Vol. 573: 61–72, 2017
https://doi.org/10.3354/meps12162

MARINE ECOLOGY PROGRESS SERIES
Mar Ecol Prog Ser

Published June 21

Influence of mixed upwelled waters on metabolic balance in a subtropical coastal ecosystem: São Sebastião Channel, southern Brazil

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ABSTRACT: Planktonic gross primary production (GPP), community respiration (CR) and net community production (NCP) were evaluated for the first time in the central part of the Southern Brazilian Bight (SBB), in the São Sebastião Channel (SSC); and their response to the South Atlantic Coastal Water (SACW) intrusion was investigated. Sampling experiments took place during late spring 2015 and summer 2016 at a fixed station in the southern portion of the SSC. Although both chlorophyll *a* and nutrient concentrations were characteristic of oligotrophic and low productive ecosystems, all volumetric planktonic metabolism experiments showed high net and gross primary productions rates. Depth-integrated NCP and GPP values had a global mean ±SE of 31 ± 10.5 and 106.7 ± 11.5 mmol O_2 m^{-2} d^{-1}, respectively. The communities tended to be net heterotrophic (i.e. GPP/CR < 1) at volumetric GPP values less than 7.10 mmol O_2 m^{-3} d^{-1}. The intrusion of nutrient-rich and -mixed water (partly SACW) observed during the summer experiments had a strong influence on autotrophic planktonic communities, enhancing their NCP rates about 3-fold. Nutrient supply by terrestrial inputs and by upwelled SACW intrusion in the SSC enables high primary production to be sustained with a dominance of autotrophic communities.

KEY WORDS: Plankton · Metabolism · Southern Brazilian Bight · SACW

INTRODUCTION

Despite covering only 7% of the surface of the global ocean, coastal ecosystems play essential roles in the biogeochemical cycles of marine environments. Indeed, they receive considerable inputs of organic matter (e.g. Carder et al. 1989, Siegel et al. 2002) and nutrients through runoff and groundwater discharge (e.g. Cloern 2001), and because of that show generally substantial biological activities when compared to the open ocean. Furthermore, coastal marine ecosystems are greatly threatened by anthropogenic activities and climate change impacts, both expected to increase in the following years (IPCC 2007). One of the consequences of global warming for continental shelves and adjacent coastal areas is that the wind fields are altered (Harley et al. 2006), which in turn may modify both the frequency and intensity of coastal upwelling events (Bakun 1990). Coastal upwelling events have been described as a relevant factor controlling phytoplankton primary production and phytoplankton biomass (Daneri et al. 2000, Moncoiffé et al.

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2000, Arístegui et al. 2004, Arbones et al. 2008). The episodic nutrient inputs driven by coastal upwelling have been reported to be positively correlated with planktonic community respiration (CR) (Arbones et al. 2008, but see Moncoiffé et al. 2000) and might play an essential role regulating the metabolic balance. The metabolic balance of planktonic communities refers to the balance between gross primary production (GPP) and community respiration (CR), defining whether plankton communities act as net CO_2 sources (CR > GPP) or sinks (CR < GPP) in a given ecosystem. Thus, considering the importance of coastal ecosystems in biogeochemical cycles, their complexities and the effect of climate on their planktonic communities, it is essential to extend our knowledge on these ecosystems.

The southeastern Brazilian Bight (SBB) contains a relatively wide continental shelf characterized by seasonal advance and retreat of warm and salty surface Tropical Water (TW; temperature >20°C and salinity >36) and colder and more nutrient-rich subsurface South Atlantic Central Water (SACW; temperature <18°C and salinity between 35 and 36), both present in the Brazil Current (Emilsson 1961, Cerda & Castro 2014). Intermittent coastal upwelling events transporting SACW nearshore occur from the north (off Cabo Frio) to the south of the SBB (Santos), and tend to be seasonal (Emilsson 1961, Matsuura et al. 1980, Castro 2014). Mainly during spring and summer, the prevailing northeasterly winds displace surface waters offshore and promote deep SACW intrusion over the continental shelf, while during winter the frequent passages of atmospheric cold front systems favor downwelling winds (Castro et al. 1987, Castro 1996).

The São Sebastião Channel (SSC) is located in the central part of the SBB, between the continent and São Sebastião Island (Fig. 1), and is dominated yearround by Coastal Water (CW) with variable thermohaline characteristics (Peres 2013). During spring and summer, SACW intrusion has been detected in the deeper layers of the South entrance of the SSC (Braga & Müller 1998, Castro & Miranda 1998, Silva et al. 2005). This coastal ecosystem is considered to be meso-oligotrophic, but with enhanced phytoplankton biomass during SACW intrusions (summer) (Saldanha 1993). The SSC is a natural barrier to the open ocean, with specific hydrodynamic properties, and is vulnerable to human activities (i.e. the presence of an important oil harbor and marine outfall). While this coastal ecosystem has been mainly studied for its hydrodynamic properties (Castro 1990, Coelho 1997, Silva et al. 2005, Dottori & Castro 2009, Dottori et al. 2015) and its benthic biodiversity (see Amaral et al. 2010), very little information exists about the pelagic plankton communities of this ecosystem. Indeed, to date, only 4 studies have evaluated the planktonic biomass and its distribution along the SSC (Gaeta et al. 1990, Gianesella-Galvão et al. 1997, Gianesella et al. 1999, Peres 2013), and only one estimated the particulate primary production of the planktonic community (Gaeta et al. 1990). Recently, Giannini & Ciotti (2016) examined the relationships between phytoplankton chlorophyll size fractions and photo-physiological parameters in the SSC.

Here, we evaluate for the first time the metabolic balance of planktonic communities (GPP, CR and net community production [NCP]) in the central portion of the SSC and investigate its response to SACW intrusion. In accordance with previous planktonic biomass and primary production studies in this region (Gaeta et al. 1990, Gianesella-Galvão et al. 1997, Gianesella et al. 1999, Peres 2013), we expected to observe relatively low metabolic rates and predominance of heterotrophic communities when SACW intrusion was absent and higher metabolic rates shifting to predominance of autotrophic communities during SACW intrusion. We examined this over sev-

