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**ESTUDO DE INTERAÇÃO ENTRE FITOPLÂNCTON E MICROPLÁSTICO NA
BAÍA DE TAMANDARÉ, PE, BRASIL**

Recife
2020

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TAMANDARÉ, PE, BRASIL**

Dissertação apresentada à Coordenação de Pós-Graduação em Oceanografia da Universidade Federal de Pernambuco (UFPE), como requisito parcial à obtenção do título de Mestre em Oceanografia.

Área de concentração: Oceanografia Biológica

Orientadora: Profa. Dra. Maria da Glória Gonçalves da Silva Cunha

Coorientador: Dr. Pedro Augusto Mendes de Castro Melo

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RESUMO

A comunidade fitoplanctônica e os microplástico foi estudada na Baía de Tamandaré (Pernambuco, nordeste do Brasil) em três ambientes (baía, pluma e recife), para identificar a variação quantitativa (sazonal e espacial) associado aos microplásticos. As amostras foram coletadas com uma rede de plâncton (20 μm) para a análise qualitativa e com garrafa de Kitahara para análise quantitativa, em dois períodos sazonais, estação seca (novembro / 2017, fevereiro e outubro / 2018) e estação chuvosa (abril, julho e agosto / 2018). Em relação à estrutura do fitoplâncton, o filo Bacillariophyta apresenta maior riqueza, seguido por Miozoa e Cyanobacteria. Dentre os táxons identificados, *Navicula* spp., *Trichodesmium* sp. e *Coscinodiscus* spp. foram dominantes. A densidade celular variou entre 1.000 cel.L⁻¹ (estação seca) a 324.000 cél.L⁻¹ (estação chuvosa). A área mais representativa foi o recife (61.555,55 cel.L⁻¹), seguido pela baía (53.611,11 cel.L⁻¹) e pluma (25.333,33 cel.L⁻¹). Foram encontrados microplásticos com formato filamentoso e nas cores transparente, azul, vermelha e preta. A proporção célula do fitoplâncton e microplástico é de 6:1. A maior concentração de microplásticos é na pluma e na baía, com partículas aderidas ao fitoplâncton. Os resultados confirmam uma estrutura fitoplanctônica semelhante aos demais ecossistemas tropicais e a ocorrência de microplásticos nas amostras de plâncton, indicando a fragmentação e o transporte do rio dessas partículas na área pesquisada. Estudos complementares são necessários para verificar se existe algum padrão na distribuição e proporção de microplásticos em ambientes semelhantes e diferentes ao do local do estudo atual, e se afetam as espécies de fitoplâncton.

Palavras-chave: Vida marinha. Poluição. Plástico. Microalgas.

ABSTRACT

The phytoplankton community and microplastics was studied in Tamandaré Bay (Pernambuco, northeastern Brazil) in three environments (bay, plume and reef), to identify the quantitative variation (seasonal and spatial) of phytoplankton associated with microplastics. The samples for phytoplankton were collected, they were collected with a plankton net (20µm) for the qualitative analysis and with a Kitahara bottle for quantitative analyses during dry (November/2017, February, October/2019) and rainy season (April, July and August/2018), every two months. In relation to the phytoplankton structure, the Bacillariophyta shows the most richness, followed by Miozoa and Cyanobacteria. Among the taxa *Navicula* spp., *Trichodesmium* sp. and *Coscinodiscus* spp. were dominants. The cell density varied between 1,000 cell.L⁻¹ (Dry season) to 324,000 cell.L⁻¹ (Rainy season). The most representative area was the reef (61,555.55 cell.L⁻¹), followed by bay (53,611.11 cell.L⁻¹) and plume (25,333.33 cell.L⁻¹). Were found microplastics with filamentous shape and in transparent, blue, red, and black colors. The bay area had the greatest number of plastic particles, followed by plume and reef and this could be explained due the transport through the river and plastic fragmentation. The ratio between the number of phytoplankton and microplastic is approximately 6:1. Microplastics adhered to phytoplankton was observed. results confirm a phytoplankton structure similar to the other tropical ecosystems and the occurrence of microplastics in plankton samples indicating the fragmentation and transport of these particles in the study area. Complementary studies are needed to verify if there is a pattern of distribution of microplastics and how they affect the phytoplankton species in Tamandaré Bay.

Keywords: Marine life. Pollution. Plastic. Microalgae.

