PLANKTON STRUCTURE a in SHALLOW COASTAL ZONE AT ADMIRALTY BAY, KING GEORGE ISLAND, WEST ANTARCTIC PENINSULA (WAP): PICO, NANO AND MICROPLANKTON AND CHLOROPHYLL BIOMASS

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Denise Rivera Tenenbaum^{1,*}, José Juan Barrera-Alba^{1,**}, Renatha Barboza Duarte¹, Márcio Murilo Barboza Tenório^{1,***}

1 Laboratório de Fitoplâncton Marinho, Instituto de Biologia, Universidade Federal do Rio de Janeiro – UFRJ, Rio de Janeiro, RJ, Brazil *e-mail: *deniser@biologia.ufrj.br; **juanalba@biologia.ufrj.br; ***marcio.tenorio@biologia.ufrj.br*

Abstract: The phytoplankton composition and biomass are being monitored in Admiralty Bay, Antarctic Peninsula since 2002 to detect possible interannual changes on a long-term perspective. In this report, we present the preliminary results of the 2009/2010 monitoring program regarding phytoplankton size-structure and biomass. Even if mean microplankton densities were similar between December 2009 and February 2010, diferent phytoplankton groups dominated each sampling period. Pennate diatoms showed highest contribution in December, whereas athecate dinoflagellates were the most abundant microplanktonic group in February. Pico and nanoplankton were only detailed during the second sampling period, and results showed that phytoplankton were dominated by cells <10 μ m (~10⁴ and 10⁷ cells.L⁻¹, respectively). The shift in phytoplankton structure pointed out by the *dominance of pico- and nano-size cells in phytoplankton and heterotrophic dinoflagellates in late summer must be confirmed by continuing the long-term monitoring program and the implementation of microvariation sampling effort to identify the factors that are actually influencing phytoplankton populations in this environment.*

Keywords: microbial community, size-fraction structure, Antarctic coastal zone, PROANTAR

Introduction

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The West Antarctic Peninsula (WAP) waters undergo extreme seasonal fluctuations in terms of light regime, sea-ice concentration and productivity (Delille, 2004). The WAP has experienced a significant rise in air temperature during the last 50 years (±0.56 °C per decade) (Marshall *et al*., 2002). Monitoring of biodiversity in shallow waters (30 m) at Admiralty Bay was implemented in 2002 by PROANTAR (Brazilian Antarctic Program) during the PROANTAR Operation XX (OPERANTAR) aiming to study the effects of environmental factors (natural and anthropogenic) on the microplanktonic community structure, through analysis of long-term temporal series. These activities were

undertaken until 2010, through four surveys, including samplings in both early and late austral summer. Recent studies showed that in the Admiralty Bay, picoplankton and nanoplankton are the dominant groups, with microplankton diatoms as the second group in abundance. Between the decades of 1990 and 2000, several studies showed a decline in diatom contribution (Kopczynska, 2008), in relation to those observed in the continental shelf region. Based on these facts, since 2009 new approaches to phytoplankton monitoring have been established, including the analysis of size-fractioned pigments by spectrofluorometry, and the analysis of density and biovolume of pico- and nanoplankton

by epifluorescence microscopy, and furthermore through a higher sampling frequency effort. Additionally, the composition of microphytobenthos species will be carried out to study the effects of environmental changes on this community in the nearshore Antarctic ecosystem.

In the present study we show preliminary results during the OPERANTAR XXVIII, between December 2009 and February 2010.

Materials and methods

Study area

Admiralty Bay (62° 03'-12' S and 58° 18'-38' W), located at King George Island, is a deep fjord-like embayment with 500 m maximum depth at its centre (Rakusa-Suszczewski *et al*., 1993). The waters from the bay mix with the deep oceanic waters from Bellingshausen and Weddell Seas at its southern opening, which connects to the Bransfield Strait (Rakusa-Suszczewski, 1980; Lipski, 1987). The maximum depth varies between 60 m along the shores and 500 m in the centre of the bay. Deep currents generated by tides, frequent upwellings, vertical mixing of the entire water column and current velocities of 30-100 cm.s−1 in the 0-100 m surface stratum are characteristic of the bay (Rakusa−Suszczewski, 1993). In the context of water column production, Admiralty Bay at nearshore can be considered as "high nutrient – low chlorophyll" (HNLC) Platt *et al*. (2003) showing high inorganic dissolved nitrogen (16.6-46.9 µM) and phosphate (0.2-9.9 µM) concentrations, while chlorophyll levels are lower than 1.7 µg.L–1 (Lange *et al*., 2007).

