Spatial distribution of microbial biomass and activity (bacterivory and bacterial production) in the northern Weddell Sea during the austral summer (January 1994)

Dolors Vaqué*, Juan I. Calderón-Paz, Núria Guixa-Boixereu, Carlos Pedrós-Alió

Institut de Ciències del Mar-CMIMA (CSIC), Departament de Biologia Marina i Oceanografia, Passeig Marítim de la Barceloneta 37-49, 08003-Barcelona, Spain

ABSTRACT: Changes in bacterial production, bacterivory and microbial biomass (autotrophic and heterotrophic) were determined in a complex area characterized by different physico-chemical regimes during the austral summer (January 1994, ECOANTAR-94 cruise). The main goal was to explore the importance of bacterivory as a factor potentially controlling bacterial production in such an extreme environment. The area studied was located in the eastern part of the Bransfield Strait, between 59.5–64° S and 58–50° W. It covered part of the South Shetland Islands shelf and extended northwards to the Weddell-Scotia Confluence and southwards towards the ice edge. Stations were grouped into 4 zones under the influence of different hydrographic conditions: IEZ, influenced directly by the ice edge zone; WS, presenting typical characteristics of the Weddell Sea; FR, located within the Bransfield Strait frontal system and its continuation in the Weddell-Scotia Confluence; and BSO-WSC, containing stations influenced by waters originating in the Bellingshausen Sea and the Weddell-Scotia Confluence. We determined the spatial distribution of microbial biomass (chlorophyll a, bacteria, heterotrophic and phototrophic nanoflagellates and ciliates), bacterial heterotrophic production, and bacterivory. The highest values of chlorophyll a concentration and primary production as well as the lowest grazing rates (0.3 µg C l^{-1} d^{-1}) and bacterial production values were found at IEZ, together with the lowest temperatures. In contrast, the highest bacterial losses (4.7 µg C l^{-1} d^{-1}) coincided with high heterotrophic nanoflagellate biomass and bacterial production at the Weddell-Scotia Confluence and waters of Bellingshausen Sea origin (BSO-WSC). These waters showed the highest temperatures, and low chlorophyll a concentration and primary production values. Taking into account the whole data set, bacteria and heterotrophic nanoflagellate biomass explained 68% of the variability in grazing rates. Our results indicated that bacterial losses due to protists represented a large proportion of bacterial production (average 67% d^{-1}). However, the variability detected in the study area was very large (from undetectable to >100%). Therefore, bacterivory does not seem to be the main factor in controlling bacterial production in all the zones studied.

KEY WORDS: Temperature · Hydrographic characteristics · Bacterial abundance · Bacterial production · Protist abundance · Grazing rates · Weddell Sea · Antarctica

INTRODUCTION

Abundance values reported for bacteria and protists in Antarctic waters (see Huntley et al. 1991 and references therein) are comparable to those of temperate marine systems (i.e. Kanhoe Bay, Landry et al. 1984; Delaware Estuary, Coffin & Sharp 1987; coastal Mediterranean Sea, Vaqué et al. 1997). However, in Antarctic coastal areas, there seems to be either no response (Rivkin et al. 1991), or a very weak one (Karl et al. 1991, Bird & Karl 1999) in bacterial biomass to phytoplankton blooms. Thus, bacterial biomass may be an

order of magnitude lower than predicted on the basis of relationships derived from lower latitudes (Bird & Kalff 1984). In addition, Bird & Karl (1999) found that at high latitudes bacterial production was a small fraction of primary production, around 10 times less than that expected by the relationship of Cole et al. (1988) in warmer waters. Different explanations have been given for this weak bacterial response: differential effects of low temperature on microbial metabolism (Pomeroy & Deibel 1986), higher substrate requirement at freezing temperatures (Wiebe et al. 1993, Pomeroy & Wiebe 2001), or direct competition for limiting organic substrates (Karl & Bird 1993). In temperate areas bacterial production is generally controlled by substrate availability, while bacterial biomass is controlled by predation, and the balance between growth and consumption determines the actual biomass found (Pace 1988, Vaqué et al. 1994, Pedrós-Alió et al. 2000). Hence, if bacterial assemblages in Antarctic waters have a lower production and biomass than expected, we pose the question of whether protists play an important role in the regulation of bacterial biomass and production, and in turn whether bacteria could be an important carbon source for their immediate predators. However, microbial productivity and indeed the importance of the microbial food web can vary depending on the physico-chemical characteristics of the water column in different areas of the Southern Ocean. Thus, temperature, distribution of organic and inorganic nutrients, photosynthetically available radiation (PAR), and the presence of ice are also significant factors in controlling the rates of microbial processes (Sullivan et al. 1990, Kang et al. 1997). Large spatial and seasonal variability in microbial productivity and biomass has been found in different geographical locations (open sea, continental shelves, coastal embayments, pack ice, etc.). This has been shown in previous studies such as EPOS (e.g. Veth et al. 1992, Lancelot et al. 1993), AMERIEZ (e.g. Nelson et al. 1989, Sullivan et al. 1990), and RACER (e.g. Bird & Karl 1990). Since the early 1980s, data on hydrography and biological processes in Antarctic waters have increased. Nevertheless there are relatively few studies on the importance of grazing on bacteria by protists, as one of the potential factors that could control bacterial production and biomass.

Our main goal was to explore the spatial distribution of heterotrophic microbial biomass and to determine the importance of bacterivory as a factor potentially controlling bacterial production in a hydrographically variable area. We determined hydrographic (i.e. temperature, salinity) and biological parameters such as chlorophyll *a* (chl *a*), primary production (Figueiras et al. 1998), bacterial and protist abundance and biomass, and heterotrophic microbial activities (grazing rates and bacterial production) at selected stations. First, we

determined whether there were significant differences in each biological variable among different areas. Second, we also investigated the hydrographical or biological variables that could best explain changes in bacterivory. Third, we evaluated whether bacterivory exerted an important control on bacterial productivity and whether the bacterial carbon consumed by protists was enough to explain their actual biomass and growth.

MATERIALS AND METHODS

Study area and sampling. Cruise ECOANTAR-94 was carried out during the austral summer (January 9 to February 4, 1994) on board BIO 'Hespérides'. The area studied during this cruise was located in the eastern part of the Bransfield Strait, between 59.5–64°S and 58–50°W (Fig. 1). It covers part of the South Shetland Islands shelf and extends northwards to the Weddell-Scotia Confluence and southwards towards the ice edge. Stations were grouped into 4 zones under the influence of different hydrographic conditions: Stns 1, 3, 4, 6, 7 and 119, influenced directly by the ice edge zone (IEZ); Stns 8, 9, 10, 11, 17, 18, 19, 38, 63, 91 and 93, typical of the Weddell Sea (WS); Stns 12, 15, 16, 56 and 76, located within the Bransfield Strait frontal system and its continuation in the Weddell-Scotia Conflu-
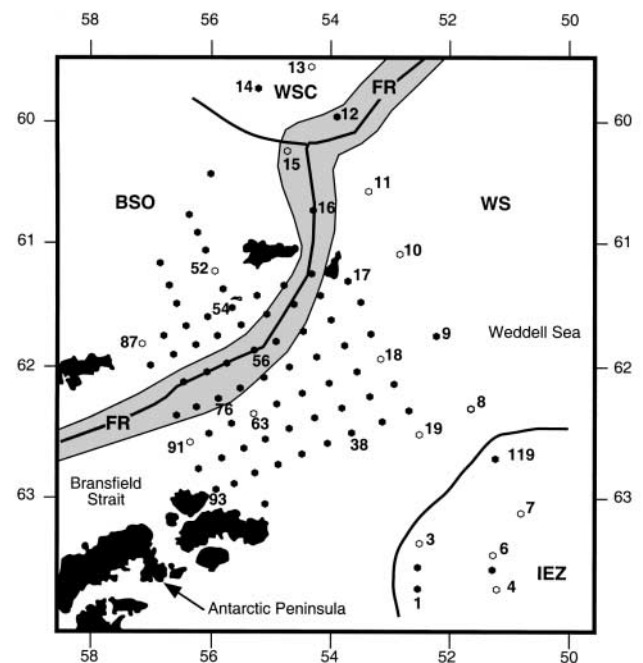

Fig. 1. Location of the stations sampled and the approximate boundaries of the different hydrographic areas (bold lines). Open symbols correspond to the stations where grazing determinations were carried out. BSO: waters of Bellingshausen Sea origin; FR: Bransfield Strait frontal system; IEZ: ice edge zone; WS: Weddell Sea; WSC: Weddell-Scotia Confluence

ence (FR); and Stns 13, 14, 52, 54 and 87, with warmer (>1.2°C) and less saline (<34.2‰) waters, influenced by waters originating in the Bellingshausen Sea and the Weddell-Scotia Confluence (BSO-WSC). Profiles of salinity, temperature and fluorescence were obtained using a conductivity, temperature, depth profiler (CTD) EG&G model MkIIIC WOCE between 5 and 1000 m depth.