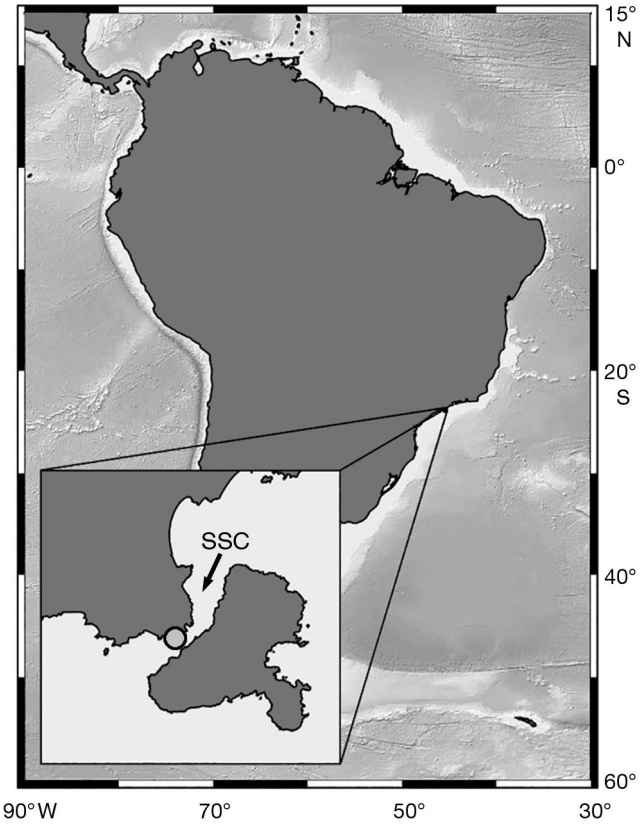

Fig. 1. Location of the studied station (grey circle, inset) in the São Sebastião Channel (SSC), southeast Brazil

eral sampling periods during late spring 2015 (October, November and December) and summer 2016 (January, February and March) at a fixed station in the central portion of the SSC.

MATERIALS AND METHODS

Experimental set-up

The sampling was carried out at one of the sites of the Brazilian Coastal Monitoring System (SiMCosta) Project (www.simcosta.furg.br) where an oceanographic buoy has been moored at the 15 m isobath (23° 49′ 50″ S and 45° 25′ 19″ W; Fig. 1). Water samples were collected at this fixed station early in the morning (08:00 h, local time) using a 5 l Go-FLO sampling bottle at 3 fixed depths: 1, 7 and 12 m. Sampling design with fixed depths was established due to difficulties in sampling and incubating at specified irradiance levels. During late spring 2015 and summer 2016, water was sampled on 2 days in October (14/10, 20/10), 7 days in November (09–11/11, 16–19/11), 1 day in December (08/12), 8 days in January (18–21/01, 25–28/01), 4 days in February (22–25/02) and 1 day in March (01/03). The seawater was carefully transferred to 5 l carboys, using a silicon tube, for subsequent subsampling and analysis of GPP, CR, NCP, bacterial abundance (BA), chlorophyll *a* (chl *a*) concentration, inorganic nutrient concentrations and coloured dissolved organic matter (CDOM), as a proxy for dissolved organic matter. During March, sample volume was sufficient only for planktonic metabolism, and thus environmental parameters were not determined.

Metabolism rates

Community metabolism (GPP, CR and NCP; mmol O_2 m^{-3} d^{-1}) was estimated from changes in oxygen concentration over 24 h in water samples containing the communities sampled at the 3 fixed depths. Dissolved oxygen concentration was measured by Winkler titration, allowing 0.1% precision in oxygen determination using a potentiometric electrode and automated endpoint detection (Oudot et al. 1988) performed by a Metrohm Titrino plus 848. Six 100 ml opaque 'dark' and 6 transparent 'light' borosilicate glass bottles were carefully filled with water from each sampled depth. The water samples were protected by a dark screen to avoid exposure to solar irradiance before the onset of the incubation. For each depth, 5 replicates were immediately fixed for the measurement of the initial oxygen concentration. The 6 'dark' and the 6 'light' bottles of each depth were incubated *in situ* for 24 h at their corresponding depths. CR and NCP were calculated from changes in dissolved oxygen concentration after incubation of samples under 'dark' and 'light' conditions, respectively, and GPP was calculated by solving the mass balance equation GPP = NCP + CR. The integrated metabolism rates were calculated by the trapezoid method, from 1 to 12 m depth. Occasionally, metabolism experiments failed, and yielded 'negative' planktonic CR (i.e. oxygen concentrations measured in dark bottles at the end of the incubation were higher than the initial values) or 'negative' planktonic GPP (i.e. CR > NCP). These estimates were not considered here and a total of 5 of 75 samples were excluded. Therefore, the number of experiments where GPP, NCP and CR rates have been presented here is not equal.

Chlorophyll *a* concentration

Sub-samples of 250 or 500 ml of seawater were filtered for the quantification of chl *a* (mg m^{-3}) onto GF/F filters. The pigment was extracted in 5 ml 90% acetone + DMSO (dimethyl sulfoxide) solution (6:4; Shoaf & Lium 1976) for at least 24 h in the dark and at −20°C, and quantified using a calibrated Turner Trilogy fluorimeter, through the non-acidification method (Welschmeyer 1994). A calibration factor obtained from a commercial chl *a* standard (Sigma-Aldrich) was used to calculate the final chl *a*.

Bacterial abundance

Total BA counts (cell ml^{-1}) were performed by flow cytometry, following Tarran & Bruun (2015). Triplicate subsamples of 2 ml for each depth were drawn from the carboys, filtered in a 20 µm mesh and fixed with microscopy grade glutaraldehyde (Grade I, 25% in H_2O, Merck) at a final concentration (v/v) of 0.25%. Samples were protected from light and heat until return to the laboratory (approximately 30 min), where they were flash frozen in liquid nitrogen. Analysis was carried out within 24 h of sampling in an Accuri C6 cytometer (Becton Dickinson) equipped with a factory-standard optical system. Maintenance and calibration were performed according to the manufacturer's specifications, and acquired volumes from each sample were estimated from mass differ-

ence of the samples before and after the analysis using a precision balance (AUX220, Shimadzu), with an empirical correction for pump start/end cycle.

Coloured dissolved organic matter

Light absorbance by CDOM was measured throughout the UV and visible spectral domains using a WPI UltraPath liquid waveguide spectrophotometer along a 50 cm pathlength. Samples were filtered through a Polycap AS capsule filter (0.2 µm) and triplicate filtrates were collected in amber Qorpak bottles previously acid cleaned and sterilized. The spectral absorbance of the filtered water was measured between 320 and 750 nm immediately or within 3 h of sampling. Freshly produced, UV-irradiated Milli-Q water was used as a reference within 1 h of sampling. The spectral average of optical density among triplicates was converted into absorption coefficients, $a_{CDOM}(\lambda)$, in m^{-1}. An exponential function was fitted by nonlinear regression to each CDOM absorption spectrum between 350 and 600 nm, following the equation $a_{CDOM}(\lambda) = a_{CDOM}(\lambda_r) \exp[-S_{CDOM}(\lambda-\lambda_r)]$, where λ_r is a reference wavelength (nm), $a_{CDOM}(\lambda_r)$ (m^{-1}) is the absorption estimate at λ_r and S_{CDOM} (nm^{-1}) is the spectral slope of the $a_{CDOM}(\lambda)$ spectrum (Bricaud et al. 1981). In this study, the reference wavelength is 443 nm and $a_{CDOM}(443)$ represents the magnitude of CDOM absorption (directly proportional to its concentration), and S_{CDOM} is indicative of the CDOM composition (Bricaud et al. 1981).

