated from the regression line. Total excretion by the whole assemblage was then estimated by summation. We stress that this is an estimation method. Because we could not incubate the smallest fraction, their excretion rates were obtained from extrapolation beyond the size limits of the species incubated.

This approach was used to estimate the total ammonium regenerated by mesozooplankton in the top 50-m layer in the eastern and western monitoring areas

Table 6 Estimation of ammonium regenerated by copepods and small euphausiids within the upper 50 m of the eastern and western monitoring areas. *Italicised* values are those just for the species and stages which were incubated, and the total estimate is in bold. All values are means, and the ranges across the five eastern and six western area stations are in parentheses

Monitoring area	Abundance no. 1,000 m^{-3}	Biomass g DM m^{-2}	Ammonium excretion mmol m^{-2} day^{-1}
Eastern	*283 (96–512)*	*3.7 (0.43–6.5)*	*0.82 (0.32–1.8)*
	2,140 (974–5,040)	**4.0 (0.69–7.0)**	**0.97 (0.39–1.9)**
Western	*287 (219–411)*	*2.7 (0.75–4.7)*	*0.50 (0.21–0.83)*
	3,030 (1510–6,660)	**3.1 (0.97–5.4)**	**0.95 (0.45–1.8)**

Table 7 Estimation of the role of metazoan zooplankton in regenerating ammonium in the eastern and western monitoring areas

Rate variable	Eastern area	Western area	Reference, method of calculation
Median and range of primary production (g C m^{-2} day^{-1})	0.45 (0.069–1.2)	1.2 (0.32–8.9)	Owens et al (1991) Atkinson et al. (1996a) Pakhomov et al. (1997) Gilpin et al. (unpublished data)
Mean *f* ratio	0.46	0.46	From Owens et al. (1991)
Estimated median ammonium demand (mmol m^{-2} day^{-1})	3.0	8.1	This study, see text
Ammonium regenerated by copepods and small euphausiids in the top 50-m layer (mmol m^{-2} day^{-1})	0.97	0.95	This study, from Table 6
Estimate of % contribution of copepods and small euphausiids to ammonium demand	32%	12%	This study, based on the values in the two rows above
Estimate of % contribution of *Euphausia superba* to ammonium demand [a]	16–50% [b]	3–4% [b]	Atkinson and Whitehouse (2000)

[a] Estimate based on krill excretion experiments from two summer South Georgia surveys; the biomasses of krill obtained during three seasons of acoustic surveys; and "ammonium demand" of phytoplankton estimated in the same manner as in this study

[b] These percentage values for the eastern and western monitoring areas had been inadvertently transposed in Table 2 of Atkinson and Whitehouse (2000), and are presented in the correct order here

(Fig. 1). These two areas have been sampled intensively during December–January in recent years, providing a baseline for this study. Within both boxes, mesozooplankton biomass in the top 50 m was substantial (Table 6), being dominated by *Calanoides acutus* CV, with important contributions from *Rhincalanus gigas* CIII–CVI♀ and small euphausiids. Total mesozooplankton abundance was also high ($>2{,}000$ ind m^{-3}).

In addition to the biomass-dominant zooplankton which were incubated, numerous smaller copepods and pteropods were not, owing to methodological difficulties (see Materials and methods, Experiments). The small copepods (chiefly *Oithona similis*, and small copepodites of *Ctenocalanus* spp. and *Drepanopus forcipatus*) dominated numerically but contributed little to biomass (Table 6). Length–mass regressions based on our previous unpublished data at South Georgia as well as the present work (Table 2) were used to estimate the DM of the small copepods. Their excretion rate was then calculated from their DM using the regression in the left hand panel of Fig. 3.

The small-copepod fraction was estimated to provide a large proportion ($>30\%$) of the total ammonium produced by copepods and small euphausiids in the top 50-m layer (Table 6). Small copepods were particularly numerous in the western area, and here they were estimated to excrete a similar amount to that of the larger copepods and small euphausiids. These ammonium-regeneration rates can be compared with estimates of uptake rate by phytoplankton (Table 7). No primary-production measurements are available for this survey, so values from previous summers are compared with our values of zooplankton biomass and excretion rates (Table 7). Median primary-production values were converted to a median "ammonium demand" by applying a Redfield ratio of C:N uptake of 100:15 and an f ratio (i.e. nitrate uptake as a fraction of nitrate plus ammonium uptake) of 0.46, a value previously obtained for the two regions in summer (Owens et al. 1991). These estimates suggest a major role for metazoan zooplankton in regenerating ammonium in the eastern area, where summer primary production is often lower than in the western area.

