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Inventory

Phytoplankton composition from Araçá Bay and São Sebastião Channel, São Paulo, Brazil

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Abstract: Despite its small area, Araçá Bay (AB) holds cultural, historical, and economic value and displays great benthic biodiversity. Thus, it is crucial to monitor its environmental health, including knowing the main groups of phytoplankton and their temporal variability. The shallow waters of Araçá Bay are continuously modified by the complex hydrography of the adjacent São Sebastião channel (SSC), challenging standard experimental designs for phytoplankton collection. Here we report changes in phytoplankton composition at intervals of five to six weeks from September 2013 to August 2014 in both Araçá Bay and SSC. Samples were collected twice daily for three consecutive days to increase taxonomic resolution. Our goal was to provide an inventory of species occurrences to aid future public policies and environmental management of the area. Analyses revealed high species richness and 166 different phytoplankton taxa. Diatoms and dinoflagellates were always numerically dominant, but taxa occurrence changed markedly. Diatoms of the genera *Pseudo-nitzschia* were abundant during spring and summer concurrently to signatures of South Atlantic Central Water in the SSC, while *Thalassiosira* occurred when waters displayed relatively lower salinity. The inventory demonstrated several potentially harmful species of microalgae and cyanobacteria, strongly suggesting investments in monitoring programs in this area that currently experience an increase in population.
Keywords: *Biodiversity;* Pseudo-nitzschia; Thalassiosira; *São Paulo coast; coastal marine environments.*

Composição do fitoplâncton da Baía do Araçá e Canal de São Sebastião, São Paulo, Brasil

Resumo: Apesar de sua pequena área, a baía do Araçá (AB) possui grande valor cultural, histórico e econômico, e biodiversidade bentônica. Assim, é fundamental monitorar sua saúde ambiental, que inclui conhecer os principais grupos de fitoplâncton e sua variabilidade temporal. As águas rasas da baía do Araçá são continuamente modificadas pela hidrografia complexa do canal de São Sebastião (SSC), desafiando desenhos experimentais convencionais para coleta de fitoplâncton. Aqui relatamos mudanças sazonais na composição do fitoplâncton, em intervalos de 4 a 6 semanas, de setembro de 2013 a agosto de 2014 na baía do Araçá e no SSC, sendo coletadas duas vezes ao dia por três dias consecutivos em cada campanha de amostragem para aumentar a resolução taxonômica. Nosso objetivo foi fornecer um inventário de ocorrência de espécies para auxiliar futuras políticas públicas e gestão ambiental na área. As análises revelaram alta riqueza de espécies e 166 táxons fitoplanctônicos diferentes. Diatomáceas e dinoflagelados foram numericamente dominantes, mas a ocorrência de táxons mudou acentuadamente entre observações. As diatomáceas do gênero *Pseudo-nitzschia* foram abundantes durante a primavera e o verão concomitantemente às assinaturas da Água Central do Atlântico Sul no CSS, enquanto *Thalassiosira* ocorreu durante períodos de salinidade relativamente mais baixa. O inventário demonstrou várias espécies potencialmente nocivas de microalgas e cianobactérias, sugerindo fortemente investimentos para programas de monitoramento nesta área que vem registrando aumento populacional contínuo.
Palavras-chave: *Biodiversidade; Pseudo-nitzschia; Thalassiosira; litoral paulista; ambientes marinhos.*

Introduction

Phytoplankton communities vary according to the physicochemical conditions of the water (Margalef 1967), but knowledge on the specific composition of these communities remain challenging (Basterretxea et al. 2020). The occurrence and the dominance of a given phytoplankton species reflect its adaptation to the environment (e.g., Anderson et al. 2002, Kremer et al. 2017, Moser et al. 2017, Ryabov et al. 2021). Hence some large-scale generalizations about the taxonomic variability and abundance of phytoplankton can be made in the ocean. Nearshore, however, environmental conditions vary over time scales of hours to days, and the same is true for phytoplankton diversity, for which observations require intense sampling effort. The quantification of species in the world ocean (Sournia, 1991) is a laborious work. Although new instruments and techniques (e.g. Sosik & Olson, 2007) are now available, microscopy analyses remain invaluable for their validation. The availability of phytoplankton species inventories is essential at urbanized coastal sites as they subsidize environmental management actions.

The São Sebastião channel (SSC), located in the north portion of the São Paulo state coast, between the municipalities of São Sebastião and Ilhabela, is partially inserted in the Marine Environmental Protection Area of the North Coast of the State of São Paulo. In the central portion of the channel, a shallow tidal plain (average depth of 1.5 m) limited by rocky flanks (Amaral et al. 2010) is known as Araçá Bay. The bay has an extensive intertidal region, which can be fully exposed to the air and exceeds 300 m in length during low spring tides (Amaral et al. 2018), with a large area of the plain being immersed and submerged within the same tidal cycle (Siegle et al. 2018). Araçá Bay is of esteemed value to the local population because, in addition to harboring high biological diversity (Amaral et al. 2010), it is a stronghold of artisanal fishers who traditionally use small vessels for fishing or leisure (Amaral et al. 2018). In the past years, the north coast of São Paulo experienced increasing population growth and environmental impacts (Xavier et al. 2016), including the discharge of untreated sewage. Indeed, there is evidence that the interaction of the São Sebastião channel with the continent plays an important role in the exchange of nutrients (Gubitoso et al. 2008) on primary productivity (Regaudie et al. 2017).

Available phytoplankton studies in SSC waters consisted of surveys reporting changes in biomass (i.e., chlorophyll concentration) or relative abundance of major taxonomic groups and their relationships with nutrient concentrations (Muller-Melchers 1955, Gianesella et al. 1999, Saldanha-Corrêa & Gianesella 2003). A review of phytoplankton studies carried out along the São Paulo coast provided a comprehensive inventory of the species present from 1913 to 2006 (Villac et al. 2008). However, no further diversity studies are available. More recent analyses of changes in chlorophyll concentration fractionated by size classes (Giannini & Ciotti 2016) and main taxonomic groups (Ciotti et al. 2018a) derived from efforts during the Araçá Thematic FAPESP project (https://biota-araca.org/index.html), conducted from September 2013 to August 2014 and showed the importance of diatoms when phytoplankton biomass increased. The present study is also derived from the Araçá Project phytoplankton dataset (Tocci 2016) and focuses on detailed taxonomic descriptions of phytoplankton in Araçá Bay and SSC, using light microscopy. The main objective is to update the phytoplankton taxa for this region, report their relative occurrence frequencies, and describe differences between the species found in Araçá Bay and the adjacent waters in the São Sebastião channel.