Nutrients

Sub-samples also filtered using a Polycap AS capsule filter (0.2 µm) were preserved in polyethylene flasks (–20°C) to measure the concentration of dissolved inorganic nutrients (µmol l^{-1}). Phosphate (P-PO_4^{3-}) and silicate (Si-$Si(OH)_4$) were determined by a colorimetric method as described in Grasshoff et al. (1983) using a BioSpectro SP 220 spectrophotometer (precisions = 0.01 µmol P-PO_4^{3-} l^{-1} and 0.02 µmol Si-$Si(OH)_4$ l^{-1}). Nitrate (N-NO_3^-) was determined by reduction to nitrite (N-NO_2^-) in a Cd/Cu column, and both total nitrite and initial nitrite were determined using a Bran-Luebbe AutoAnalyzer II using standard methods (Tréguer & Le Corre 1975). Precisions were 0.01 µmol N-NO_2^- l^{-1} and 0.02 µmol N-NO_3^- l^{-1}. Dissolved ammonium concentration (N-NH_4^+) was determined with a precision of 0.05 µmol N-NH_4^+ l^{-1} (Tréguer & Le Corre 1975), but due to sample volume limitations, analysis could only be performed for 60% of the samples, mainly representing summer conditions.

Environmental variables

Vertical profiles of temperature, salinity, dissolved oxygen concentration, photosynthetically active radiation (PAR) and turbidity were measured using an AAQ-Rinko water quality profiler (JFE Advantech). Visible (400–700 nm; W m^{-2}) and UV (10–400 nm; index factor) spectral radiations and rainfall (mm) were recorded in 10 min intervals by a meteorological station (Vantage Pro 2, Davis) installed about 300 m from the sampling site. Cumulative rain was estimated during 24 h the day before sampling. Secchi depth (m) was also determined for each sampling day.

Data analysis

All means are noted in the manuscript with their respective standard error (mean ± SE). Variables were tested with the Shapiro–Wilk test at the 0.05 significance level, with all, with the exception of NCP, showing significant departures from normality. Therefore, the Wilcoxon test was used to evaluate the statistical significance of the comparisons. In addition, a principal components analysis (PCA; Pearson 1901) was applied to the set of depth-specific physical, chemical and biological data to explore association among variables. For these analyses, temperature, salinity, turbidity, incident PAR and dissolved oxygen concentration were extracted from CTD profiles at corresponding depth. Meteorological variables (cumulative rain, solar radiation and daily UV doses from 00:00 to 23:50 h) were extracted from the meteorological station for each sampling day. They describe external conditions and are the same for all depths. These data were thus combined with measured data: planktonic metabolism, chl *a*, BA, Si-$Si(OH)_4$, N-NO_3^- + N-NO_2^-, P-PO_4^{3-}, Secchi depth, $a_{CDOM}(443)$ and S_{CDOM}. PCAs were calculated for the whole experiment except for March, when we were able to observe planktonic metabolism only. To get a normal distribution and place all measurements on a common scale, each variable was log-transformed and standardized by subtracting its mean from each value and dividing it by the standard deviation. PCA results were summarized in a 2-dimensional biplot for each sampled depth.

RESULTS

Sampling field

The variability of the main environmental parameters and variables is described in Table 1. The sampled depths of 1, 7 and 12 m received about 67 ± 4%, 16 ± 3% and 4 ± 1%, respectively, of the surface incident PAR (µmol photon m^{-2} d^{-1}). These seawater samples can be considered as a good representation of the euphotic layer considering that the euphotic zone depth is commonly approximated as the depth at which only 1% of the surface PAR remains (Kirk 1994). During late spring, observations of seawater temperature, salinity, density (σ_T) and dissolved oxygen concentration were homogeneous from the surface to 12 m depth, characterizing a well-mixed water column, while stratification developed in summer (Fig. 2), as generally seen in the SSB for this season (Cerda & Castro 2014). Indeed, during summer, surface waters were warmer (25.0 ± 0.4°C) and less salty (34.4 ± 0.1) than at 12 m depth (19.6 ± 0.5°C and 35.4 ± 0.1). Considering the entire water column, seawater was warmer and fresher during spring than during summer (Wilcoxon test, $p < 0.0001$ and $p = 0.0004$, respectively; Table 1).

Nutrient concentrations were relatively low during the whole study period, except for silicate concentration, with a mean about 13 µmol Si-Si$(OH)_4$ l^{-1}. Silicate concentration was significantly higher (22.9 ± 4.2 µmol Si-Si$(OH)_4$ l^{-1}) while nitrite + nitrate concentration significantly lower (0.5 ± 0.1 µmol N-NO_2 + N-NO_3 l^{-1}) during late spring than during summer (6.8 ± 0.3 µmol Si-Si$(OH)_4$ l^{-1} and 1.5 ± 0.3 µmol N-NO_2 + N-NO_3 l^{-1}; Wilcoxon test, $p < 0.0001$ and $p = 0.0165$, respectively; Table 1). Phosphate concentration did not differ between the two seasons (Wilcoxon test, $p = 0.0763$; Table 1) and in general was low (0.3 ± 0.0 µmol P-PO_4^{3-} l^{-1}). Because of sample volume limitations, we could not compare ammonium concentration data between seasons, but during summer its concentration was relatively low (0.6 ± 0.1 µmol N-NH_4^+ l^{-1}). Values for the CDOM parameters, $a_{CDOM}(443)$ and S_{CDOM}, showed little variation during the whole study. S_{CDOM} did not show a significant difference between seasons, with an average value of 0.0199 ± 0.0001 (Wilcoxon test, $p = 0.4979$), whereas $a_{CDOM}(443)$ was significantly higher during late spring (0.0734 ± 0.0053 m^{-1}) than summer (0.0453 ± 0.0022 m^{-1}) (Wilcoxon test, $p < 0.0001$; Table 1). Chl *a* and bacterial abundance had average values of 2.2 ± 0.1 mg m^{-3} and 13.1 ± 0.3 10^5 cell ml^{-1}, respectively, with similar ranges in both seasons (Table 1). As observed for seawater temperature and salinity, chl *a* was similar throughout the water column during late spring. However, chl *a* was significantly higher at 12 m depth (0.7 ± 0.1 mg m^{-3} in surface water and 2.0 ± 0.3 mg m^{-3} at 12 m depth; Wilcoxon test, $p < 0.0001$) during summer. During summer, colder waters with high salinity were characterized by higher chl *a* concentrations than in warmer and less salty waters (Spearman's correlation coefficients: salinity versus temperature, $\rho = -0.89$, $p < 0.0001$; chl *a* versus temperature $\rho = -0.70$, $p < 0.0001$; chl *a* versus salinity, $\rho = 0.62$, $p < 0.0001$).