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1 INTRODUÇÃO

O fitoplâncton é um grupo polifilético formado por organismos microscópicos unicelulares (algas, cianobactérias e bactérias), adaptados a viverem em ambientes aquáticos, sejam em águas continentais ou no meio marinho. Estima-se que existam cerca de quatro mil espécies de organismos do fitoplâncton marinho, apresentando uma grande diversidade de formas e tamanhos (REYNOLDS, 2006).

A diversidade na forma e tamanho do fitoplâncton está relacionada a sua capacidade de utilização de recursos e à susceptibilidade a processos de perda, podendo então as espécies ser classificadas de acordo com suas propriedades funcionais (REYNOLDS *et al.*, 2002; WEITHOFF, 2003; NASELLI-FLORES *et al.*, 2007; FINKEL *et al.*, 2010; KRUK *et al.* 2010).

É importante conhecer quais fatores governam sua dinâmica, já que o fitoplâncton é responsável pela maior parte da produção primária dos ambientes aquáticos e é indispensável para manter o ecossistema marinho equilibrado, além de serem responsáveis pela manutenção da vida como um todo através da participação nos ciclos biogeoquímicos (NAIR *et al.*, 2008).

O uso desses organismos como bioindicadores é eficaz, devido a sensibilidade e rápida respostas às alterações físico-químicas no ambiente, substâncias tóxicas e poluentes que esses seres vivos apresentam (PARMAR *et al.*, 2016). O plástico é um dos diversos poluentes presentes que o fitoplâncton tem contato.

Plástico é um produto bastante utilizado pela humanidade, devido às suas propriedades: Leve, resistente e barato. Em 2015, aproximadamente 6.300 toneladas de plásticos foram geradas, destes, 9% foram reciclados, 12% foram incinerados e os 79% restantes foram acumulados em lixões ou no meio ambiente. Se a produção e o descarte de plásticos atual continuar a crescer, em torno de 12.000 toneladas de lixo plástico estará em lixões ou no ambiente natural por volta do ano 2050. (GEYER *et al.*, 2017).

Esses materiais são despejados de forma indevida e assim contaminam o ambiente marinho; 80% dessa poluição é feita através de lixo terrestre, principalmente os deixados em praias (ANDRADY, 2011).

Esses poluentes podem chegar ao meio marinho por ser carregados pelos rios ou sistemas de drenagens das cidades, e pelo descarte ou perda em transportes aquáticos (GREGORY, 1977). THORNTON & JACKSON, 1998, avaliaram na praia de Clifford, na nova Jersey, diversos tipos de detritos com forma, tamanho e estado de fragmentação diferente, e foi observado que esses plásticos eram carregados pelo vento.

A poluição por plástico é um problema mundial que afeta todo o ambiente marinho, com maior concentração de poluentes em regiões costeiras e em giros oceânicos (COSTA, 2009), mas é possível encontrar esses poluentes em sedimentos e corais no fundo marinho do Mar Mediterrâneo (WOODALL et al., 2014), no Ártico, e em profundidade variando de 2340 a 5570 m (BERGMANN et al., 2017).

Esses materiais plásticos causam diversos danos a uma diversidade de organismos tais como aves, mamíferos, répteis e peixes (CARPENTER *et al.*, 1972; ROTHSTEIN, 1973; BARROS, ODELL & PATTON, 1990; MOSER e LEE, 1992; SECCHI e ZAZZUR, 1999; TOMÁS et al., 2002), tais como: movimentação limitada, ferimentos externos e internos, obstrução do trato digestivo, perda de apetite, alterações hormonais e problemas na reprodução (LAIST, 1987; SHAW e DAY, 1994).

Os poluentes plásticos podem apresentar dimensões pequenas, variando de 1 micrometro a 5 milímetro, e com esse tamanho são chamados de microplásticos (FRIAS & NASH, 2019).

O microplástico apresentar dois tipos de origem nos meios aquáticos. Os microplásticos de origem primária são aqueles que já são produzidos com tamanhos minúsculos, como pellets virgens que serão utilizados para o desenvolvimento de objetos plásticos cotidianos, e partículas em cosméticos, na indústria têxtil ou em abrasivos, e são descartados indevidamente ou carreados pelo sistema de esgoto para o meio ambiente (COLE et al., 2011; VAN WEZEL et al., 2016).

Os microplásticos de origem secundária são resultantes da fragmentação de objetos plástico presentes no ambiente marinho, através de fotodegradação pela radiação ultravioleta do sol e pela abrasão das ações das ondas (COLE et al., 2011). Essas partículas podem sofrer fragmentação até tamanhos menores que 1 micrômetro, e nesses tamanhos essas partículas podem ser classificadas como nanoplástico (LAMBERT & WAGNER, 2016; TER HALLE et al., 2017).