Materials and Methods

The phytoplankton *checklist* is composed of samples derived from three oceanographic stations located in the interior of Araçá Bay (AB)

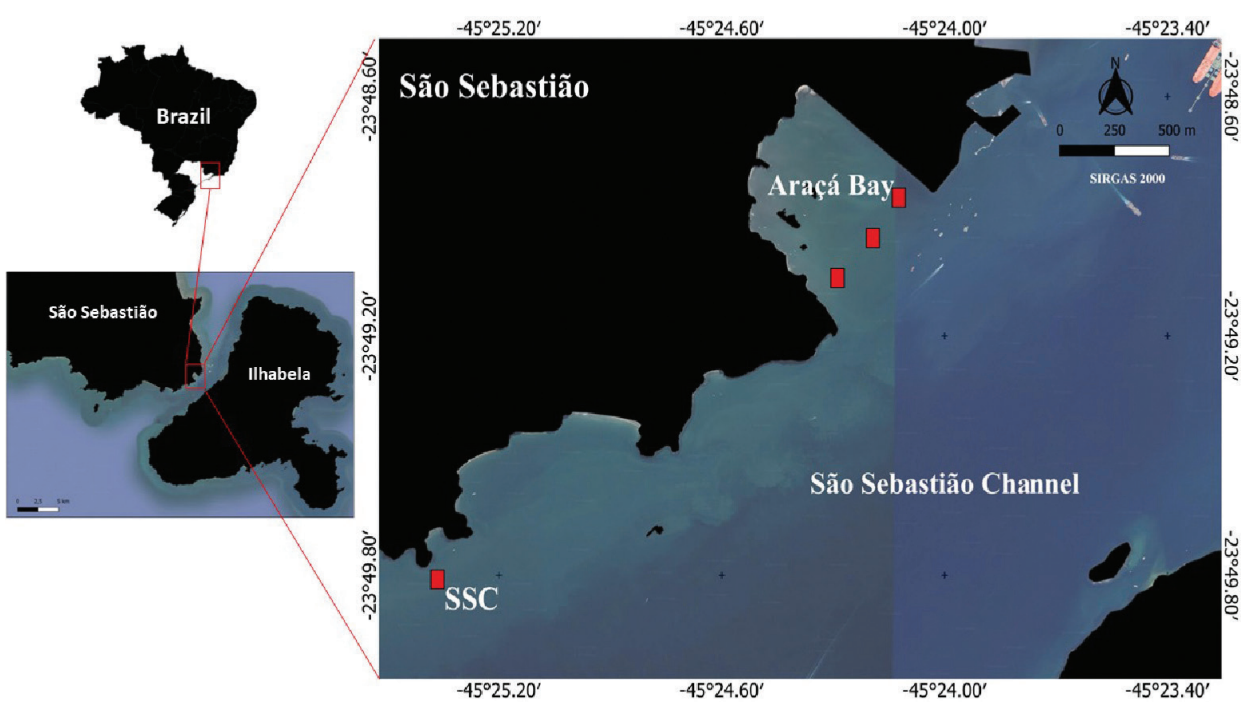

Figure 1. Location of the sampling sites at Araçá Bay (AB) and São Sebastião Channel (SSC) near the oceanographic buoy of SIMCosta Project.

and a single station located in the southern portion of the São Sebastião Channel (SSC) at the 15 m isobath (Figure 1). Nine surveys occurred between September 2013 and August 2014 every five to six weeks, in the morning and afternoon of three consecutive days (Ciotti et al. 2018b) to increase the probability of observing the variable hydrodynamics of SSC and rarer phytoplankton taxa. We used a Sontek Castway CTD to vertically profile the temperature and salinity at each station and a 5 L Van Dorn bottle to collect water for analyses of inorganic nutrients, chlorophyll-a, and phytoplankton cell enumeration. Three water samples were combined to represent AB and SSC to increase the representativeness of the occurring taxa analysis. AB samples refer to the combination of three independent stations located at isobaths between 1.5 and 2 m, while SSC refers to three successive deployments of the Van Dorn bottle (Tocci, 2016). The composite samples were further concentrated (2 L to about 100 mL) by reverse filtration with a 5 µm nylon mesh and preserved with formaldehyde neutralized in hexamethylenetetramine (0.4 %). Climatological data on precipitation rates were consulted on the CPTEC-INPE website (http://clima1.cptec.inpe.br/).

Cell enumeration and taxonomic analysis used Üthermol sedimentation chambers of 5 mL or 10 mL and an inverted optical microscope ZEISS-Axio® Observer D1 equipped with phase contrast and differential interference contrast (DIC). Only cells with maximum linear dimension MLD > 5 µm were counted, and identifications reached the lowest possible taxonomic level (genus and species) only for cells with MLD > 10 µm, with the help of specialized literature (e.g., Tomas 1997, Tenenbaum et al. 2004, Tenenbaum et al. 2006, Garcia & Odebrecht 2009, Haraguchi & Odebrecht 2010). Names and synonyms were checked and updated by queries of the *Algaebase* database (Guiry & Guiry 2021), and the diatom classification followed the work by Medlin & Kaczmarska (2004). The records for *Pseudo-nitzschia* followed the nomenclature of Hasle (1965) that divided the colony-forming species of the genus *Nitzschia* into two complexes: the "delicatissima" – for cells with widths equal to or smaller than 3 µm, and the «seriata» for cells wider than 3 µm. These two complexes were later combined and updated to the genera *Pseudo-nitzshia* (Hasle 1994).

The relative occurrence frequencies of taxa were calculated based on the method described by Matteucci & Colma (1982), which considers the overall number of occurrences of a taxon (65 samples for AB and 65 for SSC samples), following the categories: very frequent (VF) > 70%; frequent (F) ≤ 70% – >40%; infrequent (I) ≤ 40% – > 10%; and sporadic (S) <10%.

Results

Seawater temperature varied from 19.4 to 29.4° C in Araçá Bay (AB) and from 16.4 to 29.7°C in São Sebastião Channel (SSC), while salinity ranged from 30.8 to 36.6 in AB and from 30.9 to 35.7 in SSC (Table 1). A mixture of South Atlantic Central Water (SACW, thermohaline

Table 1. Environmental Variables measured during samplings at **São Sebastião Channel (SSC)** and **Araçá Bay (AB).** Minimum (Min), maximum (Max), average and standard deviation (SD), see Figure 1 for locations.