Planktonic metabolism

Volumetric NCP, CR and GPP rates showed global means for the whole period sampled of about 2.7 ±

Table 1. Mean ± SE, minimum, maximum and number of estimates (N) of environmental (temperature, salinity, solar irradiance, nutrient concentrations, magnitude of coloured dissolved organic matter absorption [$a_{CDOM}(443)$]; spectral slope of the $a_{CDOM}(\lambda)$ spectrum [S_{CDOM}]) and biological (chl *a* and bacterial abundance [BA]) parameters during late spring and summer sampling in the São Sebastião Channel

	Late spring			Summer		
	Mean ± SE	Range	N	Mean ± SE	Range	N
Temperature (°C)	24.3 ± 0.1	23.1–25.3	26	22 ± 0.5	16.9–28.2	39
Salinity	33.7 ± 0.1	32.3–34.4	26	35 ± 0.1	33.2–35.6	39
Solar radiance (W m^{-2})	190.3 ± 19.2	107.7–261.6	8	171.9 ± 21.8	61.7–308.1	14
Nitrite + nitrate (µmol l^{-1})	0.5 ± 0.1	0.2–1.3	23	1.5 ± 0.3	0.1–10.1	39
Silicate (µmol l^{-1})	22.9 ± 4.2	5.3–81.8	23	6.8 ± 0.3	2.9–14.6	39
Phosphate (µmol l^{-1})	0.3 ± 0.0	0.2–0.5	23	0.3 ± 0.0	0.1–0.8	39
Ammonium (µmol l^{-1})	0.4 ± 0.1	0.2–0.5	2	0.6 ± 0.1	0.1–2.5	36
$a_{CDOM}(443)$ (m^{-1})	0.0734 ± 0.0053	0.037–0.1302	20	0.0453 ± 0.0022	0.0174–0.0789	37
S_{CDOM} (nm^{-1})	0.0195 ± 0.0002	0.0165–0.0222	20	0.0201 ± 0.0003	0.0176–0.0241	37
Chl *a* (mg m^{-3})	2.2 ± 0.1	1.4–3.9	23	2.3 ± 0.2	0.7–4.7	39
BA (10^5 cell ml^{-1})	12 ± 0.5	9.2–16.4	23	13.7 ± 0.4	7.9–19.6	39

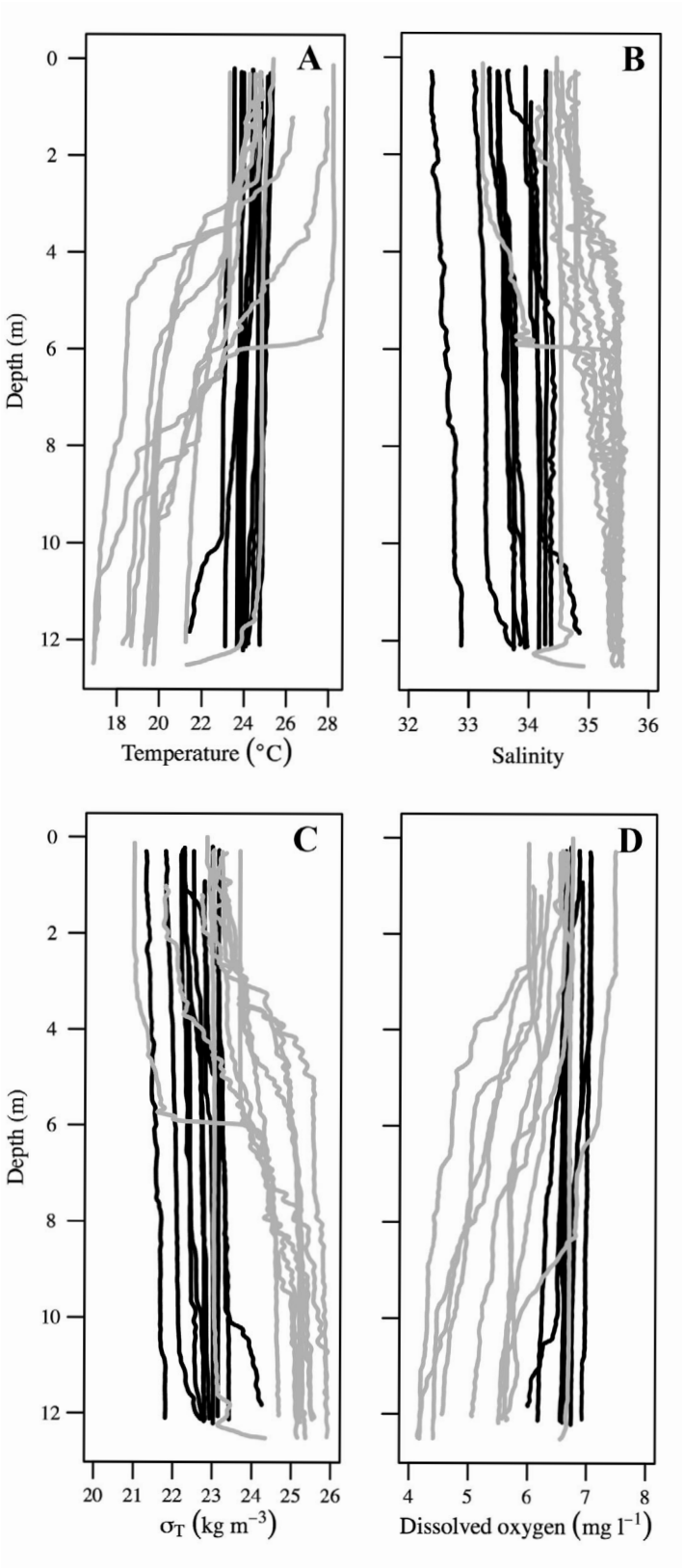

Fig. 2. (A) Salinity, (B) temperature (°C), (C) σ_T (density, kg m^{-3}) and (D) dissolved oxygen (mg l^{-1}) profiles from surface to 12 m depth of the water column during late spring (black lines) and summer (grey lines)