Chl *a* concentration, abundance and biomass of bacteria and protists. Samples for biological variables and nutrient concentration were taken from the surface to 150 m depth at 10 m intervals with 10 l Niskin bottles attached to a rosette sampler system.

Chl *a* concentration was estimated fluorometrically (Strickland & Parsons 1972). Samples (100 ml) were collected on 25 mm GF/F glass fiber filters and immediately frozen at –70°C. For more details see Berdalet et al. (1997). Six samples for bacterioplankton and nanoflagellate abundance were taken from the surface to below the deep chlorophyll maximum (DCM) at 10 to 20 m intervals at each of the indicated stations (Fig. 1). Water samples of 100 ml (for pico- and nanoplankton) were preserved with glutaraldehyde (1% final conc.) and stored from a few to 48 h at 4°C until staining and filtration. Subsamples of 10 to 20 ml (bacterioplankton) and 30 to 40 ml (nanoflagellates) were stained with DAPI for 5 min (Porter & Feig 1980, final conc. 1 µg ml⁻¹) and filtered through 0.2 and 0.6 µm black-stained polycarbonate filters to collect bacterioplankton and nanoflagellates respectively. Filters were then mounted on a slide with a drop of immersion oil and frozen at –20°C until examination by microscope after the cruise (~1 mo). Abundance of these microorganisms was determined by epifluorescence with a microscope at 1250× (Nikon Optiphot). For routine vertical profiles 1 replicate was counted. For counts of grazing experiments 2 replicates were counted. Ciliate abundance was examined in 1 l samples (from selected stations and depths) preserved in acid Lugol's solution (1% final conc.), and settled in 100 ml sedimentation chambers for at least 48 h before enumeration. Samples were examined at 200× or 400× magnification using an inverted microscope (Axiovert 35, Zeiss). One replicate was counted per sample. Ciliates were identified to genus level when possible (Lee et al. 1985). Ciliates were grouped into naked oligotrichs (*Halteria*, *Strombidium*, *Strobilidium*, *Laboea* and *Tontonia*), loricate oligotrichs (e.g. tinitinids), haptorids (e.g. *Mesodinium*), and others (including mainly scuticociliates). The average cell volume of bacteria was obtained from samples of a single station (Stn 19 at 10 m), and was measured using an image analysis system, following the protocol of Massana et al. (1997). This cell volume was transformed to carbon using the equation of Norland (1993), pg C = 0.12 × (cell volume)$^{0.7}$.

To determine the size of protists we measured their lengths and widths. Cell volumes were estimated by assuming the nearest geometrical figure. Average cell volumes of nanoflagellates were obtained by measuring 20 cells from samples of 3 different stations (Stns 7, 52 and 91). Carbon content was estimated using a literature carbon conversion factor, 0.22 pg C µm⁻³ (Børsheim & Bratbak 1987). The average ciliate cell volume for each identified group was converted to carbon equivalents using the experimentally derived factor for Lugol's fixed marine oligotrichs, 0.2 pg C µm⁻³ (Putt & Stoecker 1989). Tintinnid carbon was estimated using the experimentally determined factor of 0.053 pg C µm⁻³ (Verity & Langdon 1984). Chl *a* was converted to carbon by using a factor of 40 (Li et al. 1993).

Primary and bacterial heterotrophic production. The primary production data used in this paper are taken from Figueiras et al. (1998), and were obtained following the methodology of Steeman Nielsen (1952). Bacterial heterotrophic production (BHP) was estimated by the tritiated leucine incorporation technique. The method described in Kirchman (1993) was used with slight modifications. A stock solution of L-(4.5³H)leucine (153 Ci mmol⁻¹, Amersham) at a final concentration in samples of 10 nM ('hot' leucine diluted 1/10 with non-radioactive leucine) was used. One formaldehyde-killed control and 2 replicates of 20 ml were incubated for approximately 4 h, in the dark and at 0°C. Incubations were stopped by adding formaldehyde (4% final conc.) and stored at 4°C and in the dark until processed (<48 h). The whole volume was then filtered through polycarbonate 0.2 µm pore diameter filters. The filters were placed in scintillation vials with 5 ml of scintillation cocktail. Radioactivity was measured on a Beckman LS 6000 scintillation counter by previously dissolving the filter in 0.5 ml ethyl-acetate and adding 5 ml of Wallac Optiphase Hisafe 2 scintillation cocktail. Conversion of leucine incorporation to cells produced per mol was carried out with conversion factors of 1.44×10^{17} cells mol⁻¹ and 3.61×10^{17} cells mol⁻¹ determined with several cultures from Stns 4 (IEZ), and Stns 52 and 87 (BSO-WSC), respectively (Calderón-Paz 1997). The first was used in the IEZ, and the second for all other stations.

Bacterivory. Grazing on bacteria was determined at 15 stations representative of the 4 hydrographic areas considered (see Table 2). At 11 stations, grazing was measured at 2 depths (surface and DCM), and at 4 stations at only 1 depth (surface). Estimates of grazing by protists were determined by disappearance of fluorescent minicells (*Escherichia coli* strain X-1488, Genetic Stock Center, Yale University), following Pace et al. (1990). Minicells were added to 1 l samples in duplicate (previously filtered through 150 µm to avoid large predators) at 5 to 20% of natural bacteria concentra-

tions. Formaldehyde-killed controls were run every time. Incubations were carried out in the dark at 0°C for 48 h. Incubations were stopped with ice-cold glutaraldehyde (1% final conc.). The average cell volume of the minicells was 0.065 μm^3, similar to the average volume of natural bacteria (0.070 μm^3). Minicell and bacterial abundance were determined at the beginning of the experiment and at 48 h by epifluorescence microscopy (in duplicates). Nanoflagellates were counted at the beginning of the experiment. Calculations of bacteria consumed were done following the model of Salat & Marrasé (1994), where specific grazing rates (g, d^{-1}) are:

$$g = -1/t \cdot \ln(Mi_t/Mi_0)$$

where t = incubation time; Mi_t = number of minicells at final time; and Mi_0 = number of minicells at initial time. Bacterial net growth rates (a, d^{-1}) are:

$$a = 1/t \cdot \ln(BN_t/BN_0)$$

Finally, total bacterial grazing rates (G, cells l^{-1} d^{-1}) are:

$$G = g/a \cdot (BN_t - BN_0)/t$$

where t = incubation time; BN_t = bacterial number at the end of the experiment; and BN_0 = bacterial number at the beginning of the experiment.

Net increase in bacterial abundance equals net BHP (NBHP, bacteria l^{-1} d^{-1}), and can be obtained from the difference between the number of natural bacteria at time t (BN_t, cells l^{-1}) and at time zero (BN_0, cells l^{-1}):

$$NBHP = (1/t) \cdot (BN_t - BN_0)$$

Gross bacterial heterotrophic production (GBHP, cells l^{-1} d^{-1}) was calculated as the sum of total grazing (G) plus NBHP

$$GBHP = NBHP + G$$

These calculations were based on the assumption that all losses of bacteria during the incubations were due to grazing by protists.

Finally, specific ingestion rates (bacteria per heterotrophic nanoflagellate [HNF] h^{-1}) were calculated from total grazing rates (G) divided by HNF abundance, assuming that HNF were the only bacterivores.

HNF carbon requirements. Carbon requirements (in μg C l^{-1} d^{-1}) for HNF growth were estimated with the following equation:

$$Carbon\ requirement = \mu Bv\,C/0.40$$

where 0.40 = assumed growth efficiency (Sanders et al. 1992), μ (d^{-1}) = HNF growth rate, Bv (μm^3 l^{-1}) = HNF assemblage biovolume, and C (C, μm^{-3}) = carbon content per HNF cell. Unfortunately we did not have data on the HNF gross growth rate, so instead we used HNF net growth rate (μ, d^{-1}) values that were obtained

by counting their abundance at the beginning and at the end of the incubations of grazing experiments at Stns 52 and 91.