		SSC				AB			
		Min	Max	Average	SD	Min	Max	Average	SD
Temperature	°C	19.4	29.7	24.4	± 2.37	19.4	29.4	24.5	± 2.23
Salinity	–	30.9	35.7	34.3	± 1.21	30.8	36.6	34.4	± 1.13
Ammonia (NH_4)	$\mu mol.L^{-1}$	0.01	1.19	0.33	± 0.25	0.06	6.59	0.82	± 0.82
Nitrate plus Nitrite ($NO_3^- + NO_2^-$)	$\mu mol.L^{-1}$	0.01	0.77	0.27	± 0.14	0.01	2.02	0.52	± 0.33
Phosphate (PO_4)	$\mu mol.L^{-1}$	0.09	0.71	0.32	± 0.15	0.11	0.89	0.41	± 0.17
Silicate ($Si(OH)_4^{-4}$)	$\mu mol.L^{-1}$	0.47	6.25	03.08	± 1.25	0.64	5.62	3.63	± 1.33
Chlorophyll-a (Chla)	$mg.m^{-3}$	0.58	7.18	02.02	± 1.24	01.02	7.56	2.66	± 1.3

Table 2. Percentage of occurrence and summary statistics of the density ($cel.L^{-1}$) of the phytoplanktonic groups (> 5 µm), minimum (Min), maximum (Max), average, and standard deviation (SD) values. Taxonomic groups: centric diatom (CD) including both Coscinodiscophyceae and Mediophyceae, pennate diatom (PD), unarmored dinoflagellate (ND), armored dinoflagellate (TD), silicoflagellate (SI), flagellate (FL), coccolithophorid (CO) and cyanobacteria (CY). São Sebastião Channel (SSC) and Araçá Bay (AB).

Group	SSC					AB				
	%	Min	Max	Average	SD	%	Min	Max	Average	SD
CD	33.6	2.10^3	4.10^5	4.10^4	$\pm 6.10^4$	33.9	4.10^2	3.10^5	5.10^4	$\pm 7.10^4$
PD	67	9.10^2	1.10^6	9.10^4	$\pm 2.10^5$	40	3.10^2	6.10^5	6.10^4	$\pm 8.10^4$
ND	3.9	0	4.10^4	5.10^3	$\pm 8.10^3$	2	0	2.10^4	3.10^3	$\pm 5.10^3$
TD	10	0	8.10^4	1.10^4	$\pm 2.10^4$	3	0	3.10^4	5.10^3	$\pm 4.10^3$
SI	1.1	0	1.10^4	1.10^3	$\pm 2.10^3$	0.3	0	3.10^3	5.10^2	$\pm 7.10^2$
FL	12.7	0	5.10^4	2.10^4	$\pm 1.10^4$	16.7	6.10^2	9.10^4	3.10^4	$\pm 2.10^4$
CO	2.8	0	6.10^4	4.10^3	$\pm 1.10^4$	2.6	0	8.10^4	4.10^3	$\pm 1.10^4$
CY	2.5	0	3.10^4	3.10^3	$\pm 6.10^3$	1.5	0	4.10^4	2.10^3	$\pm 6.10^3$

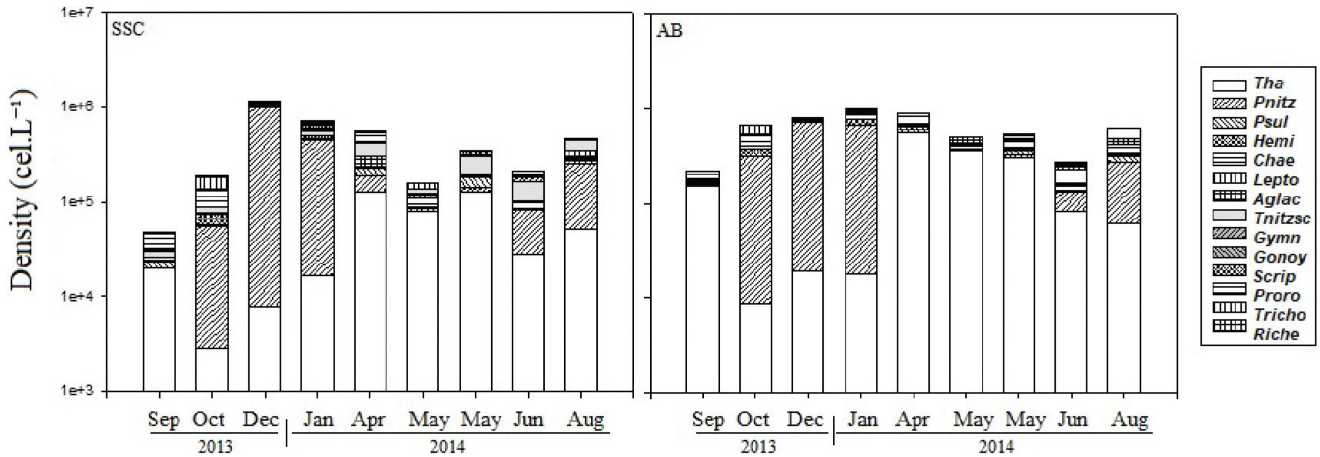

Figure 2. Variation in the density of the predominant phytoplanktonic taxa collected every 5–6 weeks between September 2013 and August 2014. Taxa: *Thalassiosira* sp.1 (Tha), *Pseudo-nitzschia* spp. (Pnitz), *Paralia sulcata* (Psul), *Hemiaulus* spp. (Hemi), *Chaetoceros* spp. (Chae), *Leptocylindrus* spp. (Lepto), *Asterionellopsis glacialis* (Aglac), *Thalassionema nitzschioides* (Tnitzsc), Gymnodiniales (Gymn), *Scrippsiella* spp. (Scrip), *Prorocentrum* spp. (Proro), *Trichodesmium* spp. (Tricho) and Richelia *intracellularis* (Riche). São Sebastião Channel (SSC) and Araçá Bay (AB).