These are crude calculations to determine whether or not metazoan zooplankton are important regenerators of ammonium in the South Georgia system. Their importance is likely to vary widely according to variations in zooplankton biomass, rates of primary production, and f ratio. For instance, the studied summer was unusual in that a bloom occurred in the eastern area as well as in the western area. Whether the zooplankton biomasses we found are typical is unknown. High standing stocks have been found previously in the western area (Ward et al. 1995; Atkinson et al. 1996a, but the few observations in the eastern area suggest that copepods are often depleted here and krill are more abundant than in the west (Pakhomov et al. 1997; Atkinson et al. 1999).

Notwithstanding these caveats, we have been conservative in our estimates of ammonium-regeneration rate. They are based on ~15-h incubations in filtered sea water, which could lead to up to ~50% underestimates of in situ rates. Further, only the copepods and small euphausiids are included in the present estimate. Antarctic krill are numerous in the eastern area in some seasons, and a previous study (Atkinson and Whitehouse 2000) suggested their importance in ammonium regeneration here (Table 7). Other taxa not included are pteropods, amphipods and chaetognaths which could increase the biomass by 30% (Ward et al. 1995; Atkinson et al. 1996a; Pakhomov et al. 1997), as well as the micrometazoans not retained by our 200-μm-mesh nets. Nevertheless, this study suggests that metazooplankton excretion may at times be an important process at South Georgia.

Acknowledgements We first thank the officers and crew of the R.R.S. "James Clark Ross" for their professional and expert help at sea. R.S. Shreeve gave us guidance and practical help with the elemental analysis and J.H. Priddle provided encouragement at all stages of this study. We also thank L.C. Gilpin for unpublished primary production data. For comments on the manuscript we are grateful to R.S. Shreeve, A.G. Hirst, J.L. Watkins, E.J. Murphy, P. Ward, and three anonymous referees.

References

Alcaraz M, Saiz E, Fernandez JA, Trepat I, Figueiras F, Calbet A, Bautista B (1998) Antarctic zooplankton metabolism: carbon requirements and ammonium excretion by salps and crustacean zooplankton in the vicinity of the Bransfield Strait during January 1994. J Mar Syst 17:347–359

Antezana T, Ray K (1984) Active feeding of *Euphausia superba* in a swarm north of Elephant Island. J Crustac Biol 4 [Spec No 1]:142–155

Atkinson A (1996) Subantarctic copepods in an oceanic, low-chlorophyll environment: ciliate predation, food selectivity and impact on prey populations. Mar Ecol Prog Ser 130:85–96

Atkinson A, Shreeve RS (1995) Response of the copepod community to a spring bloom in the Bellingshausen Sea. Deep-Sea Res II 42:1291–1311

Atkinson A, Whitehouse MJ (2000) Ammonium excretion by Antarctic krill *Euphausia superba* at South Georgia. Limnol Oceanogr 45:55–63

Atkinson A, Shreeve RS, Pakhomov EA, Priddle J, Blight SP, Ward P (1996a) Zooplankton response to a phytoplankton bloom near South Georgia, Antarctica. Mar Ecol Prog Ser 144:195–210

Atkinson A, Ward P, Murphy EJ (1996b) Diel periodicity of subantarctic copepods: relationships between vertical migration, gut fullness and gut evacuation rate. J Plankton Res 18:1387–1405

Atkinson A, Ward P, Hill A, Brierley AS, Cripps GC (1999) Krill–copepod interactions at South Georgia, Antarctica. II. *Euphausia superba* as a major control on copepod abundance. Mar Ecol Prog Ser 176:63–79

Atkinson A, Whitehouse MJ, Priddle J, Cripps GC, Ward P, Brandon MA (in press) South Georgia, Antarctica: a productive, cold-water pelagic ecosystem. Mar Ecol Prog Ser

Båmstedt U (1985) Seasonal excretion rates of macrozooplankton from the Swedish west coast. Limnol Oceanogr 30:607–617

Båmstedt U, Tande KS (1985) Respiration and excretion rates of *Calanus glacialis* in Arctic waters of the Barents Sea. Mar Biol 87:259–266

Banse K (1995) Zooplankton: pivotal role in the control of ocean production. ICES J Mar Sci 52:265–277

Biggs DC (1982) Zooplankton excretion and NH_4^+ cycling in near-surface waters of the Southern Ocean. I. Ross Sea, austral summer1977–1978. Polar Biol 1:55–67

Boyce SJ, Clarke A (1997) Effect of body size and ration on specific dynamic action in the Antarctic plunderfish, *Harpagifer antarcticus* Nybelin 1947. Physiol Zool 70:679–690

Brandon MA, Murphy EJ, Whitehouse MJ, Trathan PN, Murray AWA, Bone DG, Priddle J (1999) The shelf break front to the east of the sub-Antarctic island of South Georgia. Cont Shelf Res 19:799–819