1.1, 6.9 ± 0.4 and 10.1 ± 1.1 mmol O_2 m^{-3} d^{-1}, respectively. The mean volumetric GPP/CR ratio (i.e. ratio estimating the balance between GPP and CR) was about 1.5 ± 0.2 mmol O_2 m^{-3} d^{-1}. Volumetric NCP, GPP and GPP/CR ratio were significantly higher at 1 m depth than at 7 and 12 m depth (Wilcoxon test, $p < 0.0001$), whereas CR was not significantly different among depths (Wilcoxon test, $p = 0.1856$) (Fig. 3). Volumetric plankton metabolism rates (NCP, GPP and CR) showed great variation along the water column and over time (Fig. 3). Volumetric plankton metabolism rates were not significantly different at 1 and 12 m depth between late spring and summer (Wilcoxon test, $p > 0.05$; Fig. 3). Although volumetric CR at 7 m depth was not significantly different between the two seasons (Wilcoxon test, $p = 0.8674$), volumetric GPP and NCP rates at 7 m depth were significantly different between late spring and summer (Wilcoxon test, $p = 0.0178$ and $p = 0.0376$, respectively).

We did not observe a significant relationship between volumetric plankton metabolism rates and environmental variables (temperature, salinity, chl *a*, BA, nitrite + nitrate, silicate and phosphate concentrations) during the whole experiment period; however, note that experiments in March were not included here due to lack of concurrent environmental data. However, some concurrent patterns were observed through PCAs (Fig. 4). At the surface (Fig. 4A), volumetric planktonic metabolism was correlated with chl *a* and related to turbidity. Salinity tended to be positively related to nitrite + nitrate concentration and negatively related to temperature, rainfall and $a_{CDOM}(443)$. At 7 m depth (Fig. 4B), volumetric NCP and GPP tended to be positively related to salinity and inversely related to temperature and silicate concentration. At the bottom depth (Fig. 4C), CR tended to be positively related to phosphate and nitrite + nitrate concentration. Volumetric NCP and GPP were weakly related to turbidity and temperature. All planktonic communities (100%) were autotrophic at the surface, 62% at 7 m depth and 14% at 12 m (Fig. 4A, B and C, respectively).

During the whole experimental period, depth-integrated GPP and NCP were about 2 times more variable over time than depth-integrated CR (Fig. 5). Depth-integrated NCP values ranged from –47 mmol O_2 m^{-2} d^{-1} to 152.3 mmol O_2 m^{-2} d^{-1}, with a global mean of 31 ± 10.5 mmol O_2 m^{-2} d^{-1}, and depth-integrated GPP ranged from 50.2 mmol O_2 m^{-2} d^{-1} to 213.5 mmol O_2 m^{-2} d^{-1}, with a global mean of 106.7 ± 11.5 mmol O_2 m^{-2} d^{-1}. During summer, the depth-integrated metabolic rate variations tended to be higher than during the late spring sampling period (Fig. 5), but not significantly so (Wilcoxon test, $p > 0.05$).

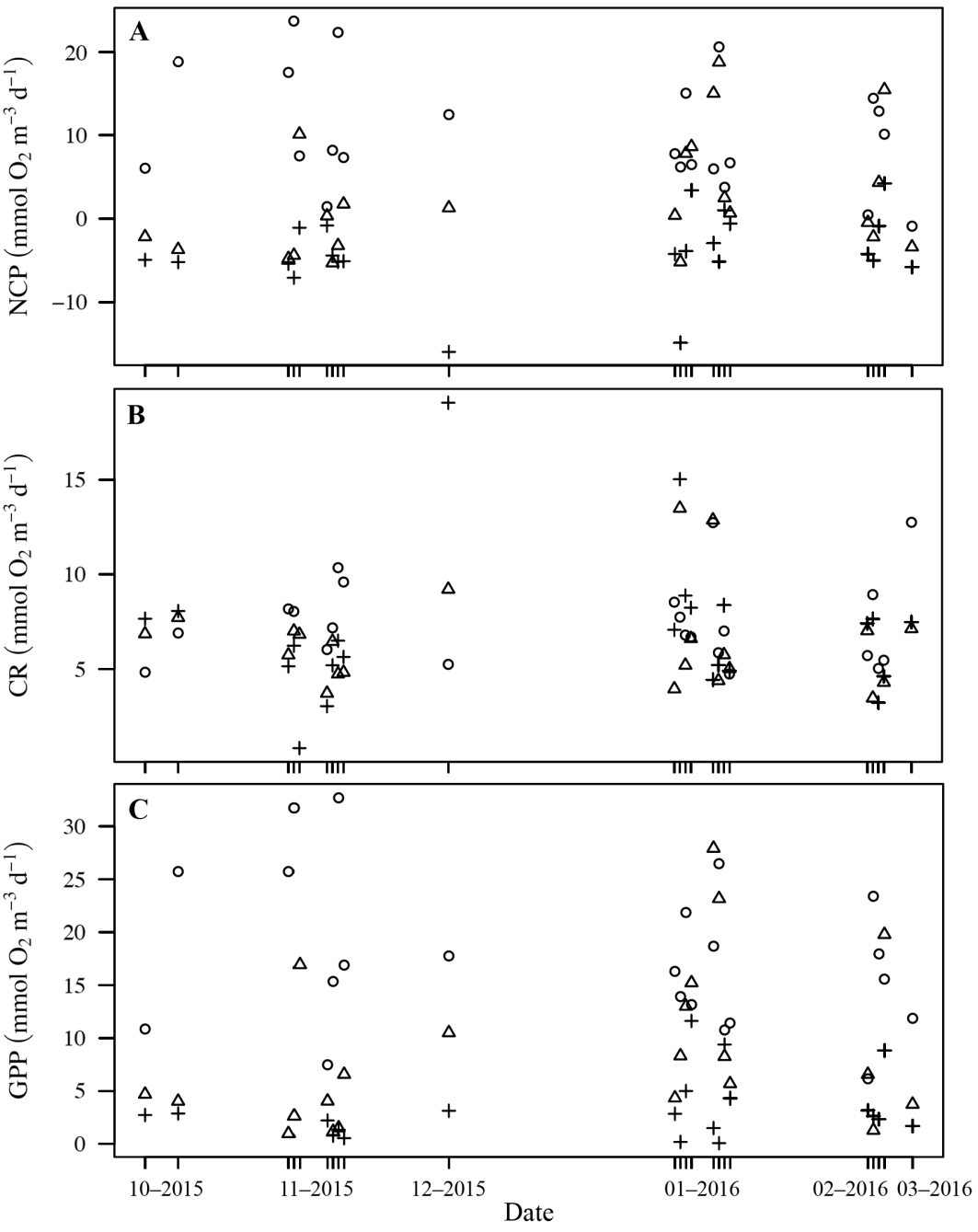

Fig. 3. Variation over sampling time of volumetric (A) net community production (NCP), (B) community respiration (CR) and (C) gross primary production (GPP) for the 3 sampled depths (circles: 1 m; triangles: 7 m; plus signs: 12 m). Each tick mark on *x*-axis represents a sampling event (e.g. in October 2015, seawater was sampled on 2 days; in November 2015, on 7 days)

The threshold of GPP (i.e. the amount of GPP that defines autotrophic and heterotrophic communities) was derived from the significant relationship between volumetric NCP and GPP ($r^2 = 0.89$, $p < 0.01$), and was estimated at 7.10 mmol O_2 m^{-3} d^{-1} (Fig. 6A). CR was, in general, less variable and lower than GPP. Fifty-seven percent of all GPP/CR ratio data was higher than 1 (Fig. 6B).