Sampling

The analysis of microplankton and chlorophyll was performed from aliquots of the 5 L water samples collected with a Van Dorn bottle from surface, middle water column and near the bottom (≈ 30 m) at five stations in December 2009 and in February 2010. The fractionation analysis of pico- and nanoplankton was performed only at three stations (AR, MP and CF) in February 2010, when three surveys were done. The Admiralty Bay location and the position of the sampling stations are shown in Figure 1.

At the same time, temperature and salinity measurements were carried out by the Laboratório de Química Orgânica Marinha (LabQOM), Instituto Oceanográfico da Universidade de São Paulo (The Marine Organic Chemistry Laboratory of the Oceanographic Institute of the University of São Paulo).

Fixation and preparation of samples

For microplankton ($>$ 20 μ m), 1 L aliquots were fixed with buffered formaldehyde (2% f.c.). In the laboratory, samples were analysed using the settling technique (Utermöhl, 1958) in an Olympus IX70® inverted microscope at 400x magnification.

For pico- $\left($ <2 μ m) and nanoplankton $\left($ <20 μ m), aliquots of 250 mL were stored in dark bottles and fixed with 0.22 µm filtered glutaraldehyde (2% f.c.) at 4 °C until analysis. 5 and 30 mL were stained with DAPI (4',6-diamidino-2-phenylindole) at a final concentration of $0.01 \mu g.L^{-1}$ (Martinussen & Thingstad, 1991), during 10 minutes and filtered respectively by 0.22 µm (picoplankton) and 1.0 µm (nanoplankton) polycarbonate black membrane filters (Poretics®), and mounted on microscope slides between layers of immersion oil. Slides were stored at –20 °C. Analyses were performed using an Olympus BX51® epifluorescence microscope with 1,000 x magnification. The number of heterotrophs was calculated based on the total counted using DAPI (UV filter combination) minus the number of autotrophs analysed by autofluorescence (Blue filter combination).

For chlorophyll biomass, 2 L aliquots were filtered through Whatman® GF/F filters for pigments analyses, while 0.8-2 L was used for the size structure study. In the latter case, water sampled at 3 depths was fractionated by serial filtration on 10 μm and 2 μm polycarbonate filters and GF/F. The filters were folded, placed into a 1.2 mL cryotube and immediately quick-frozen in liquid nitrogen (−196 °C) and stored at −80 °C. Concentrations of chlorophyll *a* and phaeophytin *a* were assessed using a modified version of Neveux and Lantoine's (1993) method.

In order to normalize distributions and eliminate zero values, the biological data was transformed using $log10(x + 1)$. Differences among surveys and sampling

Figure 1. Study area (modified from Moura, 2009) with the position of the sampling sites: Ferraz Station (CF), Botany Point (BP), Machu Picchu (MP), Point Thomas (PT), Arctowski (AR).

stations were tested by a One-Way ANOVA with a Kruskal-Wallis test ($p < 0.05$). Spearman's correlation factor was also calculated.

Results

Microplankton and total chlorophyll biomass between early and late summer

Although salinity showed little variation between sampling periods, values were on average lower in February 2010 (33.9 \pm 0.2) than in December 2009 (34.2 \pm 0.1). During the early summer, the water was relatively colder $(-0.13 \pm 0.11 \degree C)$ than during late summer $(0.68 \pm 0.25 \degree C)$. Although no great differences in salinity and temperature between sampling stations during each period, on Machu Picchu (MP) the lowest salinity and temperature were observed in December 2009, while at EACF the lowest values for both variables were registered in January 2010 (Figures 2a, b). Total chlorophyll biomass was on average

higher in early summer $(0.34 \mu g.L^{-1})$ than in late summer $(0.20 \mu g.L^{-1})$, no significant differences were observed among sampling stations inside each sampling period (Figures 2c, d).

An average cellular density of $3 \times 10^3 \pm 0.3 \times 10^3$ cells. L⁻¹ was observed for microplankton, with little variation between sampling periods ($\approx 10^3$ cells. L⁻¹). The contribution is shared by the diatoms (mainly at the beginning of summer with 56%) and dinoflagellates (at the end of the summer with 68%). Among diatoms the pennate type was predominant (90% in December 2009 and 70% in February 2010). Athecate forms, especially heterotrophs, were more abundant among dinoflagellates during February (69%), while thecate forms representing ~70% of total dinoflagellates in December. Among sampling sites, microplankton registered maximum cellular density at MP due to the predominance of pennate diatoms during December 2009 (Figure 2e).

Sampling station

Figure 2. Results of salinity (continuous line), temperature (dotted line), chlorophyll *a* concentration (µg.L⁻¹) and microplankton density (cells.L⁻¹) at December 2009 (a, b and c) and February 2010 (d, e and f).