Temperature corrections. Bacterial production and bacterivory rates were always determined at 0°C. *In situ* temperatures, however, varied between –1.8 and 2.5°C. Corrections for *in situ* temperatures were done using experimentally determined Q_{10} values. The latter were obtained following the equation $Q_{10} = 10^{10 \cdot B}$, where B is the regression coefficient of the exponential regression between the rate and temperatures (i.e. Choi & Peters 1992). The Q_{10} values used for tritiated leucine incorporation were obtained from different experiments in the study areas described in Calderón-Paz (1997). The Q_{10} values used were Q_{10} = 5.37 in IEZ and WS, 4.46 in FR, and 3.55 in BSO-WSC waters. Q_{10} values for bacterivory were obtained from a temperature experiment carried out in the Weddell Sea (Stn 91, Vaqué unpubl.). Water samples were incubated at 5 different temperatures (–1.1, –0.5, 3.5, 7.5 and 11.5) and the Q_{10} obtained (8.65) was used for all stations.

Data analysis. All data except for temperatures were log transformed in order to equalize variance.

Differences of each variable between zones were tested with ANOVA. Relationships between variables were explored with Pearson correlation coefficients. To determine the main factors responsible for grazing rate variability we used multiple regression.

Data of different variables were integrated for the photic zone (down to 1% of surface light). The integrated values (m^{-2}) were then divided by the depth of the water column used as a reference, thus giving a weighted mean. This has the advantage of making the results comparable, despite the very different thickness of the photic layers at different stations (from 34 m at Stn 119 to 117 m at Stn 13).

RESULTS

Hydrography

A detailed study of the hydrography of the area was presented by López et al. (1995). A brief description is given here and the main hydrographic zones are shown in Fig. 1. A most relevant feature was the frontal system located in the center of the Bransfield Strait (FR) separating waters of Bellingshausen Sea origin (BSO, to the west and north) and Weddell Sea origin (WS, to the south and east). Another frontal system was located northeast of the study area and corresponded to the Weddell-Scotia Confluence (WSC). Finally, southeast of the study area close to the ice edge zone (IEZ), the salinity and temperature values indicated the presence of melt water at the surface (Berdalet et al. 1997).

Table 1. Average, maximum and minimum values of different variables determined in the 4 zones. IEZ: ice edge zone; WS: Weddell Sea; FR: Bransfield Strait frontal system; BSO-WSC:Bellingshausen Sea and Weddell-Scotia Confluence; HNF: heterotrophic nanoflagellates; PNF: phototrophic nanoflagellates; BHP: bacterial heterotrophic production; n: number of determinations; nd: not detectable

Variable	IEZ Average Min.–Max.	n	WS Average Min.–Max.	n	FR Average Min.–Max.	n	BSO-WSC Average Min.–.Max	n
Temperature (°C)	−1.24 −1.79−0.09	67	−0.26 −1.36−0.65	100	0.48 −1.06−1.65	53	1.08 −0.53−2.44	51
Chlorophyll a (µg l^{-1})	1.02 0.10−3.02	62	0.64 0.12−2.15	92	0.58 0.11−1.45	49	0.89 0.16−2.26	44
Bacteria ($\times 10^8$ cells l^{-1})	2.97 0.73−10.00	34	3.14 0.71−11.50	56	3.75 1.51−7.61	30	3.74 1.37−6.21	29
HNF ($\times 10^6$ cells l^{-1})	0.31 nd−1.33	33	1.80 0.29−5.64	41	3.30 0.77−9.71	17	4.94 0.20−11.84	14
PNF ($\times 10^6$ cells l^{-1})	2.39 0.10−7.56	33	4.45 0.35−10.99	39	4.69 0.49−9.991	17	3.68 1.20−7.99	12
BHP ($\times 10^7$ cells l^{-1} d^{-1})	3.31 nd−11.51	35	8.39 0.20−58.52	56	4.66 0.19−17.22	30	12.53 nd−33.50	30

Table 2. Temperature, chlorophyll a concentration, PNF biomass, bacteria (biomass, grazing and production estimated in the incubation bottles), HNF biomass, specific ingestion rates (Bact HNF^{-1} h^{-1}), and ciliate biomass determined at selected stations located in the 4 different zones. nd: not detected; –: no data

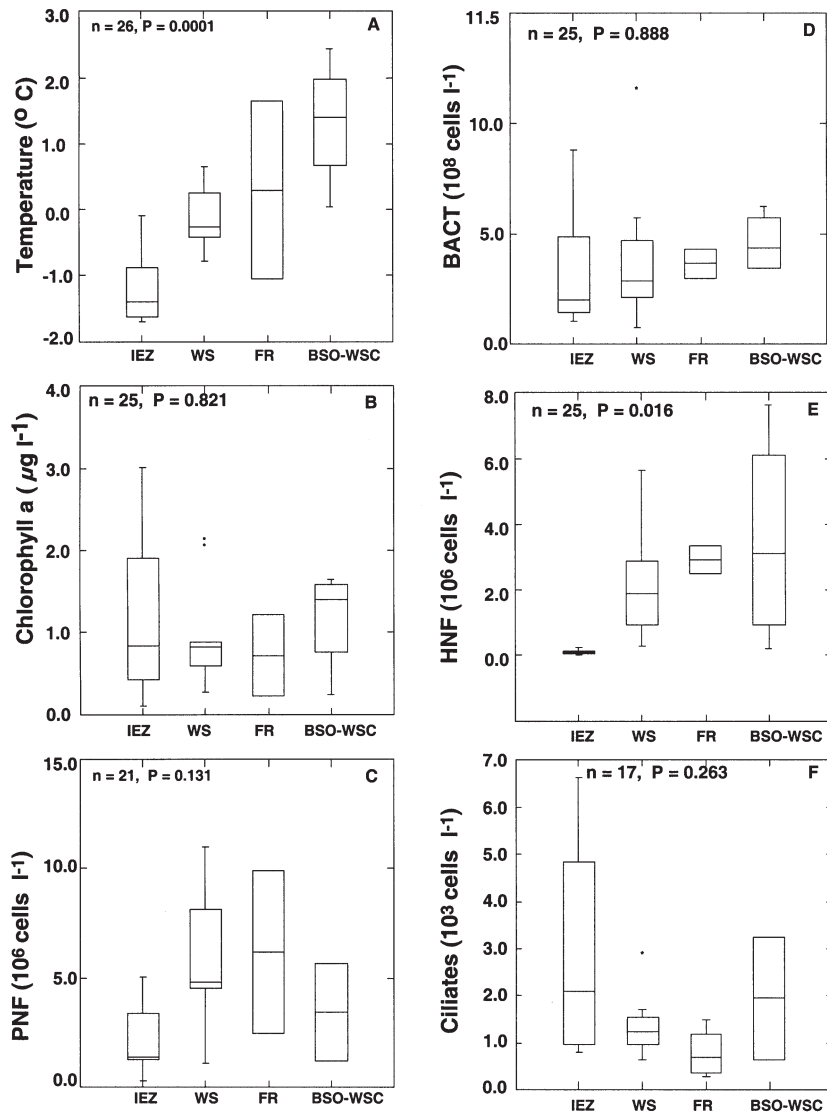| Stn	Depth (m)	Temp. (°C)	Chl (µg l^{-1})	PNF (µg C l^{-1})	Bacteria Biomass (µg C l^{-1})	Bacteria Grazing (µg C l^{-1} d^{-1})	Bacteria Production (µg C l^{-1} d^{-1})	HNF Biomass (µg C l^{-1})	HNF Ingestion rate (bacteria HNF^{-1} h^{-1})	Ciliates Biomass (µg C l^{-1})
IEZ										
3	0	−0.09	1.15	13.6	16.4	1.5	1.7	0.3	35.0	–
3	50	−1.30	2.67	7.3	13.4	1.8	3.8	0.2	66.9	–
4	0	−1.57	0.55	6.9	1.9	0.3	1.0	0.3	10.6	4.1
4	20	−1.69	0.51	7.0	4.7	0.9	0.7	0.6	12.6	–
6	0	−0.45	0.10	23.4	3.4	nd	1.3	0.1	nd	–
6	56	−1.48	1.13	4.2	2.1	nd	1.6	0.7	nd	–
7	0	−1.31	3.02	27.8	4.1	1.3	1.7	0.8	12.4	16.2
7	60	−1.70	0.35	1.4	3.1	0.6	0.5	0.2	30.6	0.6
WS										
8	0	−0.23	2.06	34.1	21.5	2.5	0.9	6.9	3.0	–
8	40	−0.27	2.15	24.8	8.4	4.0	7.9	3.3	9.6	–
10	0	−0.26	0.82	60.4	5.3	3.0	5.1	12.7	1.9	–
10	30	−0.33	0.69	44.8	5.0	3.4	3.9	7.6	3.6	–
11	0	0.65	–	27.6	3.3	nd	1.5	13.8	nd	–
11	40	0.58	0.87	10.2	3.1	nd	1.8	3.3	nd	1.4
18	0	0.41	0.89	25.0	1.3	nd	6.0	19.9	nd	1.9
18	50	0.40	0.82	25.2	7.2	nd	0.2	7.5	nd	1.8
19	0	0.09	0.27	47.6	4.6	nd	1.2	6.4	nd	1.1
19	60	−0.78	0.37	5.8	3.5	nd	2.3	3.2	nd	2.7
63	0	−0.60	0.53	–	9.1	nd	nd	1.0	nd	–
91	0	−0.50	0.64	–	10.7	nd	7.2	6.2	nd	–
FR										
15	0	1.65	0.22	54.5	8.0	2.5	3.2	8.8	2.2	2.8
15	70	−1.06	1.21	13.5	5.6	0.8	0.7	11.77	0.5	–
BSO-WSC										
13	0	2.44	0.23	31.1	11.6	4.7	4.5	16.1	2.3	–
13	70	0.04	1.30	6.6	9.8	3.8	3.5	26.9	1.1	0.3
52	5	1.49	1.52	–	6.4	2.1	3.4	5.9	2.8	–
87	5	1.30	1.65	–	6.4	0.7	1.4	0.7	7.5	–