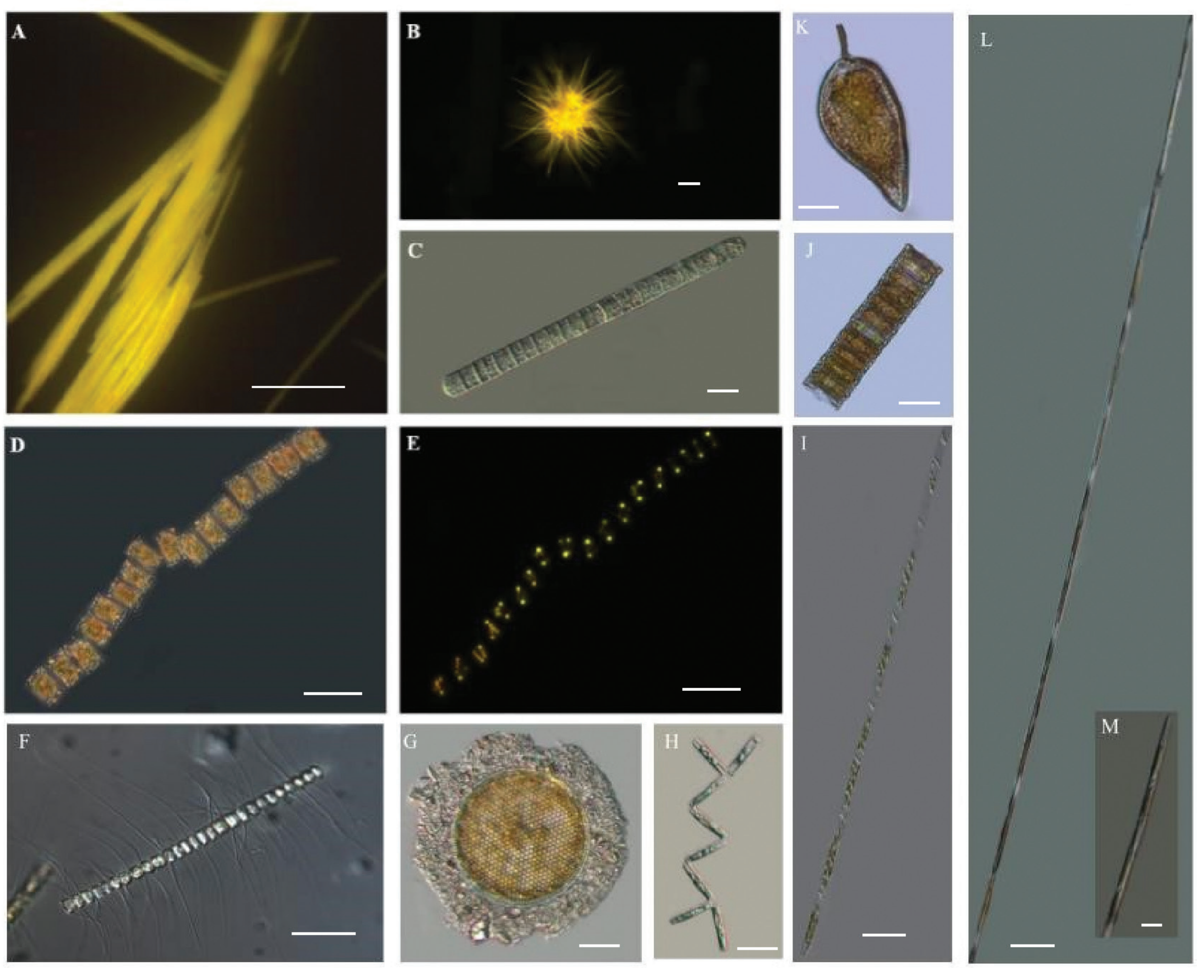

Figure 3. Diazotrophic cyanobacteria of the genus *Trichodesmium,* aggregates in bundles know as "tuffs" (**A**), in spherical "puffs" (**B**) and as a single trichome (**C**). Diatom *Hemiaulus membranaceus* (**D**) and diazotrophic cyanobacteria *Richelia intracellularis* (**E**) within it. Diatoms *Chaetoceros* cf. *debilis* (**F**), *Thalassiosira* sp.1 (**G**), *Leptocylindrus danicus* (**I**) and *Paralia sulcata* (**J**). Diatoms *Thalassionema nitzschioides* (**H**), *Pseudo-nitzschia* "*seriata* complex" sp.1 (see methods for definition) (**L, M**). Armored Dinoflagellate *Prorocentrum micans* (**K**). **A, B** epifluorescence microscopy image at 10x magnification; **E** epifluorescence microscopy image at 20x magnification; **I, L** DIC microscopy image at 200x magnification and **C, F, G, H, M** at 40x magnification; **D, K, J** phase-contrast microscopy image at 400x magnification. Scale bar: **A, B, D, E, F** = 50 µm; **C, G, H, I, J, K, L** = 10 µm; **M** = 05 µm.

Table 3. Taxonomic classification of the phytoplankton community observed in Araçá Bay (AB) and at São Sebastião Channel (SSC), between September 2013 and August 2014, see locations in Figure 1. Relative frequencies at each point: VF = very frequent, F = frequent, I = infrequent, S = sporadic; (MDL > 10 µm for the majority of taxa identified up to genera – species level).

Classification	Relative frequency	
	SSC	AB
Phylum Bacillariophyta		
Class Mediophyceae		
Subclass Biddulphiophycidae		
Order Biddulphiales		
Family Biddulphiaceae		
Biddulphia biddulphiana (J.E. Smith) Boyer	S	I
Family Bellerocheaceae		
Climacodium frauenfeldianum Grunow	I	I
Order Briggerales		
Family Streptothecaceae		
Helicotheca tamesis (Shrubsole) M.Ricard	S	–
Subclass Chaetocerotophycidae		
Order Chaetocerotales		
Family Chaetocerotaceae		
Bacteriastrum cf. *hyalinum* Lauder 1864	S	S
Bacteriastrum delicatulum Cleve	–	S
Chaetoceros aequatorialis Cleve	S	S
Chaetoceros affinis Lauder	S	I
Chaetoceros brevis F.Schütt	S	S
Chaetoceros concavicornis L.A.Mangin	S	S
Chaetoceros compressus Lauder	S	S
Chaetoceros curvisetus Cleve	I	I
Chaetoceros danicus Cleve	S	S
Chaetoceros cf. *debilis* Cleve	F	F
Chaetoceros decipiens Cleve	I	I
Chaetoceros didymus Ehrenberg	I	I
Chaetoceros lorenzianus Grunow	I	I
Chaetoceros peruvianus Brightwell	S	S
Chaetoceros subtilis Cleve	S	S
Order Hemiaulales		
Family Hemiaulaceae		
Cerataulina pelagica (Cleve) Hendey	I	I
Eucampia zodiacus Ehrenberg	S	S
Hemiaulus hauckii Grunow ex Van Heurck	I	I
Hemiaulus membranaceus Cleve	I	I
Hemiaulus sinensis Greville	S	I
Family Isthmiaceae		
Isthmia cf. *nervosa* Kütz	–	S
Subclass Cymatosirophycidae		
Order Cymatosirales		
Family Cymatosiraceae		
Cymatosira lorenziana Grunow	S	S
Subclass Thalassiosirophycidae		
Order Lithodesmiales		
Family Lithodesmiaceae		
Lithodesmium undulatum Ehrenberg	S	S
Ditylum brightwellii (T.West) Grunow	S	S
Order Thalassiosirales		
Family Thalassiosiraceae		
Detonula pumila (Castracane) Gran	–	S
Thalassiosira sp. 1	VF	VF
Thalassiosira sp. 2	F	F
Thalassiosira cf. *decipiens* (Grunow) Jørgensen	S	F
Thalassiosira gravida Cleve	S	S
Thalassiosira punctigera (Castracane) Hasle	F	F
Thalassiosira cf. *minuscula* Krasske	I	I
Family Skeletonemataceae		
Skeletonema cf. *costatum* (Greville) Cleve	I	I
Family Lauderiaceae		
Lauderia annulata Cleve	I	I
Order Stephanodiscales		
Family Stephanodiscaceae		
Cyclotella cf. *litoralis* Lange & Syvertsen	S	I
Cyclotella cf. *striata* (Kützing) Grunow	S	S
Cyclotella cf. *stylorum* Brightwell	I	I
Order Eupodiscales		
Family Odontellaceae		
Odontella aurita (Lyngbye) C.Agardh	S	I
Family Parodontellaceae		
Trieres mobiliensis (Bailey) Ashworth & E.C.Theriot	S	I
Class Coscinodiscophyceae		
Order Asterolamprales		
Family Asterolampraceae		
Asteromphalus flabellatus (Brébisson) Greville	S	S
Order Coscinodiscales		
Family Coscinodiscaceae		
Coscinodiscus asteromphalus Ehrenberg	S	I
Coscinodiscus granii L.F.Gough	S	S
Coscinodiscus wailesii Gran & Angst	S	
Family Heliopeltaceae		
Actinoptychus senarius (Ehrenberg) Ehrenberg	I	I