SACW influence

During the summer sampling, 10 experiments out of 13 had temperatures lower than 20°C and salinities higher than 35 near the bottom layer (12 m depth). Nutrient concentrations were about 2- to 4-fold higher in these 10 experiments than in the others. Thus, the bottom layer seemed to receive an

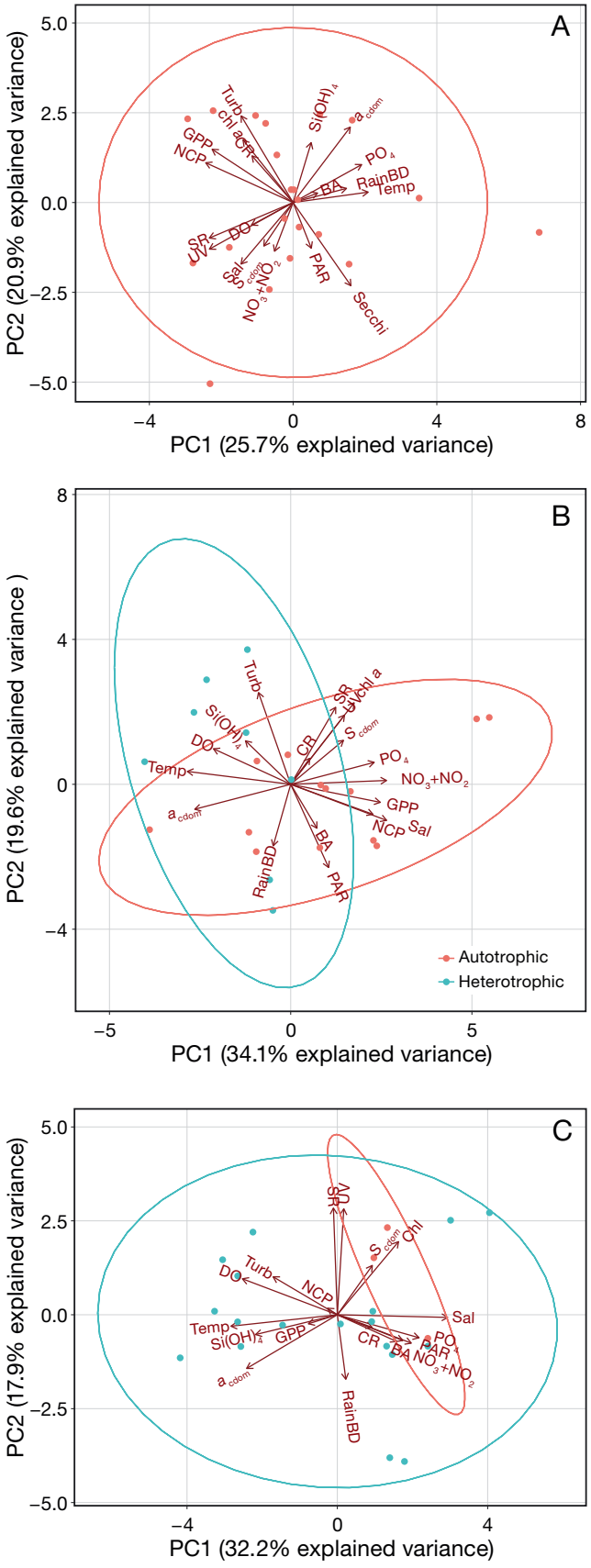

Fig. 4. Biplots of results from principal components analysis (PCA) at (A) 1 m, (B) 7 m and (C) 12 m depth on environmental parameters (seawater temperature [Temp]; salinity [Sal]; cumulative rain the day before the sampling [RainBD]; UV dose [UV]; solar radiation [SR]; photosynthetically active radiation reaching corresponding depth [PAR]; turbidity [Turb]; Secchi depth [Secchi]; nitrate + nitrite concentration [NO_3+NO_2]; phosphate concentration [PO_4]; silicate concentration [Si]; dissolved oxygen concentration [DO]; magnitude of coloured dissolved organic matter absorption [a_{CDOM}]; spectral slope of the $a_{CDOM}(\lambda)$ spectrum [S_{CDOM}]) and biological parameters (chlorophyll *a* concentration [chl *a*]; bacterial abundance [BA]; gross primary production [GPP]; net community production [NCP]; community respiration [CR]). Secchi depth was represented only for surface PCA. When the volumetric NCP rates are <0, the dots are in blue (heterotrophic) and when volumetric NCP rates are >0, the dots are in pink (autotrophic). The blue and pink ellipses encompass 95% of the data that are heterotrophic and autotrophic, respectively

upwelled cool and salty nutrient-rich and -mixed water (partly SACW) for these experiments. On the contrary, the rest of the experiments during spring and summer had the same characteristics as CW (salinity <34.5 and temperature >20°C). These observations are consistent with previous descriptions of the SACW presence in the SSC (Braga & Müller 1998, Castro & Miranda 1998, Silva et al. 2005), with sporadic southern intrusions of nutrient-rich cold and salty SACW waters during spring and summer, although their frequency is higher in summer.