Fig. 2. Box and whisker plots of different variables in each area. (A) Temperature, (B) chlorophyll *a* concentration, (C) phototrophic nanoflagellate abundance (PNF), (D) bacterial abundance (BACT), (E) Heterotrophic nanoflagellate abundance (HNF) and (F) Ciliate abundance. Horizontal lines within boxes indicate the median of the distribution, and the box limits are the 25% quartiles of the data. The whiskers cover the entire data range, except for outliers (•), some of which are off-scale. n: number of data, P: level of significance for the ANOVA test. Data corresponding to selected stations and depth where grazing rates were measured

Spatial distribution of temperature, microbial biomass, and bacterial activities

Mean values and range of temperature, chl *a* concentration, bacterial and nanoflagellate abundance and BHP are shown in Table 1. The highest values of chl *a* concentration (3.02 µg l^{-1}) and the lowest temperatures (–1.8°C) were found in the IEZ. Mean temperature values increased from IEZ to BSO-WSC. Both

maximal and minimal values of bacterial abundance were found in WS, although mean values were slightly higher in FR and BSO-WSC than in IEZ and WS (2.97 to 3.75 × 10^8 cells ml^{-1}). BHP, phototrophic nanoflagellate (PNF) and HNF abundance showed the lowest values in IEZ, maximum values for BHP and PNF were recorded in WS and maximum values for HNF abundance were recorded in BSO-WSC (Table 1). Mean values of HNF increased from the IEZ to BSO-WSC (0.31 × 10^6 to 4.94 × 10^6 cells l^{-1}). Average BHP reached the highest values in BSO-WSC (12.53 × 10^7 l^{-1} d^{-1}) and mean PNF abundance in FR (4.69 × 10^6 cells ml^{-1}) (Table 1). Thus, all variables except for chl *a* showed the lowest mean values at the lowest temperatures (IEZ).

All variables measured at the same stations and depths where grazing rates were determined are shown in Table 2, and Figs. 2 & 3. Gross bacterial heterotrophic production (GBHP) data presented in these figures are different from those shown in Table 1. Here GBHP was calculated as the sum of grazing rates and net production within the incubation bottles in order to obtain values comparable to bacterivory data (see 'Discussion'). Bacterivory was not always detectable. For instance, in the WS area we could only obtain a grazing rate at 2 of the 7 stations visited. The highest total bacterivory rate was found in the BSO-WSC (Stn 13, 4.7 µg C l^{-1} d^{-1}), and the lowest (when detectable) was found in the IEZ (Stn 4, 0.3 µg C l^{-1} d^{-1}). However, specific ingestion rates showed maximum values in the IEZ (Stn 3, 67 bacteria HNF^{-1} h^{-1}), coinciding with the lowest temperature (Table 2). Temperature, chl *a*, microbial biomass, and GBHP for the selected stations followed similar patterns (among zones) to those obtained for the whole data set for each zone (Table 1, Figs. 2 & 3A). Thus, bacterivory obtained in these selected stations should be representative of the different zones considered. Median bacterial and HNF abundance followed a similar pattern to that of temperature, increasing from the IEZ to BSO-WSC zones (Fig. 2A,D,E). Temperature and HNF abundance both showed significant differences among zones (Fig. 2A,E). Chl *a* concentration showed a similar trend to that of ciliate abundance, with a gradual decrease
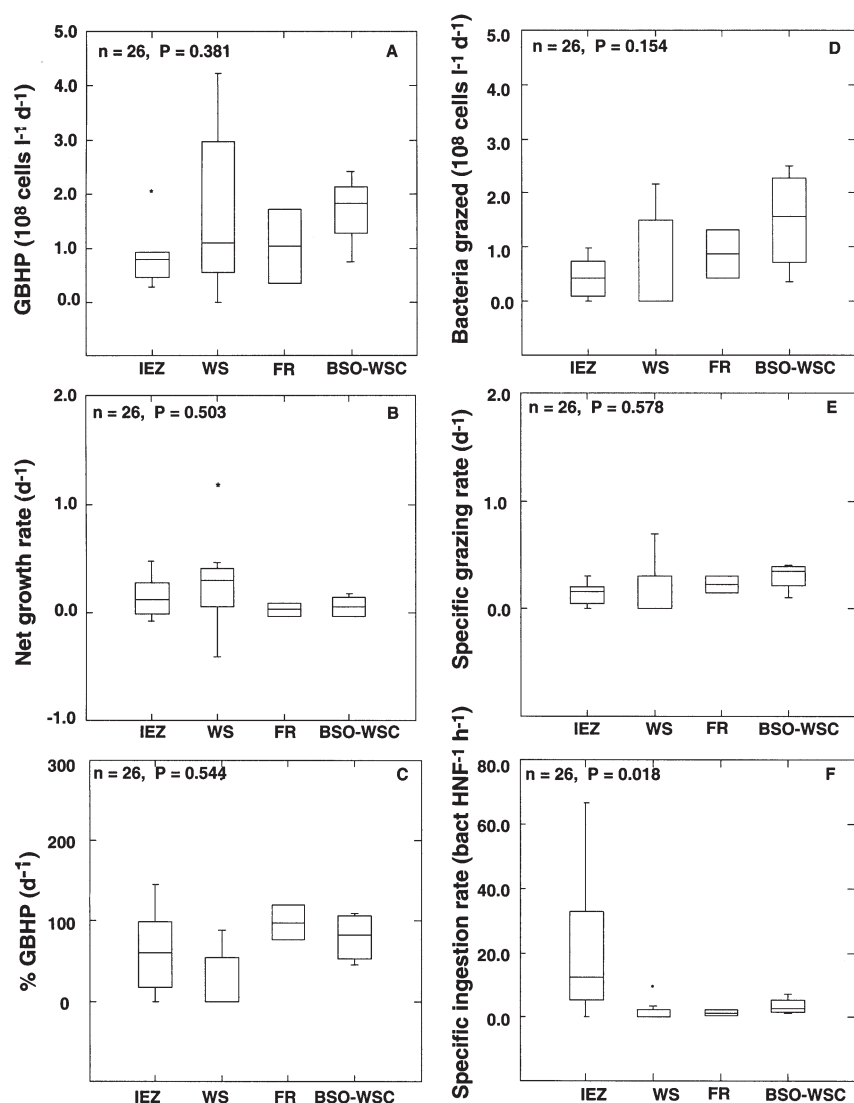
Fig. 3. Box and whisker plots of different variables in each area. (A) Gross bacterial heterotrophic production (GBHP), (B) bacterial net growth rate, (C) percent of grazed bacterial production, (D) total bacteria grazed, (E) specific grazing rate and (F) specific ingestion rate. Explanations for box plots are given in Fig. 2. n: number of data, P: level of significance for the ANOVA test. Data corresponding to selected stations and depth where grazing rates were measured

Regarding heterotrophic microbial activities, we detected that the median GBHP and net growth rate (Fig. 3A,B) showed a slight increase in WS, while total grazing rates showed the highest value in BSO-WSC (Fig. 3D, Table 2). Specific grazing rates showed very similar values in all zones (Fig. 3E), while specific ingestion rates were only high in the IEZ (Fig. 3F). Finally, despite the great variability between stations, bacterivory represented a high proportion of GBHP in the FR (98 ± 22%) and BSO-WSC zones (79 ± 26%), and a lower proportion in the IEZ (62 ± 42%) and in WS (43 ± 79%).