Chapelle G, Peck LS, Clarke A (1994) Effects of feeding and starvation on the metabolic rate of the necrophagous Antarctic amphipod *Waldeckia obesa* (Chevreux, 1905). J Exp Mar Biol Ecol 183:63–76

Checkley DM Jr, Dagg MJ, Uye S (1992) Feeding, excretion and egg production by individuals and populations of the marine, planktonic copepods, *Acartia* spp. and *Centropages furcatus*. J Plankton Res 14:71–96

Conover RJ, Huntley M (1991) Copepods in ice-covered seas – distribution, adaptations to seasonally limited food, metabolism, growth patterns and life cycle strategies in polar seas. J Mar Syst 2:1–41

Donnelly J, Torres JJ, Hopkins TL (1994) Chemical composition of Antarctic zooplankton during austral fall and winter. Polar Biol 14:171–183

Dubischar CD, Bathmann UV (1997) Grazing impact of copepods and salps on phytoplankton in the Atlantic sector of the Southern Ocean. Deep-Sea Res II 44:415–433

Fransz HG, Gonzalez SR (1995) The production of *Oithona similis* (Copepoda: Cyclopoida) in the Southern Ocean. ICES J Mar Sci 52:549–555

Gardner WS, Paffenhöfer G-A (1982) Nitrogen regeneration by the subtropical marine copepod *Eucalanus pileatus*. J Plankton Res 4:725–734

Gifford DJ (1993) Protozoa in the diets of *Neocalanus* spp. in the oceanic subarctic Pacific Ocean. Prog Oceanogr 32:223–237

Glibert PM, Miller CA, Garside C, Roman MR, McManus GB (1992) NH_4^+ regeneration and grazing: interdependent processes in size-fractionated $^{15}NH_4^+$ experiments. Mar Ecol Prog Ser 82:65–74

Hagen W (1988) On the significance of lipids in Antarctic zooplankton (in German). Ber Polarforsch 49:1–129. English version (1989): Can Transl Fish Aquat Sci 5458:1–149

Hagen W, Schnack-Schiel SB (1996) Seasonal lipid dynamics in dominant Antarctic copepods: energy for overwintering or reproduction? Deep-Sea Res I: 139–158

Harris RP, Malej A (1986) Diel patterns of ammonium excretion and grazing rhythms in *Calanus helgolandicus* in surface stratified waters. Mar Ecol Prog Ser 31:75–85

Hernández-Leon S, Torres S, Gómez M, Montero I, Almeida C (1999) Biomass and metabolism of zooplankton in the Bransfield Strait (Antarctic Peninsula) during austral spring. Polar Biol 21:214–219

Huntley ME, Nordhausen W (1995) Ammonium cycling by Antarctic zooplankton in winter. Mar Biol 121:457–467

Ikeda T (1985) Metabolic rates of epipelagic zooplankton as a function of body mass and temperature. Mar Biol 85:1–11

Ikeda T, Mitchell AW (1982) Oxygen uptake, ammonia excretion and phosphate excretion by krill and other Antarctic zooplankton in relation to their body size and chemical composition. Mar Biol 71:283–298

Ikeda T, Hing-Fay E (1981) Metabolic activity of zooplankton from the Antarctic ocean. Aust J Mar Freshw Res 32:921–930

Ikeda T, Hing-Fay E, Hutchinson SA, Boto GM (1982) Ammonia and inorganic phosphate excretion by zooplankton from inshore waters of the Great Barrier Reef, Queensland. I. Relationship between excretion rate and body size. Aust J Mar Freshw Res 33:55–70

Ikeda T, Torres JJ, Hernández-León S, Geiger SP (2000) Metabolism. In: Harris R, Wiebe P, Lenz J, Skjoldal HR, Huntley M (eds) ICES Zooplankton methodology manual. Academic Press, London, pp 455–532

Johnson MA, Macaulay MC, Biggs DC (1984) Respiration and excretion within a mass aggregation of *Euphausia superba*: implications for krill distribution. J Crustac Biol 4 [Spec No 1]:174–184

Kiørboe T, Tiselius PT (1987) Gut clearance and pigment destruction in a herbivorous copepod, *Acartia tonsa*, and determination of in situ grazing rates. J Plankton Res 9:525–534

Lopez MDG, Huntley ME (1995) Feeding and diel vertical migration cycles of *Metridia gerlachei* (Giesbrecht) in coastal waters of the Antarctic Peninsula. Polar Biol 15:21–30

Miller CA, Glibert PA (1998) Nitrogen excretion by the calanoid copepod *Acartia tonsa*: results of mesocosm experiments. J Plankton Res 20:1767–1780