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Classification	Relative frequency	
	SSC	AB
Family Hemidiscaceae		
Azpeitia sp.1	S	S
Family Leptocylindraceae		
Leptocylindrus danicus Cleve	I	I
Leptocylindrus minimus Gran	I	I
Order Rhizosoleniales		
Family Probosciaceae		
Proboscia alata (Brightwell) Sundström	S	S
Family Rhizosoleniaceae		
Dactyliosolen fragilissimus (Bergon) Hasle	I	I
Dactyliosolen phuketensis (B.G.Sundström) G.R.Hasle	–	S
Guinardia delicatula (Cleve) Hasle	I	I
Guinardia flaccida (Castracane) H.Peragallo	I	I
Guinardia striata (Stolterfoth) Hasle	I	I
Neocalyptrella robusta (G.Norman ex Ralfs) Hernández-Becerril & Castillo	S	S
Rhizosolenia hebetata Bailey	S	S
Rhizosolenia hyalina Ostenfeld	S	S
Rhizosolenia styliformis T.Brightwell	S	S
Sundstroemia setigera Medlin, L.K., Boonprakob, A., Lundholm, N. & Moestrup	I	S
Sundstroemia pungens Medlin, L.K., Boonprakob, A., Lundholm, N. & Moestrup	S	I
Order Triceratiales		
Family Triceratiaceae		
Triceratium favus Ehrenberg	I	I
Order Paraliales		
Family Paraliaceae		
Paralia sulcata (Ehrenberg) Cleve	F	F
Subclass Corethrophycidae		
Order Corethrales		
Family Corethraceae		
Corethron sp.1	I	I
Class Bacillariophyta (incertae sedis)		
Order Bacillariophyta (incertae sedis)		
Family Bacillariophyta (incertae sedis)		
Neomoelleria cornuta (Cleve) S.Blanco & C.E.Wetzel	I	I
Class Bacillariophyceae		
Order Bacillariales		
Family Bacillariaceae		
Bacillaria paxillifera (O.F.Müller) T.Marsson	S	S
Cylindrotheca closterium (Ehrenberg) Reimann & J.C.Lewin	F	VF

Classification	Relative frequency	
	SSC	AB
Denticula sp.1	S	–
Fragilariopsis doliolus (Wallich) Medlin & P. A. Sims	I	I
Nitzschia sp.1	S	I
Nitzschia longissima (Brébisson) Ralfs	S	I
Nitzschia incurva var. *lorenziana* R. Ross	I	F
Pseudo-nitzschia spp	VF	VF
Pseudo-nitzschia “delicatissima complex” sp.2	I	I
Pseudo-nitzschia “*seriata* complex*”* sp.1	F	F
Pseudo-nitzschia “*seriata* complex*”* sp.2	F	F
Tryblionella sp.1	I	F
Order Cocconeidales		
Family Cocconeidaceae		
Cocconeis sp.1	I	I
Order Cymbellales		
Family Cymbellaceae		
Cymbella sp.1	S	S
Order Naviculales		
Family Diploneidaceae		
Diploneis cf. *bombus* (Ehrenberg) Ehrenberg	S	I
Diploneis didymus (Ehrenberg) Ehrenberg	–	S
Diploneis cf. *smithii* (Brébisson) Cleve	–	S
Diploneis weissflogii (A.W.F.Schmidt) Cleve	F	F
Family Naviculaceae		
Haslea wawrikae (Husedt) Simonsen	I	I
Haslea cf. *trompii* (Cleve) Simonsen	I	I
Navicula sp.1	VF	F
Family Plagiotropidaceae		
Meuniera membranacea (Cleve) P.C.Silva	I	I
Family Pleurosigmataceae		
“*Pleurosigma/Gyrosigma*” Complex	F	VF
Family Stauroneidaceae		
Stauroneis sp.1	S	I
Order Fragilariales		
Family Fragilariaceae		
Fragilaria sp.1	S	I
Order Licmophorales		
Family Licmophoraceae		
Licmophora tincta (C.Agardh) Grunow	I	F
Order Thalassiophysales		
Family Catenulaceae		
Amphora sp.1	I	F