Relationships between variables

Temperature and biological variables

Pearson correlation analysis showed that temperature was significantly correlated with HNF, PNF abundance, total grazing rates (when bacterivory ≠ 0) and GBHP (Table 4). These results indicate that temperature could be important in enhancing heterotrophic microbial activities, and in increasing the abundance and biomass of nanoflagellates (Table 4). On the other hand temperature was not correlated at all with chl *a* concentration, or with bacterial abundance.

Total grazing rates and microbial variables

Taking into account only values of bacterivory ≠ 0, grazing rates on bacteria were significantly correlated with abundance of bacteria, HNF and PNF (Table 4). These results suggest that grazing on bacteria increased when both prey and predators increased. We explored the potential factors responsible for changes in grazing rates by multiple regression analysis. Bacteria and HNF abundance (cells l^{-1}) explained 68% of grazing rate variability. The equation was:

$$\log \text{grazing rates} = 1.80 + 0.55\log(\text{bacterial abundance}) + 0.25\log(\text{HNF abundance})$$
$$(n = 16, R^2 = 0.676; p_{\text{constant}} = 0.30;$$
$$p_{\text{Bact}} = 0.018; p_{\text{HNF}} = 0.004)$$

from the IEZ to FR, and increasing again in the BSO-WSC (Fig. 2B,F). Finally, PNF abundance increased from the IEZ to FR and declined in the BSO-WSC (Fig. 2C). Maximum abundance and biomass (Fig. 2F, Table 2) of ciliates were detected at Stn 7 at the surface, and minimum values of abundance were detected at Stn 13 (70 m). Naked oligotrich ciliates (heterotrophic and mixotrophic) dominated the assemblage. Loricate oligotrichs (tintinnids) were less abundant and not always present. The phototrophic *Mesodinium* spp. was not very abundant when present and its contribution in terms of biomass was low (Table 3).

Table 3. Average abundance and biomass of different groups of ciliates found in each zone (as percent of the total ciliate abundance or biomass). SE given in parentheses; nd: not detectable

Zone (Stns)	Naked oligotrichs		Tintinnids		Others		*Mesodinium*	
	Abundance	Biomass	Abundance	Biomass	Abundance	Biomass	Abundance	Biomass
IEZ	82.02	75.76	6.46	18.26	7.15	5.23	4.36	0.75
(4, 7, 119)	(8.21)	(10.23)	(2.43)	(13.42)	(2.68)	(4.31)	(3.83)	(0.19)
WS	91.76	94.40	1.10	1.78	1.82	1.87	5.32	1.81
(9, 11, 18, 19)	(8.68)	(5.09)	(1.67)	(3.79)	(3.18)	(3.90)	(4.71)	(1.37)
FR	88.22	96.22	nd	nd	10.53	3.68	1.26	0.11
(12, 15, 16)	(14.08)	(5.12)			(14.89)	(5.20)	(1.78)	(0.15)
BSO-WSC	95.28	78.21	1.69	0.51	2.60	21.20	0.42	0.08
(13, 14)	(0.37)	(1.38)	(1.69)	(0.51)	(1.75)	(1.97)	(0.42)	(0.08)

Bacterial abundance and predators (e.g. HNF) accounted for most of the variability in bacterivory, despite the fact that temperature was correlated with total grazing rates.

Transect from the IEZ to BSO-WSC

To illustrate the changes of each variable across the different zones, one transect was carried out from the IEZ to BSO-WSC (~400 km). Temperatures showed the widest range in this transect (–1.7 to 2.4°C). Integrated values of biological variables are shown in Fig. 4. The highest integrated chl *a* concentration and primary production values were observed close to the IEZ (see Berdalet et al. 1997, Figueiras et al. 1998, and our Fig. 4A,C). Lower values were observed in the rest of this transect, at the stations occupied by WS waters and also in BSO-WSC (Stn 13). From Stns 9 to 13 (WS and FR) PNF biomass was the most important compo-

Table 4. Correlation coefficient (R) and level of significance (p) found between biological variables, with temperature and total grazing rates. BACT (log bacterial abundance, cells l^{-1}), HNF and PNF (log heterotrophic and autotrophic nanoflagellate abundance, cells l^{-1}), GBHP (log bacterial heterotrophic production, cells l^{-1} d^{-1}), GRZ (log total grazing rates, cells l^{-1} d^{-1}). Numbers in bold are significant values; n: number of cases. na; not applicable (GBHP cannot be correlated with GRZ because total grazing rates is a component of bacterial production)

Variable	Temp.			GRZ		
	R	p	n	R	p	n
BACT	0.263	0.194	26	**0.618**	**0.011**	16
HNF	**0.560**	**0.004**	24	**0.704**	**0.002**	16
PNF	**0.570**	**0.007**	21	**0.631**	**0.016**	14
GBHP	**0.409**	**0.046**	24	na	na	
GRZ	**0.509**	**0.044**	16	–	–	–

nent of phytoplankton biomass (Fig. 4A). The PNF assemblage was dominated by the free-living nanoflagellate *Phaeocystis* sp. with an average cell volume of around 25 μm^3. Integrated bacterial carbon and HNF biomass followed similar trends. Both increased up to Stn 8 (Fig. 4B), decreased at Stn 9 and increased again at the following stations (Fig. 4B). Bacterial carbon exceeds HNF carbon in the stations influenced by the ice edge. Away from the latter the opposite situation was found (Fig. 4B). Primary production reached its maximum at Stn 119, while BHP was highest at Stn 8, coinciding with the highest bacterial biomass (Fig. 4B,C). BHP (μg C l^{-1} d^{-1}) represented 5.7% (Stn 119) to 107% (Stn 13) of primary production (Fig. 4C). All these results indicate that in the IEZ and surrounding waters phytoplankton activity and biomass were enhanced, while bacterial and HNF biomass and BHP were fairly important at the end of the transect (Stns 12 to 13, FR and BSO-WSC respectively). In addition, bacteria showed a peak in biomass and activity at Stn 8.

Different profiles of bacterial and HNF abundance, grazing rates and GBHP (at 2 depths), for 5 of the stations along this transect are shown in Fig. 5. Bacterial abundance profiles achieved the lowest and highest values at Stns 7 (IEZ) and 8 (WS) respectively, while HNF abundance showed minimum values at Stn 7 (IEZ) and maximum values at Stn 13 (BSO-WSC). Ciliate abundance and biomass at the surface showed decreasing values along the transect (Table 2). Grazing rates on bacteria had the lowest values at Stns 11 (undetectable) and 7. Considering surface values, consumed bacteria increased from Stns 7 to 13, except for Stn 11 (Fig. 5). However, for the DCM, similar grazing rates were recorded for Stns 8, 10 and 13, and again the lowest values were obtained at Stns 7 and 11 (not detected). GBHP and grazing rates were balanced at Stns 7 and 13, and at Stn 10 in the DCM, while GBHP exceeded grazing on bacteria in the DCM of Stn 8 and on the surface at Stn 10 by about 50% and more than 100% at Stn 11 (where grazing was undetectable).