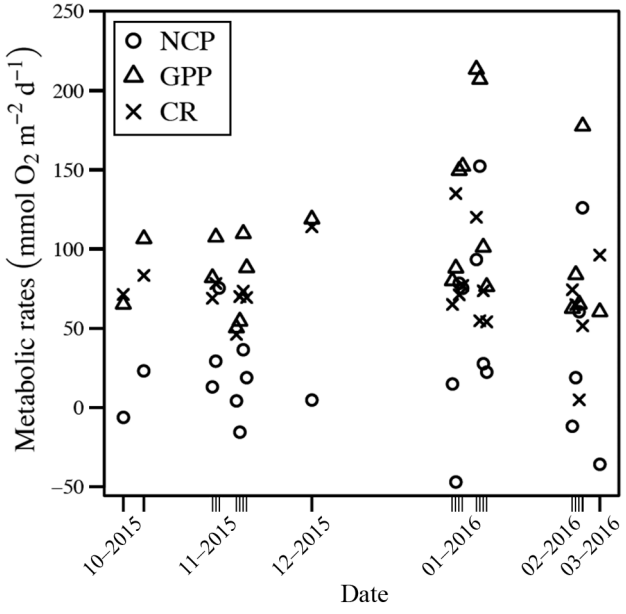

Fig. 5. Variation over sampling time of depth-integrated net community production (NCP), community respiration (CR) and gross primary production (GPP) (mmol O_2 m^{-2} d^{-1})

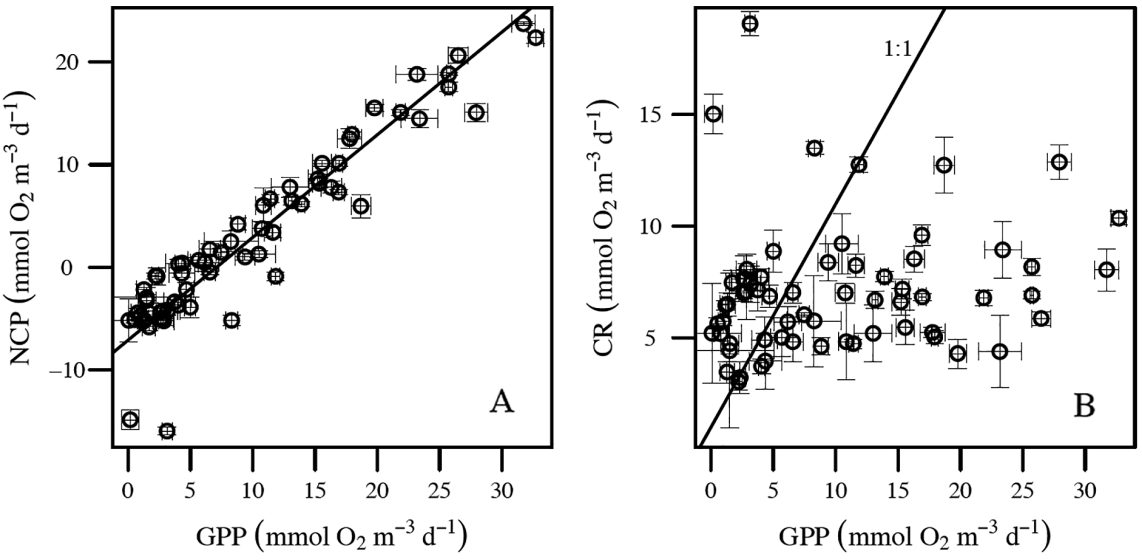

Fig. 6. Volumetric net community production (NCP; A) and volumetric community respiration (CR; B) versus volumetric gross primary production (GPP). The solid line in (A) represent the standard major axis (SMA) regression, the equation for which is: NCP = 0.99 (±0.08) GPP − 7.09 (±1.13), r^2 = 0.89, p < 0.01, n = 64. The solid line in (B) represents the 1:1 relationship. The slope of the SMA regression is not significantly different from 1 (*t*-test, p = 0.1831). Error bars are SE

Table 2. Mean ± SE and number of estimates (N) of depth-integrated net community production (NCP), community respiration (CR), gross primary production (GPP) and the GPP/CR ratio from experiments associated with the presence of mixed South Atlantic Coastal Water (SACW) or Coastal Water (CW) at 12 m depth

	Mixed SACW		CW	
	Mean ± SE	N	Mean ± SE	N
NCP (mmol O_2 m^{-2} d^{-1})	60.0 ± 18.5	10	10.8 ± 5.2	11
CR (mmol O_2 m^{-2} d^{-1})	79.24 ± 9.60	9	73.0 ± 5.2	11
GPP (mmol O_2 m^{-2} d^{-1})	139.2 ± 17.7	9	81.3 ± 8.5	9
GPP/CR (mmol O_2 m^{-2} d^{-1})	1.96 ± 0.35	9	1.12 ± 0.08	9

Depth-integrated GPP and NCP rates were significantly higher for experiments associated with the presence of mixed upwelled SACW in deeper layers (139.2 ± 17.7 and 60 ± 18.5 mmol O_2 m^{-2} d^{-1}, respectively) than for those associated with CW (81.3 ± 8.5 and 10.8 ± 5.2 mmol O_2 m^{-2} d^{-1}; Wilcoxon test, p = 0.0243 and 0.0201, respectively; Table 2). Similarly, the depth-integrated GPP/CR ratio was consistently higher under the influence of mixed upwelled SACW in deeper layers (1.96 ± 0.35 mmol O_2 m^{-2} d^{-1}) than with CW (1.12 ± 0.08 mmol O_2 m^{-2} d^{-1}; Wilcoxon test, p = 0.0380; Table 2). In contrast, depth-integrated CR, volumetric metabolism rates and chl *a* concentration did not differ between both conditions (Wilcoxon test, p > 0.05; Table 2).

DISCUSSION

Terrestrial inputs

In previous studies, the SSC was characterized as an oligotrophic ecosystem with low nutrient concentration and therefore likely unable to sustain high phytoplankton biomass and primary production (Gaeta et al. 1990, Gianesella-Galvão et al. 1997, Gianesella et al. 1999). Except for silicate, nutrient concentrations reported here were in the same range as those observed in previous studies (Gaeta et al. 1990, Gianesella-Galvão et al. 1997, Gianesella et al. 1999). Although most of our observations of silicate were within the previous published values for the region (1 to 14 µmol Si-Si$(OH)_4$ l^{-1}; Gaeta et al. 1990, Gianesella-Galvão et al. 1997, Gianesella et al. 1999, Giannini & Ciotti 2016), with a median of 8.2 µmol Si-Si$(OH)_4$ l^{-1}, the lower limit was 3 times higher and extreme concentrations were observed of up to 81.8 µmol Si-Si$(OH)_4$ l^{-1}. These high concentrations were observed during late spring. During this study, silicate concentration was inversely correlated to salinity (OLS regression, r^2 = 0.21, p = 0.0002). Indeed, silicate concentration and salinity was significantly higher and lower, respectively, in the late spring than

in summer (see 'Results' section). Marine water receives silicate inputs mainly from the lithosphere through river runoff and rainfall (Conley 2002). Although no major river flows into the SSC, its width is very narrow and delimited by only a few kilometres (7.2 km in the north, 5.6 km in the south and 1.9 km in the centre). Thus, the occurrence of strong winds, currents and/or rainfall may promote terrestrial inputs into the SSC through advection or leashing. This hypothesis is supported by positive trends observed in surface and bottom layers between silicate concentration and $a_{CDOM}(443)$ (OLS regression, surface: $r^2 = 0.12$, $p > 0.05$, bottom: $r^2 = 0.30$, $p = 0.0131$; Fig. 4A,C) and negative correlations between silicate concentration and salinity (OLS regression, surface: $r^2 = 0.23$, $p = 0.0328$, bottom: $r^2 = 0.64$, $p = 0.0002$; Fig. 4A,C). Thus, this subtropical coastal ecosystem seems to receive important freshwater inputs that may affect its planktonic community.