Os microplásticos são capazes de serem mais danosas do que os materiais maiores, por conseguirem reter poluentes e lançar aditivos químicos, como hidrocarbonetos policíclicos aromáticos (PAH) e bifenil policlorado (PCB) (ENDO et al., 2005; MATO et al., 2001; RIOS et al., 2007), para o meio ambiente, contaminando os locais e os seres vivos, quando estes ingerem essas partículas. Metais pesados também podem adsorver na superfície do microplástico (BRENNECKE et al., 2016).

Essas partículas também são capazes de causar os mesmos danos que plásticos maiores em organismos planctônicos, filtradores, detritívoros ou de menor tamanho (WRIGHT,

THOMPSON & GALLOWAY, 2013; ANDRADY, 2011; MATO *et al.*, 2001; FRIAS, MARTINS e SOBRAL, 2011). A ingestão de pequenas partículas plásticas e das substâncias tóxicas adsorvidas nesses materiais é a forma mais comum pelo qual esses poluentes afetam os organismos (COLE *et al.*, 2013).

Danos às populações (Fitoplâncton) também podem ser causados pelos poluentes plásticos, já que esses materiais podem agir como vetores de microalgas, por servir de substratos para esses organismos que podem formar florações nocivas ou não (MASÓ *et al.*, 2003).

Através do aumento na densidade dessas partículas, devido aos seres vivos fixados ou aderidos, ocorre o transporte vertical desses agregados, levando-os para locais onde normalmente não ocorrem e tornando-os disponíveis para os seres vivos que vivem em maiores profundidades ou bentônicos (WRIGHT, THOMPSON & GALLOWAY, 2013).

As interações entre o fitoplâncton e o microplástico, e os impactos decorrente dessas interações, foram majoritariamente observadas em estudos de laboratório com cultivo de microalgas (PRATA *et al.*, 2019). Foi observado que as partículas se aderem a superfície dos organismos causando danos a estrutura das células ou interferindo o metabolismo dos organismos, e são capazes de se agruparem em agregados (CHEN *et al.*, 2011; BHATTACHARYA *et al.*, 2010; SJOLLEMA *et al.*, 2016).

O presente trabalho procura adquirir mais informações acerca dessa interação, com amostras coletadas *in situ*, em três áreas da baía de Tamandaré, localizada no litoral sul do estado de Pernambuco (Brasil), para testar a hipótese de que a quantidade de microplástico é inversamente proporcional ao de fitoplâncton, ou seja, nos pontos em que o número de partículas for maior, terá menos células, mas ainda assim que o número de partículas seja significativamente menor que o número de células.

2 OBJETIVOS

Para o desenvolvimento do presente trabalho, foram elaborados objetivos, geral e específicos, para direcionar e metrificar o progresso do estudo, buscando responder as hipóteses levantadas.

2.1 OBJETIVO GERAL

Este trabalho teve como objetivo avaliar a comunidade fitoplanctônica e o microplástico presente na Baía de Tamandaré, Pernambuco, através do levantamento qualitativo e quantitativo do material de interesse nas amostras coletadas e da sua distribuição espacial e sazonal, observando a relação entre esses dois materiais, como a ocorrência de partículas aderidas às microalgas ou presentes em agregados de matéria orgânica.

2.2 OBJETIVOS ESPECÍFICOS

- a) Descrever a comunidade fitoplanctônica em níveis específicos e infraespecíficos, evidenciando a estrutura da comunidade através da sua abundância relativa, frequência de ocorrência, índice de diversidade específica e equitabilidade;
- b) Delinear os parâmetros ambientais abióticos e bióticos presente na área de estudo;
- c) Identificação do número de partículas do material não-vivo, analisando as suas características morfológicas, abundância e frequência de ocorrência;
- d) Uso de proporção matemáticas e testes estatísticos para relacionar os valores de número de células de fitoplâncton e partículas do microplástico sazonal e espacialmente.

3 ESTRUTURA DA DISSERTAÇÃO

De acordo com os objetivos e resultados obtidos ao longo da realização do presente estudo, esta dissertação está composta de um capítulo, que se refere a artigo científico e segue as normas de publicação da revista selecionada.

Capítulo 1: “PHYTOPLANKTON AND MICROPLASTICS IN TAMANDARÉ BAY, PERNAMBUCO, BRAZIL”

Este capítulo está submetido na revista científica **Marine Biology Research (ISSN: 1745-1000)**. Este estudo avaliou a interação do material pelágico vivo (Fitoplâncton) e não vivo (Microplástico) na Baía de Tamandaré (Pernambuco, Nordeste do Brasil), em três ambientes (baía, pluma e recife), em variação espacial e temporal, como também a ocorrência de partículas aderidas às microalgas ou formando agregados.