HNF carbon requirement

In several cases we observed that most of the GBHP was cropped by consumers, although a great variability was associated with this estimate. Then, we determined whether the consumed bacterial carbon was enough for HNF carbon requirements to grow.
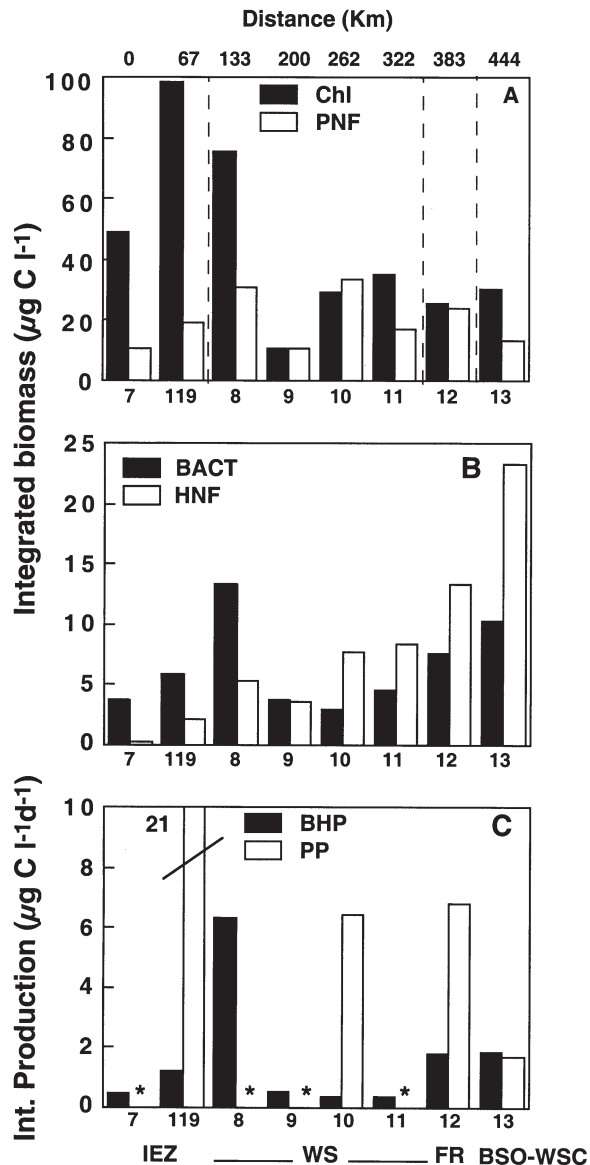

Fig. 4. Integrated parameters at stations from the IEZ to BSO-WSC. (A) Total phytoplankton and phototrophic nano-flagellate biomass, (B) bacteria (BACT) and heterotrophic nanoflagellate biomass (HNF), (C) bacterial heterotrophic production (BHP) and primary production. (∗) Stations where primary production was not determined. Distance in km and station numbers are indicated on the x-axes (above and below respectively)

HNF and bacterial average cell volumes were 16 µm^3 (small HNF around 3 µm in diameter were the most abundant forms) and 0.07 µm^3 respectively. Averages of bacterial and HNF biomass (µg C l^{-1}) in the 4 zones studied revealed that HNF carbon was lower than bacterial carbon in the IEZ, equal in the WS and BSO-WSC and slightly higher in FR (Fig. 6A). When bacterial carbon ingested was compared to the carbon requirements for net HNF growth (0.2 d^{-1}) the consumed bacterial carbon was enough for the needs of HNF growth only in the IEZ (Fig. 6B).

DISCUSSION

Methodological considerations

Bacterivory

The minicell disappearance technique (long-term incubation, 48 h) was used to measure grazing rates on bacteria by protists (Pace et al. 1990). Limitation of methods using fluorescent labeled particles (FLB, latex beads) as tracers have been widely discussed in, e.g., Vaqué et al. (1994), Landry (1994), and Leakey et al. (1996). The major concern arises on comparing our results with those obtained by other authors that used direct uptake of FLB or beads (Table 5). For instance, we did not consider that some grazing could be due to ciliates, or to other bacterial predators present in the water, despite the fact that samples were filtered by 150 µm and some small zooplankton could be present. However, measurements of direct uptake of minicells and total grazing rates were fairly comparable in the Hudson River estuary (Vaqué et al. 1992), where HNF were the most important grazers. Available data of bacterivory in Antarctic waters revealed that HNF had a higher impact than ciliates (i.e. Laybourn-Parry et al. 1996). The most representative group of ciliates found in different Antarctic waters, as well as in other marine systems, are oligotrichous ciliates (Garrison & Buck 1989, Laybourn-Parry et al. 1996, Vaqué et al. 1997), including heterotrophic and mixo-trophic forms. This ciliate assemblage could feed on different kinds of particles (e.g. bacteria, nanoflagel-lates) although they prefer organisms larger than bacteria (Capriulo et al. 1991). Taking into account that only a small proportion of the ciliates counted were strictly bacterivorous, such as small scuticocili-ates (Table 3, included in the 'other' category) as well as small oligotrich ciliates (Šimek et al. 2000), grazing on bacteria should indeed be mainly due to HNF.
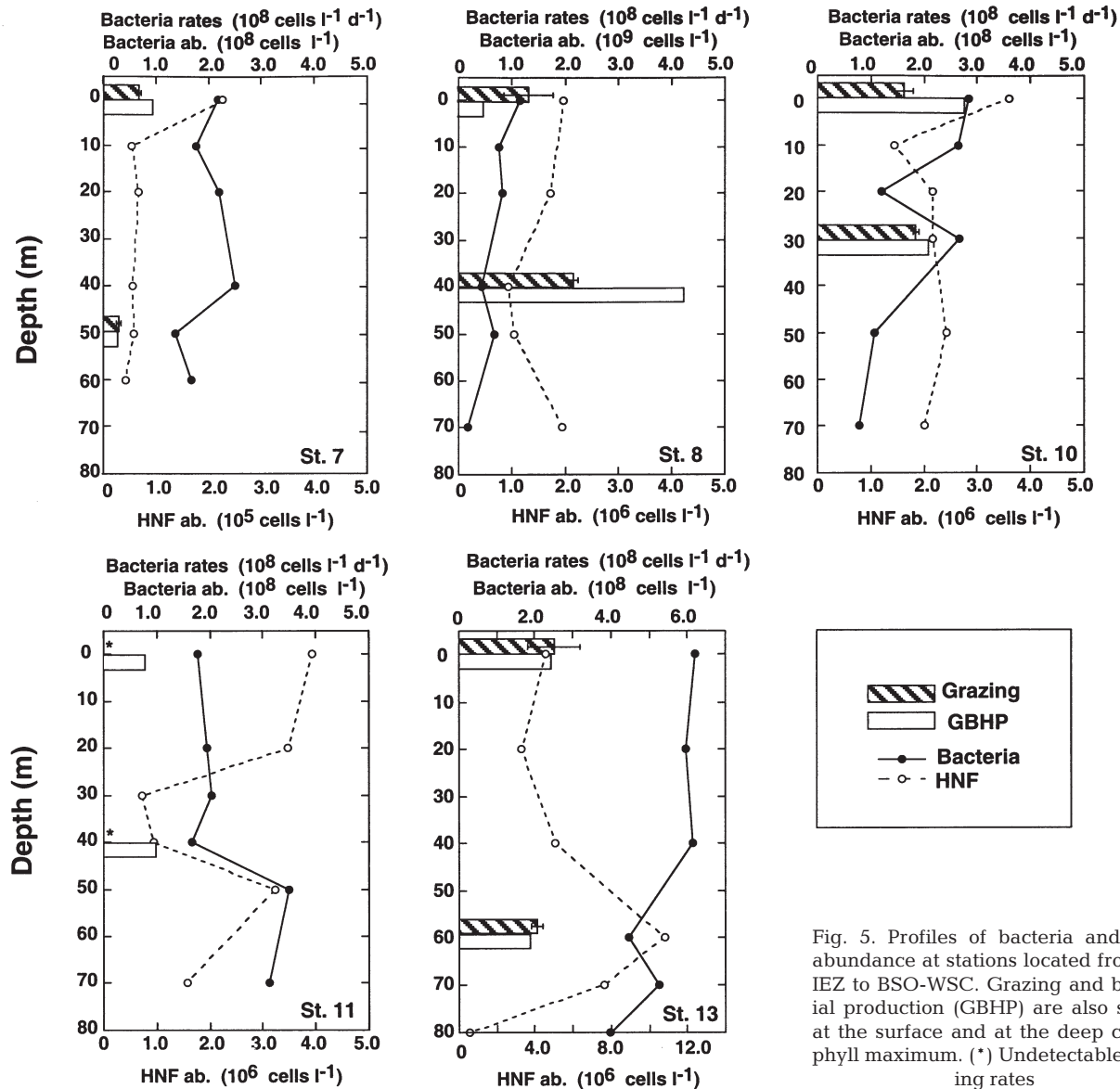
Fig. 5. Profiles of bacteria and HNF abundance at stations located from the IEZ to BSO-WSC. Grazing and bacterial production (GBHP) are also shown at the surface and at the deep chlorophyll maximum. (*) Undetectable grazing rates

Bacterial biomass and production

Archaea have been reported to constitute significant fractions of Antarctic picoplankton assemblages (De Long et al. 1994). These prokaryotes cannot be distinguished from bacteria in DAPI counts; therefore, we use the terms of bacterial abundance, biomass, bacterivory and production for convenience, and these values may well include both bacteria and *Archaea*.