Planktonic metabolism

This study quantified for the first time the planktonic metabolism in the subtropical coastal upwelling system of the SSC. Volumetric planktonic metabolism varied among the sampled depths, but it was always more productive at the surface (Fig. 3). Although somewhat higher in summer than late spring (Figs. 3 & 5), the planktonic metabolism was in general much higher than one should expect considering the meso-oligotrophic classification of this ecosystem by earlier studies. More than 80% of our observations were classified as autotrophic (NCP > 0), with high depth-integrated mean GPP of 122.5 ± 13.4 mmol O_2 m^{-2} d^{-1}, compared with lower depth-integrated mean GPP of observations classified as heterotrophic (66.1 ± 5.7 mmol O_2 m^{-2} d^{-1}). Furthermore, the threshold of GPP observed in this study was in the same range found in the European coastal sector (5.7 mmol O_2 m^{-3} d^{-1}) or in the Arctic Ocean (5.5 mmol O_2 m^{-3} d^{-1}) (Duarte & Regaudie-de-Gioux 2009), both characterized as productive ecosystems receiving nutrients mainly through allochthonous inputs.

Gaeta et al. (1990) published the only estimates of particulate primary production (PPP) for the SSC, with values from 0.2 to 6.5 mmol C m^{-3} d^{-1}. Although their study took place during autumn when SACW intrusion was missing, nutrient and chl *a* concentrations were about the same range as observed here. The PPP values observed by Gaeta et al. (1990) in the SSC were similar (1 to 6.5 mmol C m^{-3} d^{-1}) to those observed in a close subtropical coastal ecosystem north of the SSC, Ubatuba (Teixeira 1973, 1979, Tundisi et al. 1978, Gaeta et al. 1999), that is also subjected to SACW intrusion during spring and summer. These studies estimated PPP using the ^{14}C method (Nielsen 1952) after short incubations (4–6 h). Previous work has showed that primary production using the ^{14}C method is closer to GPP after a short incubation than a long one (12 h) (e.g. Nielsen 1952, Móran & Estrada 2002, Marra 2002). Although those studies used short-time incubations to estimate PPP, the GPP observed here with values from 0.1 to 32.7 mmol O_2 m^{-3} d^{-1} was much higher than PPP values observed previously. Photosynthetic quotient can be used to convert GPP values into units of carbon. Considering that photosynthetic quotients exhibit values from 1 to 2 (Williams et al. 1979, Laws et al. 2000, Hendricks et al. 2004), GPP with units of carbon would still be much higher than PPP previously observed in this region. All of these studies reported their results as phytoplankton primary production while analysing PPP. In fact, they analysed ^{14}C incorporated into particles retained in filters (Nielsen 1952) without considering ^{14}C incorporated into dissolved organic carbon, and thus they have likely underestimated primary production. Indeed, Regaudie-de-Gioux et al. (2014) observed that ^{14}C-PPP was 7 times lower than GPP measured by the dark-light method when they were used concurrently. Bender et al. (1987) explained that ^{14}C-PPP does not account for dissolved organic carbon release or respiratory losses by the community. The conclusion of previous studies that both SSC and Ubatuba coast ecosystems would be low in productivity was probably biased by the method they used. According to our results, the primary production in these subtropical coastal ecosystems is expected to be much higher than previously thought.

Influence of SACW and terrestrial inputs

Considering the entire water column, we observed the effect of mixed upwelled SACW. In fact, experiments that were characterized by bottom waters under the influence of nutrient-rich and -mixed water (partly SACW) were highly productive, with higher chl *a* content than those under the influence of CW. Furthermore, the influence of nutrient-rich and -mixed water (partly SACW) at bottom layers seemed to enhance the depth-integrated metabolism of autotrophic planktonic communities. Indeed, depth-integrated NCP of autotrophic communities

was 3-fold higher for experiments where bottom layers were under the influence of nutritive-mixed SACW than in those under the influence of CW (71.9 ± 15.8 and 19.0 ± 4.0 mmol O_2 m^{-2} d^{-1}, respectively; Wilcoxon test, p = 0.0161). Due to the low number of experiments characterized by heterotrophic communities (only 5), we were unable to perform this test for heterotrophic communities.

The nutrients supplied by terrestrial inputs and by upwelled SACW intrusion in the SSC allow sustained high primary production with a dominance of autotrophic communities. Although this subtropical coastal upwelling system may be considered as lower in nutrients than other coastal upwelling systems (e.g. off Peru, NW Africa), we observed here that strongly enhanced planktonic metabolism promotes the dominance of autotrophic communities. Considering the great ecological and biogeochemical interest in this subtropical coastal ecosystem, it is essential to perform similar studies throughout an entire year with periodic evaluations of planktonic metabolism at different points of the SSC (from the north to the south) in order to observe wind and current effects in the north and SACW intrusion in the south. Finally, considering the large latitudinal scale of the SBB and its large fisheries grounds and oil deposits, planktonic communities may vary considerably along the SBB. Other sites of the SBB deserve further sampling to investigate spatial differences in planktonic metabolism.

Acknowledgements. This research was funded by the Brazilian National Council of Research and Development (CNPq), the Araça project and the University of São Paulo (USP) – the Center for Marine Biology (CEBIMar). A.R.G. was supported by a post-doctoral fellowship 'Ciência sem fronteiras – Bolsa Atrações de Jovens Talentos' funded by the CNPq (BJT: 301179/2014-0). This research also received funds from CNPq (405133/2013-8), MMA-USP (761923/2011), Fapesp (2011/50317-5) and NP-BioMar (http://npbiomar.cebimar.usp). We thank C. B. Bellini, D. Gallo and R. Böhmer and V. G. Chiozzini for their assistance with the fieldwork and laboratory analyses, as well as CEBIMar technicians during samplings. This research is a contribution of the NP-BioMar, University of São Paulo.

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Editorial responsibility: Steven Lohrenz, New Bedford, Massachusetts, USA

Submitted: November 11, 2016; Accepted: April 15, 2017
Proofs received from author(s): May 31, 2017