Pico and Nanoplankton abundance and size‑fractioned chlorophyll in late summer

During February 2009 pico- and nanoplankton densitiy did not show significant differences among sampling sites, but differences were observed among sampling periods (*p* < 0.01). Chl*a* concentrations varying between 0.18 and 0.74 μ g.L⁻¹ were observed, with the size-fraction <10 μ m representing more than 80% of the total. An increase in Chl*a* concentrations was observed from the first to the last survey, although the fraction 2-10 µm registered higher values during the second survey (Figure 3b). Nanoplankton and picoplankton abundances showed a similar pattern, with higher mean values (8.5 \pm 2.8 \times 10⁶ and $1.2 \pm 0.2 \times 10^{11}$ cells. L⁻¹, respectively) during the second survey (Figures 3c, d). The nanoplankton community was dominated by the autotrophic (>75%) and 2-10 μ m

size-fraction, while the picoplankton was dominated by heterotrophs (~99%). Despite autotrophic cells represented only <1% of picoplankton, abundances relatively high for this group were observed $(2.1 - 8.2 \times 10^7 \text{ cells.} \text{L}^{-1})$. Autotrophic nanoplankton was positively correlated with total and <10 µm size-fraction of Chl*a* (*p* < 0.01), although autotrophic picoplankton was negatively correlated with total and <0.2 µm size-fraction of Chl*a* (*p* < 0.05).

Discussion

Microplankton and total chlorophyll biomass between early and late summer

Microplankton cellular densities and chlorophyll biomass observed in this study were low when compared to those registered for Admiralty Bay during the decades of the

Figure 3. Results at the different sampling periods during February 2010: a) salinity (continuous line) and temperature (dotted line); b) size-fractioned chlorophyll a concentration (µg.L⁻¹); c) picoplankton density (autotrophs in 10⁷ L⁻¹ cells; heterotrophs in 10⁹ cells.L⁻¹); d) nanoplankton density (10⁶ cells.L⁻¹).

1970s, 1980s and 1990s, when densities of 105 cells.L–1 were usually registered (i.e. Kopczynska, 1981; Brandini, 1993; Kopczynska, 2008). However densities were similar to those observed by Lange *et al*. (2007) in a study developed during the austral summer 2002/2003.

Dominance of diatoms over dinoflagellates in the microplankton fraction has been usually observed for Admiralty Bay (Lange et al., 2007; Kopczynska, 2008), but a diminished percentage of contribution of diatoms in the phytoplankton assemblages was observed in a study developed in 2003-2005 (Kopczynska, 2008). Our results showed that the contribution of diatoms decrease especially during late summer, while at the same time heterotrophic dinoflagellates (i.e. *Gyrodinium lachryma*) became more abundant.

Pico and Nanoplankton abundance and size‑fractioned chlorophyll in late summer

Phytoplankton community at Admiralty Bay during the late summer of 2009/2010 was dominated by pico- and nanoplankton, both in abundance and chlorophyll biomass. In previous studies the dominance of nanoflagellates and monads for this region had been observed (i.e. Kopczynska, 1980; Kopczynska, 1981; Brandini, 1993; Kopczynska, 2008). Maxima of flagellates at Admiralty Bay in windless days and little variation in atmospheric pressure was reported, which resulted in an increase of water column stability (Kopczynska, 1981). Although Kopczynska (2008) showed the co-dominance of picoplankters from inverted microscope cell counting technique at Admiralty Bay, this was the first attempt to quantify the real contribution of picoautotrophs to total phytoplankton density and biomass, and densities were in the same range of those observed in other Antarctic regions (i.e. Umani *et al*., 2005; Delille *et al*., 2007). In the nearshore coastal waters along the Antarctic Peninsula, a recurrent shift in phytoplankton community structure, from diatoms to cryptophytes, has been documented due to high temperatures along the Peninsula increasing the extent of coastal melt-water zones promoting

seasonal prevalence of cryptophytes (Moline *et al*., 2004). The dominance of pico and nano-size cells in phytoplankton, which are not grazed efficiently by Antarctic krill, will likely cause a shift in the spatial distribution of krill and may allow also for the rapid asexual proliferation of carbon poor gelatinous zooplankton, salps in particular (Moline *et al*., 2004), and probably the dominance of heterotrophic dinoflagellates observed during the late summer period of this study.

Conclusion

In the context of the regional warming trend of WAP, preliminary results of the present study showed a shift in Admiralty Bay plankton community, with significant variability both in short- and medium-term scales, from day to day and months. Low microplankton densities, dominance of dinoflagellates, mainly heterotrophs, and high contribution of autotrophs pico- and nanoplankton to total density and biomass in late summer, suggest that changes could be occurring in Admiralty Bay food web. Thus, it is necessary to continue the long-term monitoring program and the implementation of microvariation sampling effort to identify the factors that are actually influencing phytoplankton populations in this environment.

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