Bacterial production is presented in 2 ways. First, for routine samples we used bacterial production (BHP) obtained by ^{3}H-leucine incorporation (pmol l^{-1} h^{-1}) (Kirchman et al. 1985, Kirchman 1993). Conversion factors used to transform pmol l^{-1} h^{-1} into cells l^{-1} d^{-1} were obtained in the same study zones (Calderón-Paz

1997). These conversion factors were quantitatively similar to the ones estimated empirically by Bjørnsen & Kuparinen (1991) in a very close area. BHP determined by ^{3}H-leucine incorporation showed low values similar to those in the Northwestern Mediterranean Sea (Calderón-Paz 1997, Pedrós-Alió et al. 1999). These values were at the lower end of the range for warmer marine systems (White et al. 1991, Vaqué et al. 1994 and references therein). Second, bacterial production (GBHP) was also estimated in the incubation bottles where grazing rates were determined. Assuming that bacterial losses were only due to predation by protists, GBHP was calculated by adding net production plus total grazing rate throughout the 48 h of incubation. Bacterial production was estimated in this way be-
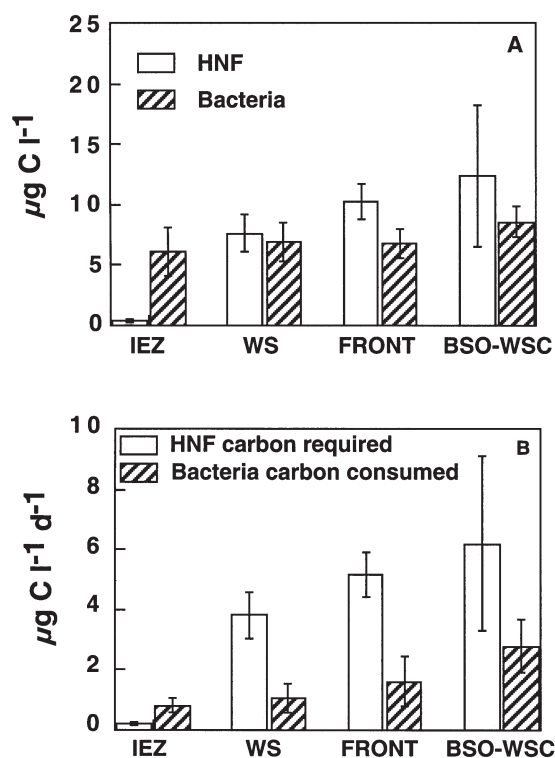
Fig. 6. (A) Comparison of bacterial and HNF biomass in the different zones, and (B) consumed bacterial carbon compared to HNF carbon requirements for growth

cause it is inappropriate to compare the 2 activities (grazing and production) obtained with different incubation times and procedures. While estimates of bacterial production by ^{3}H-leucine are measured within a short time period (~4 h), grazing determinations required much longer incubations in order to be measurable in these waters. For this reason we compared the bacteria consumed and produced within the same incubation bottles. This estimate of bacterial production (GBHP) is an underestimation because calculations are based on the conservative assumption that all losses of bacteria during the incubations were due exclusively to grazing by protists. However, other losses could be important, for example Guixa-Boixereu et al. (2002) found substantial bacterial mortality due to viruses in the Gerlache Strait.

Bacterial production obtained from ^{3}H-leucine incorporation showed systematically lower values than those obtained in the incubation bottles (Table 1, Fig. 3A). Nevertheless, bacterial production obtained by ^{3}H-leucine (BHP) and for 0 to 48 h intervals in the incubation bottles (GBHP) were significantly correlated (Pearson correlation analysis, n = 25, R = 0.433, p < 0.05). Average bacterial doubling times calculated by leucine incorporation ranged from 2.6 ± 0.4 d (BSO-WSC) to 9.4 ± 3.4 d (FR). Doubling times of bacteria in

the bottles used to measure grazing gave similar values, ranging from 3.6 ± 0.8 d (BSO-WSC) to 8.6 ± 5.5 d (FR). Since there was a significant correlation between both bacterial production estimates (BHP vs GBHP), we expected that the proportion of consumed bacteria to GBHP should be similar with the percentage of bacterial losses to BHP in the routine samples.

Patterns of microbial biomass and activity in different study zones

Chl *a* concentration and primary production were maximal near the IEZ (Table 1, Figs. 2B & 5A,C). Marginal ice zones are considered as sites of active phytoplankton growth and accumulation (Alexander & Niebauer 1981, Smith & Nelson 1985). Our values of chl *a* were similar to the ones found by Nelson & Smith (1986) and Perez et al. (1994) close to the ice edge. In contrast, we found low values for abundance of heterotrophic microorganisms (bacteria and HNF) and BHP in this zone, compared to the other study zones (Table 1). Sullivan et al. (1990) found higher heterotrophic microbial activity and biomass in the ice edge surrounding waters up to 200 km northward. These authors also found a significant correlation between bacterioplankton and phytoplankton (activity and biomass) along a transect from the IEZ to the open sea. We found an inverse pattern between these variables (compare Fig. 5 of Sullivan et al. 1990 with Fig. 4 in this study). Discrepancies could be due to the sampling seasons. Their cruises were carried out in late winter and spring when the algal bloom was in the initial stages (Kang et al. 1997) and ours during late summer, when the bloom was probably already declining. Our results indicate segregation in space and time between the development of phytoplankton and bacterial and HNF assemblages (Fig. 4). In fact, we detected an increase in BHP and a slight increase in bacterial and HNF abundance at a distance from the ice edge waters (Stn 8, Fig. 4B,C), while maximum values of primary production were achieved at the preceding station (Stn 119 from IEZ). Similar spatio-temporal variation of phyto- and bacterioplankton biomass across a transect in the Scotia-Weddell Sea area during sea ice retreat in the austral summer was observed in the EPOS expedition (Lancelot et al. 1991)

Total grazing on bacteria (when detectable) increased in parallel with temperature from the ice edge to 400 km north. However, HNF and bacterial abundance accounted for the highest percentage of the variability in grazing rates (68%). We suspect that temperature affects grazing rates indirectly, because the direct influence of temperature would rely on GBHP and HNF growth, which would be translated

Table 5. Range of bacterial and HNF abundance and biomass, and grazing rate data recorded in different Antarctic waters. Temperature values and method used are included. (1: James et al. 1995; 2: Laybourn-Parry et al. 1995; 3: Laybourn-Parry et al. 1996; 4: Putt et al. 1991; 5: Pedrós-Alió et al. 1996; 6: Leakey et al. 1996; 7: Vaqué et al. 2002). Grazing methods used: BEADS, plastic beads; FLB, fluorescent labeled bacteria. Estimated: calculated through models of Vaqué et al. (1994); MINIS: minicells; nd: not detected; –: no data