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Classification	Relative frequency	
	SSC	AB
Order Surirellales		
Family Surirellaceae		
Stenopterobia sp.1	–	S
Surirella sp.1	–	S
Subclass Fragilariophycidae		
Order Thalassionematales		
Family Thalassionemataceae		
Lioloma pacificum (Cupp) Hasle	I	I
Thalassionema frauenfeldii (Grunow) Tempère & Peragallo	I	S
Thalassionema nitzschioides (Grunow) Mereschkowsky	VF	VF
Thalassiothrix sp.1	S	S
Subclass Urneidophycidae		
Order Plagiogrammales		
Family Plagiogrammaceae		
Plagiogramma sp.1	–	S
Order Rhaphoneidales		
Family Asterionellopsidaceae		
Asterionellopsis glacialis (Castracane) Round	I	I
Family Rhaphoneidaceae		
Delphineis sp.1	F	F
Rhaphoneis sp.1	I	F
Phylum Miozoa		
Superclass Dinoflagellata		
Class Dinophyceae		
Order Dinophysiales		
Family Dinophysaceae		
Dinophysis "acuminata/sacculus" complex	I	I
Dinophysis cf. *caudata* Kent	S	–
Dinophysis microstrigiliformis Abé	S	–
Dinophysis cf. *ovum* F.Schütt	S	–
Dynophysis tripos	I	I
Ornithocercus cf. *magnificus* Stein	–	S
Order Gonyaulacales		
Family Ceratiaceae		
Tripos cf. *declinatus* (G.Karsten) F.Gómez	I	I
Tripos azoricus (Cleve) F. Gómez	S	–
Tripos furca (Ehrenberg) F. Gómez	I	I
Tripos fusus (Ehrenberg) F. Gómez	I	I
Tripos hircus (Schröder) F. Gómez	I	I
Tripos longirostrum (Gourret) Hallegraeff & Huisman	S	S
Tripos macroceros (Ehrenberg) Hallegraeff & Huisman	S	–
Tripos cf. *massiliense* (Gourret) F.Gómez	I	I
Tripos muelleri Bory	I	I
Tripos trichoceros (Ehrenberg) Gómez	S	S
Family Gonyaulacaceae		
Gonyaulax cf. *spinifera* (Claparède & Lachmann) Diesing	F	I
Family Pyrocystaceae		
Alexandrium cf. *tamarense* (Lebour) Balech	S	S
Pyrophacus horologium F.Stein	S	S
Order Gymnodiniales		
Family Gymnodiniaceae		
cf. *Gymnodinium* sp.1	VF	F
cf. *Gymnodinium* sp.2	S	S
cf. *Gymnodinium* sp.3	S	S
cf. *Gymnodinium* sp.4	S	S
Family Gyrodiniaceae		
Gyrodinium sp.1	S	S
Order Peridiniales		
Family Heterocapsaceae		
Heterocapsa rotundata (Lohmann) Gert Hansen	F	I
Heterocapsa sp.1	I	I
Family Oxytoxaceae		
Oxytoxum scolopax F.Stein	–	S
Oxytoxum crassum J.Schiller	S	–
Corythodinium tesselatum (F.Stein) Loeblich Jr. & Loeblich III	S	–
Corythodinium constrictum (F.Stein) F.J.R.Taylor	S	–
Family Podolampadaceae		
Podolampas palmipes Stein	S	S
Family Protoperidiniaceae		
Protoperidinium crassum (Balech) Balech	S	S
Protoperidinium curtipes (E.G.Jørgensen) Balech	S	–
Protoperidinium divergens (Ehrenberg) Balech	S	S
Protoperidinium leonis (Pavillard) Balech	S	–
Protoperidinium mariebouriae (Paulsen) Balech	S	–
Protoperidinium parviventer Balech	S	S
Protoperidinium pentagonum (Gran) Balech	S	S
Protoperidinium steinii (E.G.Jørgensen) Balech	I	I

Continue...

...Continuation

Classification	Relative frequency	
	SSC	AB
Family Pyrocystaceae		
Pyrocystis lunula (F.Schütt) F.Schütt	I	–
Order Prorocentrales		
Family Prorocentraceae		
Prorocentrum balticum (Lohmann) Loeblich III	F	F
Prorocentrum gracile F.Schütt	F	F
Prorocentrum micans Ehrenberg	F	F
Prorocentrum cordatum (Ostenfeld) J.D.Dodge	F	F
Prorocentrum scutellum B.Schröder	F	F
Order Thoracosphaerales		
Family Thoracosphaeraceae		
cf. *Scrippsiella*	S	S
Scrippsiella acuminata (Ehrenberg) Kretschmann, Elbrächter, Zinssmeister, S.Soehner, Kirsch, Kusber & Gottschling	VF	F
Scrippsiella spinifera G.Honsell & M.Cabrini	I	I
Phylum Ochrophyta		
Class Dictyochophyceae		
Order Dictyochales		
Family Dictyochaceae		
Dictyocha fibula Ehrenberg	F	I
Octactis octonaria (Ehrenberg) Hovasse	I	I
Phylum Haptophyta		
Class Coccolithophyceae		
Order Coccolithales		
Family Calcidiscaceae		

Classification	Relative frequency	
	SSC	AB
Umbilicosphaera cf. *sibogae* (Weber Bosse) Gaarder	F	F
Order Syracosphaerales		
Family Calciosoleniaceae		
Calciosolenia brasiliensis (Lohmann) J.R.Young	S	S
Calciosolenia murrayi Gran	S	–
Family Rhabdosphaeraceae		
Discosphaera sp.1	I	S
Discosphaera tubifera (Murray & Blackman) Ostenfeld	S	–
Rhabdosphaera sp.1	I	S
Family Syracosphaeraceae		
Calciopappus sp.1	S	–
Syracosphaera sp.1	I	S
Syracosphaera pirus Halldal & Markali	S	–
Syracosphaera prolongata Gran ex Lohmann	S	S
Phylum Cyanobacteria		
Class Cyanophyceae		
Order Oscillatoriales		
Family Microcoleaceae		
Trichodesmium erythraeum Ehrenberg ex Gomont	I	F
Trichodesmium thiebautii Gomont ex Gomont	I	F
Order Synechococcales		
Family Pseudanabaenaceae		
Pseudanabaena sp.1	I	S
Order Nostocales		
Family Nostocaceae		
Richelia intracellularis J.A.W.F.Schmidt	I	I

index 20.0 °C; 36.36, according to Miranda, 1985) and Coastal Water (CW, salinity below 35 and temperature higher than 20.0 °C) was observed during December 2013 in both AB and SSC. The CW was a mixture of oceanic water masses and continental outflows and dominated SSC in all samplings with temporally variable thermohaline characteristics (see Ciotti et al. 2018a same dataset). Maximum concentrations of ammonia, nitrate + nitrite, phosphate, silicate, and chlorophyll, were generally observed at AB, but their average values were similar at both sampling sites (Table 1). During all surveys, we observed smaller volumes of accumulated precipitation compared to the regional climatology.