4 ARTIGO 1 - PHYTOPLANKTON AND MICROPLASTICS IN TAMANDARÉ BAY, PERNAMBUCO, BRAZIL

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Abstract: The phytoplankton community was studied in Tamandaré Bay (Pernambuco, northeastern Brazil) in three environments (bay, plume and reef), to identify the quantitative variation (seasonal and spatial) of phytoplankton associated with microplastics. The samples for phytoplankton samples were collected every two months, during the dry and rainy season from November/2017 to October/2018. To collect it was used a plankton net (20µm) for the qualitative analysis and with a Kitahara bottle for quantitative analyses. In relation to the phytoplankton structure, the Bacillariophyta shows the most richness, followed by Miozoa and Cyanobacteria. Among the taxa *Navicula* spp., *Trichodesmium* sp. and *Coscinodiscus* spp. were dominants. The cell density varied between 1,000 cell.L⁻¹ (Dry season) to 324,000 cell.L⁻¹ (Rainy season). The most representative area was the reef (61,555.55 cell.L⁻¹), followed by bay (53,611.11 cell.L⁻¹) and plume (25,333.33 cell.L⁻¹). Were found microplastics with filamentous shape and in transparent, blue, red, and black colors. The bay was the area with the greatest number of plastic particles, followed by plume and reef. The ratio between the number of

phytoplankton and microplastic is approximately 6:1. Microplastics adhered to phytoplankton was observed. The plume and bay presented the higher concentration of microplastics and this could be explained due the transport through the river and plastic fragmentation. The results confirm a phytoplankton structure similar to the other tropical ecosystems and the occurrence of microplastics in plankton samples indicating the fragmentation and transport of these particles in the study area. Complementary studies are needed to verify if there is a pattern of distribution of microplastics and how they affect the phytoplankton species in Tamandaré Bay.

Keywords: Marine Life, Pollution, Plastic, Microalgae

Introduction

Phytoplankton is composed of single-celled organisms capable of photosynthesis, so it is one of the responsible for the primary marine coastal production, being the basis of the flow of nutrients and energy in the trophic web in marine and estuarine ecosystems (Eskinazi-Leça et al. 1980; Grego et al. 2004).

The phytoplankton community is used as an indicator of the physicochemical conditions of aquatic ecosystems, through variations in the composition, biomass, and productivity of the species, which respond quickly to the changes of various abiotic and biotic factors present in the environment (Aquino et al. 2014). One of the factors that can affect the behavior of microalgae is microplastics.

Microplastics are defined as particles ranging from 1 micrometer to 5 millimeters in length (Frias and Nash 2019). This plastic debris can be classified according to their origin into primary when they are originally produced with reduced dimensions and are discarded in aquatic environments directly, or secondary resulting from fragmentation of larger plastics through ultraviolet radiation and action of waves and tides (Horton and Dixon 2017), or fibers released by clothes or fishery gear.

This debris accumulates in the water and sediment, polluting the coastal regions and the oceans with a probability of interaction with a variety of organisms since plankton to the mammals (Frias et al. 2011; Ivar do Sul and Costa 2014), causing physical damage to the animals and retaining toxic substances, which when ingested, can transfer contaminants along with the trophic web (Mato et al. 2001; Teuten et al. 2009).

These plastics debris can also be used as a substrate for sessile organisms, transporting them to other locations. This type of association increases the density of pollutants and makes them available to living beings from deeper regions (Wright et al. 2013).

Most of the research about the interaction between microplastics and phytoplankton has been carried out in laboratories, and it has been discovered that these particles can cause damage in various ways when the microalgae adhere to the surface of the microplastic, interfering in the algae metabolism (Besseling et al. 2014; Bhattacharya et al. 2010; Yokota et al. 2017; Zhang et al. 2017).

There are few studies relating to the phytoplankton community and microplastics (Masó et al. 2003; Silva Cunha et al. 2019) mostly in tropical regions.

Previous studies on phytoplankton carried out on Tamandaré beach and in the adjacent estuarine areas addressed the composition, density, and biomass (Moura and Passavante 1994/1995; Rosevel da Silva et al. 2005). There is no record of studies relating to phytoplankton and microplastics in this region.