Moloney CL, Field JG (1989) General allometric equations for rates of nutrient uptake, ingestion, and respiration in plankton organisms. Limnol Oceanogr 34:1290–1299

Moloney CL, Field JG (1991) The size-based dynamics of plankton food webs. I. A simulation model of carbon and nitrogen flows. J Plankton Res 13:1003–1038

Morales CE, Bedo A, Harris RP, Tranter PRG (1993) Grazing of copepod assemblages in the north-east Atlantic: the importance of the small size fraction. J Plankton Res 13:455–472

Owens NJP, Priddle J, Whitehouse MJ (1991) Variations in phytoplanktonic nitrogen assimilation around South Georgia and in the Bransfield Strait (Southern Ocean). Mar Chem 35:287–304

Paffenhöfer G-A (1971) Grazing and ingestion rates of nauplii, copepodids and adults of the marine copepod *Calanus helgolandicus*. Mar Biol 11:286–298

Paffenhöfer G-A, Gardner WS (1984) Ammonium release by juveniles and adult females of the sub-tropical marine copepod *Eucalanus pileatus*. J Plankton Res 6:505–513

Pakhomov EA, Verheye HM, Atkinson A, Laubscher RK, Taunton-Clark J (1997) Structure and grazing impact of the mesozooplankton community during late summer 1994 near South Georgia, Antarctica. Polar Biol 18:180–192

Pasternak AF, Kosobokova KN, Drits AV (1994) Feeding, metabolism and body composition of the dominant Antarctic copepods with comments on their life cycles. Russ J Aquat Ecol 3:49–62

Peters RH, Downing JA (1984) Empirical analysis of zooplankton filtering and feeding rates. Limnol Oceanogr 36:147–159

Priddle J, Whitehouse MJ, Atkinson A, Brierley AS, Murphy EJ (1997) Diurnal changes in near-surface ammonium concentrations – interplay between zooplankton and phytoplankton. J Plankton Res 19:1305–1330

Razouls S, Du Reau G, Guillot P, Maison J, Jeandel C (1998) Seasonal abundance of copepod assemblages and grazing pressure in the Kerguelen Island area (Southern Ocean). J Plankton Res 20:1599–1614

Schnack SB, Smetacek V, Bodungen Bv, Stegmann P (1985) Utilization of phytoplankton by copepods in Antarctic waters during spring. In: Gray JS, Christiansen ME (eds) Marine biology of polar regions and effects of stress on marine organisms. Wiley, London, pp 65–81

Schnack-Schiel SB, Hagen W (1995) Life-cycle strategies of *Calanoides acutus*, *Calanus propinquus*, and *Metridia gerlachei* (Copepoda: Calanoida) in the eastern Weddell Sea, Antarctica. ICES J Mar Sci 52:541–548

Shreeve RS, Ward P (1998) Moulting and growth of the early stages of two species of Antarctic copepod in relation to differences in food supply. Mar Ecol Prog Ser 175:109–119

Skjoldal HR, Båmstedt U, Klinken J, Laing A (1984) Changes with time after capture in the metabolic activity of the carnivorous copepod *Euchaeta norvegica* Boeck. J Exp Mar Biol Ecol 83:195–210

Vidal J, Whitledge TE (1982) Rates of metabolism of planktonic crustaceans as related to body weight and temperature of habitat. J Plankton Res 4:77–84

Voronina NM (1998) Comparative abundance and distribution of major filter-feeders in the Antarctic pelagic zone. J Mar Syst 17:375–390

Ward P, Atkinson A, Murray AWA, Wood AG, Williams R, Poulet SA (1995) The summer zooplankton community at South Georgia: biomass, vertical migration and grazing. Polar Biol 15:195–208

Ward P, Shreeve RS, Cripps GC (1996) *Rhincalanus gigas* and *Calanus simillimus*: lipid storage patterns of two species of copepod in the seasonally ice free zone of the Southern Ocean. J Plankton Res 18:1439–1454

Wen YH, Peters RH (1994) Empirical models of phosphorus and nitrogen excretion by zooplankton. Limnol Oceanogr 39:1669–1679

Whitehouse MJ (1997) Automated seawater nutrient chemistry. British Antarctic Survey, Cambridge

Whitehouse MJ, Woodley VR (1987) Automated seawater nutrient analysis. British Antarctic Survey, Cambridge

Whitehouse MJ, Priddle J, Brandon MA, Swanson C (1999) A comparison of chlorophyll/nutrient dynamics at two survey sites near South Georgia, and the potential role of planktonic nitrogen recycled by land-based predators. Limnol Oceanogr 44:1498–1508

Whitehouse MJ, Priddle J, Brandon MA (2000) Chlorophyll/nutrient characteristics in the water masses to the north of South Georgia, Southern Ocean. Polar Biol 23:373–382