Phytoplankton densities were as high as 10^6 cel L^{-1} (Table 2). Despite the significance of picoplankton and nanoplankton for Brazilian coastal waters, it is worth mentioning that this inventory covered organisms greater than 5 µm. Diatoms were the predominant taxonomic group at both AB and SSC, with pennate diatoms representing 67% of phytoplankton species in the latter during the sampling period. Comparatively larger abundances of flagellates and armored dinoflagellates were noticeable at AB and SSC, respectively (Table 2).

The diatom genus *Thalassiosira* predominated during fall and winter (Figure 2), while *Pseudo-nitzschia* spp. prevailed during October and December 2013 (spring events) and January 2014 (summer) (Figure 2), after periods of high precipitation rates and when surface waters were warm and showed high phosphate concentrations. The highest densities of *Pseudo-nitzschia* spp. in October 2013 (10^5 10^5 cel.L^{-1}) were concurrent with the presence of cold waters (19.4°C, 35.5) near the SSC bottom (data presented in Ciotti et al. 2018a).

A total of 166 taxa were identified, with 86 genera, 129 species, 33 morphotypes, and 4 complexes, distributed in eight classes: Mediophyceae (42), Coscinodiscophyceae (23), Bacillariophyceae (37), Dinophyceae (48), Dictyochophyceae (02), Coccolithophyceae (10) and Cyanophyceae (04) (Figure 3 – frequent taxa). Of these, 148 taxa were in samples from within AB and 155 from SSC. *Trichodesmium* was frequently observed at AB, with the occurrence of *T. erythraeum* and *T. thiebautii* in the form of free trichomes, tufts, and puffs

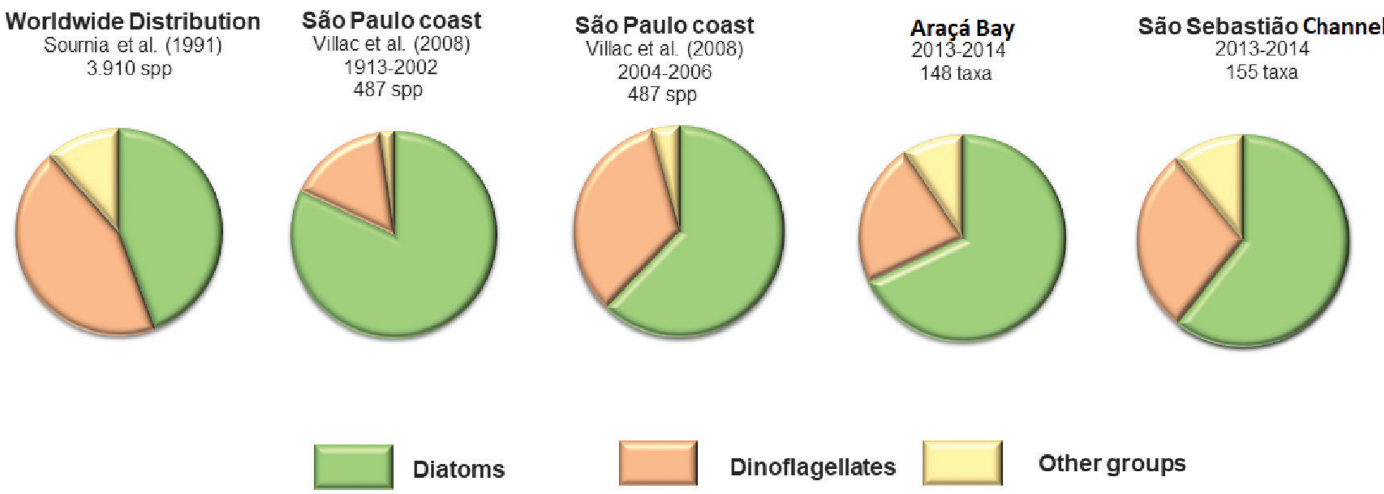

Figure 4. Relative contributions of the main taxonomic groups (diatoms, dinoflagellates and others – coccolithophorids, silicoflagellates, cyanobacteria, among others) at different levels: worldwide distribution (Sournia et al. 1991), data from 1913–2002 for the state of São Paulo (Villac et al. 2008), data from 2004–2006 for the state of São Paulo (Villac et al. 2008), data from 2013–2014 for Araçá Bay (AB) and sampling site at São Sebastião Channel (CSS). (Figure adapted from Villac et al. 2008).

(Figure 3) The cyanobacterium *Richelia intracellularis* was observed in association with diatoms of the genus *Hemiaulus* only, for the species *H. hauckii, H. sinensis*, and *H. membranaceus* predominantly and with up to 4 trichomes of *R. intracellularis* (Figura 3).

The richness of the classes differed slightly between the two sites, as did the relative frequency of each taxon (Table 3). Diatoms were the most frequent (AB = 68%; SSC = 60%), with the class Mediophyceae having the highest percentage in AB (28%; 41 taxa) and SSC (25%; 39 taxa), with the genus *Thalassiosira* being very frequent (AB = 100%; SSC = 98%). Although the frequencies of classes Coscinodiscophyceae (AB = 15%; SSC = 14%), Dictyochophyceae (AB = 1%; SSC = 1%), Coccolithophyceae (AB = 4%; SSC = 6%) and Cyanophyceae (AB = 3%; SSC = 3%) did not vary notably between sites, the taxa from these classes displayed infrequent or sporadic occurrences, with the exceptions for the frequent diatom *Paralia sulcata* and the coccolithophorid of the genus *Umbilicosphaera* at both sites, *Dictyocha fibula* at CSS and cyanobacteria of the genus *Trichodesmium* at AB. The class Dinophyceae (AB = 24%; SSC = 30%) showed a larger percentage contribution and numerical richness at SSC (46 taxa) than at AB (36 taxa), with the species *Scrippsiella acuminata* showing the highest frequency. The species *Cylindrotheca closterium* and *Thalassionema nitzschioides*, of the class Bacillariophyceae (AB = 25%; SSC = 21%), had higher frequencies in AB and the genus *Pseudo-nitzschia* in both locations (AB = 82%; SSC = 70%).

Overall, our inventory showed that diatoms and dinoflagellates represented together, over 80% of the total (Figure 4), similar to what was presented by Villac et al. (2008) for the coast of São Paulo state (diatoms 62%, dinoflagellate 34%).