Location	Season	Bacterial abundance ($\times 10^8$ l^{-1})	Bacterial biomass (μg C l^{-1})	HNF abundance ($\times 10^6$ l^{-1})	HNF biomass (μg C l^{-1})	Ingestion (bacteria) (HNF^{-1} h^{-1})	Bacterial biomass grazed (% d^{-1})	Bacterial production grazed (% d^{-1})	Temp. (°C)	Grazing method
Melt Ponds[1] (McMurdo ice shelf)	Dec 91–Jan 92	0.3–3.4	–	–	–	–	<5.0–13.6[a]	nd	5	BEADS
Crooked lake[2] (Ultra-oligotrophic Antarctic Lake)	Dec 92–Nov 93	1.2–4.5	2.0–10.0	0–0.5	0–2.4	0.2	–	0.1–9.7	2–4	FLB
Heywood lake[3]	Dec 94–Jan 95	<10.0–80.0	5–>400	1.0–17.0	<5.0–>70	0.8	–	6	2	FLB
Sombre lake (Maritime Antarctic lake)	Dec 94–Jan 95	<10.0–31.8	2–>100	0.1–7.0	5.0–8.0	0.5	–	100		FLB
Ice edge[4] (McMurdo Sound)	Nov 90–Jan 91	1.0–7.0	–	1.0–12.0	–	0.01–2.4	0.5–30.0	–	–	BEADS
Drake Passage[5]	Jan 94	0.5–7.1	2.7–5.8	0.04–0.4	1.6–2.7	–	0.5–1.1	5.0–17.8	–1.0–6.0	Estimated
Prydz Bay[6]	Dec 93–Feb 94	2.1–8.3	13.0–64.0	1.6–4.2	7.5–16.0	0.06–8.3	2.8–12.2	10.0–36.0	–1.4–(–0.4)	FLB
Antarctic Peninsula[7]										
Bellingshausen waters	Dec 95–Feb 96	3.4–3.5	5.5–5.6	0.3–2.2	0.2–1.7	1.8–6.4	11.0–26.0	35–100	–0.06–2.40	MINIS
Bransfield strait	Dec 95	3.6–6.9	5.8–11.1	0.5–0.9	0.4–0.5	0.1–7.8	0.7–14.0	0.5–174	0.00–0.17	MINIS
Gerlache strait	Dec 95–Feb 96	2.5–7.2	4.0–11.6	0.4–2.1	0.2–1.7	4.7–13.2	2.3–26.0	38–80	–1.19–0.48	MINIS
This study										
Ice edge	Jan 94	0.7–10.0	1.4–18.7	0–1.3	0–4.7	nd–66.9	nd–30.7	nd–>120.0	–1.7–(–0.09)	MINIS
Weddell Sea	Jan 94	0.7–11.5	1.3–21.5	0.3–5.6	1.0–19.8	nd–9.6	nd–69.1	nd–>120.0	–0.5–0.7	MINIS
Front	Jan 94	1.5–7.6	2.8–14.2	0.8–9.7	2.7–34.2	0.5–2.2	14.3–30.6	76.3–119.6	–1.1–1.7	MINIS
BSO-WSC	Jan 94	1.4–6.2	2.6–11.6	0.2–11.8	0.7–41.7	1.1–7.5	10.2–40.3	46.1–108.9	0.04–2.4	MINIS

[a]Grazing on bacteria due to ciliates

into an increase in bacterial and HNF abundance, and as a result total bacterivory would also be enhanced (Table 4). Several experiments carried out on different cruises, in which samples were incubated at different temperatures, showed that bacterial production, HNF and bacterial abundance and, consequently, bacterivory, increased after incubating at higher temperatures (Vaqué unpubl.). This dependency of bacterial heterotrophic activity on temperature has been demonstrated by the significant relationship between bacterial production and temperature found across systems (White et al. 1991, Vaqué et al. 1994). In addition, Choi & Peters (1992) showed that HNF isolated from cold systems increased their growth and grazing rates when the temperature was increased.

Assuming that bacteria were mainly consumed by HNF, each single HNF from 3 of the 4 zones (WS, FR, BSO-WSC) ingested from undetectable to 9.6 bacteria h^{-1}. However, almost all stations located in the IEZ showed the highest values recorded in Antarctic waters (11 to 67 HNF bacteria h^{-1}), only comparable to those obtained in Resolute Passage in the Artic under an ice layer (Laurion et al. 1995). Thus, specific ingestion values in this study (except for the IEZ) are at the low end of temperate aquatic systems, but higher than the values found in other Antarctic waters (Table 5). This disagreement could be due to the way this specific ingestion rate was calculated. Thus, several authors (e.g. Leakey et al. 1996, Bird & Karl 1999) measured the FLB directly inside the digestive vacuoles of HNF, while our rates were estimated by dividing total grazing rates by the total number of HNF. We could not discriminate between phototrophic and mixotrophic nanoflagellates and they

could be partially responsible for bacterial grazing rates (Berninger et al. 1992). Although HNF are presumably the main consumers of bacteria, we cannot rule out the possibility that other microorganisms than HNF (e.g. mixotrophic nanoflagellate ciliates, heterotrophic dinoflagellates, naupliae) could also graze directly on bacteria.

Bacterivory versus bacterial biomass and production. HNF carbon requirement

Consumed bacteria, as a percent of bacterial biomass, varied from undetectable to 69% daily (average: 17 ± 19%). As a percent of GBHP consumed bacteria ranged from undetectable to >100% daily (average: 67 ± 13%). These values were at the upper end of the range obtained in other Antarctic waters (Table 5). We observed considerable variability among stations (Table 2). For instance, at Stn 10 (WS) protists consumed 69% of bacterial biomass, and at Stn 8 at the surface (WS) >100% of GBHP. In 5 WS stations and 1 IEZ station, on the other hand, grazing could not be detected. Hence, bacterivory cannot be considered the main factor controlling bacterial production for the whole study area (Table 5). Ducklow et al. (2001) showed a strong bacterial response to the bloom in the Ross Sea. BHP was relatively low, so the implication is that removal rates failed to balance the growth. Anderson & Rivkin (2001) showed that the timing and magnitude of the phytoplankton bloom and the duration of the post bloom period exert a significant influence on the flux of bacterioplankton carbon through protozoans. For instance, they found that grazing losses of bacteria were negligible immediately before and after the phytoplankton bloom. During these periods bacteria would probably not be controlled by bacterivory and the protists present could predominantly be herbivorous. This observation would be in agreement with our results obtained in summer, a situation that can be considered post-bloom. The fact that grazed bacterial carbon did not meet HNF carbon demand (except in the IEZ zone) would be a confirmation that HNF assemblages need to graze on something other than bacteria. For instance, HNF could feed on small algae (Goldman & Caron 1985), or take up DOM of high molecular weight, as was observed in Antarctic waters by Marchant & Scott (1993). Estimates of HNF carbon demand were obtained under conditions that in principle should favor a good balance between bacterial carbon consumption and HNF carbon requirement. First, the bacterial biomass was relatively high compared to that of HNF. On the one hand, HNF showed an average cell volume of 16 µm³, which is lower than the values obtained in the Drake Passage (65.4 µm³,

Pedrós-Alió et al. 1996) and in the Western Weddell Sea (30 µm³, Kivi & Kuosa 1994). This cell volume was within the range found in the coastal sea ice in the vicinity of Davis Stn (15 to 159 µm³, Archer et al. 1996) and in coastal waters of Prydz Bay (3.3 to 34.9 µm³, Leakey et al. 1996). On the other hand, bacterial cell volume averaged 0.07 µm³. It was higher than that obtained in the Drake Passage (Pedrós-Alió et al. 1996) and within the range of values recorded by Putt et al. (1990) in McMurdo Sound and in ultra-oligotrophic Maritime Antarctic lakes (Laybourn-Parry et al. 1995, 1996). Second, HNF carbon requirements were probably underestimated, because they were based on HNF net growth rate without taking into account HNF mortality. Third, we assumed that the only bacterivores were HNF, but a portion of the bacteria grazed could go to other predators. These 3 considerations would favor a relatively high bacterial carbon to HNF requirement ratio. Despite all these considerations, only in the IEZ did grazing fulfill HNF requirements (Fig. 6B).

The ECOANTAR-94 cruise provided an opportunity to explore the variability of microbial biomass and activity in a complex area characterized by different hydrographic regimes. We studied the relevance of bacterivory as the main factor in controlling bacterial production and biomass as well as being the only carbon source for HNF growth. Despite the high variability within each zone, an increase in HNF and bacterial activity (grazing and production) from the coldest (IEZ) to warmest zones (WS, FR, BSO-WSC) was detected, while phytoplankton (production and biomass) decreased. Indeed, temperature was significantly correlated with HNF abundance and bacterial grazing and production. However, the main factors that explained most of the variability in bacterivory (68%) were HNF and bacterial abundance. Our results indicated that bacterial losses due to protists represented a substantial proportion of bacterial production (average 67% d⁻¹). However, the variability detected within areas was rather large (from undetectable to >100%). Therefore, bacterivory was not the main factor controlling bacterial production in all zones considered. This leaves ample room for alternative factors such as lysis by viruses.

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