Discussion

Our results are analogous to those presented by Villac et al., (2008), who reported 193 distinct taxa over a longer extension of the São Paulo coast (between Cananéia and Ubatuba) from 2004 to 2006. Their inventory included 120 diatoms, 65 dinoflagellates, and 3 silicoflagellates. In the present study, however, we observed larger contributions of the diatom genera *Pseudo-nitzschia, Thalassiosira, Chaetoceros, Hemiaulus, Cyclotella, Coscinodiscus, Guinardia, Rhizosolenia, Thalassionema, Cylindrotheca,* and *Leptocylindrus*, and the dinoflagellate genera *Prorocentrum, Scrippsiella, Tripos, Gymnodinium, Dinophysis,* and *Heterocapsa*. One addition to Villac et al. (2008) inventory was the diazotrophic cyanobacteria *Richelia intracellularis* (unfrequent taxa), either free or in symbiosis with diatoms at both AB and SSC. Although this result can be partially related to our sampling design, differences in environmental conditions between the two studies cannot be discarded, reinforcing the importance of frequent assessments of phytoplankton genera or species.

The 5 to 6-week interval observations revealed some temporal distinctions in the taxonomic composition of the phytoplankton. For example, the diatom genera *Thalassiosira* and *Pseudo-nitzschia* were consistently frequent (Table 3, Figure 2). However, their abundances tended to alternate. In addition, *Thalassiosira* (class Mediophyceae) was frequent when taxa richness was high, while when *Pseudo-nitzschia* (class Bacillariophyceae) was predominant, the richness of taxa was low, and their highest abundances occurred synchronically to intrusions of South Atlantic Central Water in SSC.

In temperate marine ecosystems, the succession between dominant phytoplankton taxa tends to be seasonal, leading to blooms (e.g., Cui et al. 2018, Fragoso et al. 2021). The genus *Pseudo-nitzschia*, with about 55 species (Guiry & Guiry 2021), can form blooms in coastal regions globally (e.g., Trainer et al. 2012), and some species are known to be potentially harmful by producing the neurotoxin domoic acid (Hasle 2002). The genus *Thalassiosira* contains more than 100 species (Round et al. 1990), but as for *Pseudo-nitzschia* and other genera of the class Bacillariophyceae, such as *Navicula*, *Pleurosigma,* and *Gyrosigma*, species-level identification requires scanning electron microscopy.

Our results suggest not only the establishment of urgent monitoring programs for harmful algal blooms (HABs) given the frequent potential species year-round at both sites but also that these programs need to encompass proper techniques for distinguishing taxa, as species identification by optical microscopy alone is incomplete (e.g., Hoppenrath et al. 2007, Hamsher et al. 2011, Fernandes et al. 2014, Sterrenburg et al. 2015).

In our study area, the typical physical accumulations of phytoplankton cells nearshore can episodically include organisms that advect from the open ocean guided by winds (Lugomela et al. 2002), which may be the case for the diazotrophic cyanobacteria *Trichodesmium* spp. and *Richelia intracellularis* at both sampling sites. Slicks of the genus *Trichodesmium* are commonly observed in surface waters of the Brazilian Current (Detoni et al. 2016) or in inner shelf waters (< 50 m) during the summer (Brandini et al. 1989). The occurrences could be linked to the relatively low nitrogen input from the continent, as the observations took place during a dry period (Tocci 2016), favoring the growth of diazotrophic cyanobacteria. However, at least for *Trichodesmium*, the advection of waters from offshore by mesoscale winds (Castro Filho & Mirada 1998) could be a source of these organisms for the coast. Moreover, favorable upwelling winds will favor intrusions of the South Atlantic Central Water in the SSC, not only enhancing the local concentration of nutrients and primary production rates (Regaudie et al. 2017) but also transporting diatoms, such as *Pseudo-nitzschia*, that impacted the overall taxa richness. These results indicate the need for future phytoplankton monitoring programs assessing the offshore contribution of water masses to SSC.

The observation of unfrequent taxa of tycopelagic diatoms (Table 1) at AB and SSC included the predominance of *Cylindrotheca closterium, Diploneis weissflogii*, and *Thalassionema nitzschioides.* The Bacillariophyceae *Cocconeis* sp.1, *D. didymus, D.* cf. *smithii*, *Licmophora tincta, Delphineis* sp.1*, Rhaphoneis* sp.1, and *Surirella* sp.1 showed larger densities at AB than SSC, and some species only observed at AB, such as *Diploneis didymus, Diploneis* cf. *smithii, Stenopterobia* sp.1, *Surirella* sp.1, and *Plagiogramma* sp.1, probably a result from the bay hydrodynamics that due to its shallower depth (Siegle et al. 2018) allows organisms to resuspend to the water column during each tidal cycle. This continuous exchange of phytoplankton organisms between the sediments of the bay and SSC water needs further evaluation for a better description of this ecologically important system.

Note that some of the identified taxa are mentioned in the literature as non-toxin-producing bloom formers (Odebrecht et al. 2001, Hallegraeff et al. 2003, Moestrup 2004, Villac et al. 2008), such as the diatoms *Asterionellopsis glacialis, Cerataulina pelagica, Cylindrotheca closterium, Guinardia delicatula, Leptocylindrus minimus*, and *Chaetoceros* spp.; the dinoflagellates *Tripos fusus* and *Tripus hircus*; and the silicoflagellate *Dictyocha fibula, may alternatively bloom*. However, accumulations of these species may result in many other ecologic and economic impacts (Castro et al. 2016). Additionally, results also reveal the lower diatom diversity when the genus *Pseudo-nitzschia* was abundant, which occurred during SACW intrusions in the channel.

Our results stress the demand for the urgent implementation of monitoring programs that aid public policies for environmental safety. The occurrences of taxa are known to be potentially harmful, highlighting the dinoflagellates of the genera *Alexandrium*, cf. *Gymnodinium*, *Dinophysis*, *Gonoyaulux*, and *Prorocentrum* are unsettling. Although some initiatives are in place, our results demonstrate the need for a comprehensive monitoring program that includes modernized methodologies and hydrodynamical modeling. If conditions for blooming are favored with nutrients input by sewage and warming of seawater temperatures, they may cause fish death, mollusk poisoning, and several public health problems (Hallegraeff et al. 2003).

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Author Contributions

B.R.C. Tocci: Contribution to data collection; Contribution to data analysis and interpretation; Contribution to manuscript preparation.

G.A.O. Moser: Contribution to manuscript preparation; Contribution to critical revision, adding intelectual content.

A.M. Ciotti: Substantial contribution in the concept and design of the study; Contribution to data collection; Contribution to data analysis and interpretation; Contribution to manuscript preparation; Contribution to critical revision, adding intelectual content.

Conflicts of Interest

The authors declare that they have no conflict of interest related to the publication of this manuscript.

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