Magalhães and Araújo (2012) observed that more than 50% of the waste present on Tamandaré beach (Pernambuco, Northeastern Brazil) is formed by plastic debris, and about one-third of these were found in fragmented forms, with size ranging from 5 to 10 cm in diameter.

In this context, the current research aims to identify the quali-quantitative variation (seasonal and spatial) of phytoplankton associated with microplastics in Tamandaré Bay. For this, testing the hypothesis that the amount of non-living material (microplastics) is inversely proportional to living material (phytoplankton), and it is expected that the number of particles is not significantly higher than the number of phytoplankton cells. It is also intended to observe the form of interaction between plastic debris and phytoplankton, through if particles occur adhered to microalgae or form aggregates, considering that this information allows evaluating the possibilities of microplastics become available to the organisms in the trophic web.

Material and Methods

Study area

The Tamandaré bay located in the municipality of Tamandaré, south coast of the state of Pernambuco (latitudes 8°42'602" S and 8°46'671" S and longitudes 35° 7' 29" W and 35° 2' 28" W), approximately 110km from the capital, Recife.

It is characterized by a semicircular shape, with the concavity facing the Atlantic Ocean, representing one of the most important ecosystems of the state coast (Moura 1991).

It is formed by coastal rivers of reduced dimensions, including the rivers Formoso, Mamucaba, and Ilhetas (Santos-Filho 1969; Maida and Ferreira 1997), and areas composed of Atlantic forest, mangroves, marine phanerogams meadows and a continental shelf formed by

coral reefs and beach rocks, which support a varied marine fauna and flora creating a place utilized for fishing and tourism. Because of this, it is recognized as a biodiversity hotspot.

The entire area is inserted into two overlapping conservation units (APA de Guadalupe and APA Costa dos Corais), reflecting the need for protection of this environment, aiming at a healthy human occupation (Barbosa et al. 2016).

The Long-Term Ecological Research Programme (PELD), with PELD-TAMS, is developed at the Tamandaré bay, which comprises an area of 216,574.06 hectares, which extends from the coast to the slope, to understand the patterns and processes present in the area, its temporal and spatial variations, the structure of populations and communities, and fundamental factors for the resilience of the site in the face of environmental changes and anthropic impacts, and thus assist and develop strategies necessary for the maintenance and conservation of biodiversity and the sustainable use of resources, with the participation of local society.

Sampling

In Tamandaré bay, samples were collected every from November/2017 to October/2018. The sampling was realized in two seasonal periods, dry season (November/17, February, and October/18) and rainy season (April, July, and August/18), both at low tide. Three points at three sampling sites were selected: the first, to the north, in the interior of the bay, with minimal influence of the river plume (B1, B2, B3); the second, at the southern end of the bay, with the direct influence of the estuary plume formed by the Ilhetas and Mamucaba rivers (P1, P2, P3); and the last, next to the first line of reefs (R1, R2, R3) (Figure 1).

The samples were collected with a plankton net (20µm), with a towing time of 3 minutes and fixed with neutral formaldehyde at 4% for qualitative analysis. For quantitative analyses, the samples were performed with a Kitahara bottle and fixed with Lugol at 2%. The material obtained was filed in the Laboratory of Phytoplankton (LABFITO) belonging to the collection of the Department of Oceanography of the Federal University of Pernambuco.

Laboratory analysis

The analysis of qualitative samples was realized with the aid of the AXIOSKOP microscope provided with photomicrography equipment. The identification of the phytoplankton was made up to the lowest possible taxonomic level. The identification of taxa was made through specialized bibliography (Balech 1988; Chreti  not-Dinnet et al. 1990; Per  gallo and Per  gallo 1897-1908; Prescott 1978; Tomas 1997; Hoppenrath et al. 2009). The

taxonomic classification and scientific names were checked through the international database AlgaeBase (Guiry and Guiry 2020).

The analysis of the microplastic composition in the collected samples was focused on morphological characteristics of the particles (Color, Length and Shape). To minimize the contamination of the samples and the equipment used in the analysis, some precautions were taken: The processing area was kept clean and free from dust or any kind of particles. The material used, like the slides, cover slip and the sedimentation chamber were washed with distilled water and let to air dry.

The quantitative analysis followed the methodology described by the Utermöhl (Hasle 1978; Edler 1979). After homogenization, 10 ml of each sample were placed in sedimentation chambers, colored with Bengal Rose, and left for sedimentation for 24 hours. The counts were performed in an increase of 450 X, with the aid of an inverted microscope (Zeiss, AXIOVERT). This microscope was adapted with phase contrast and ocular with a micrometric ruler. The count of the entire sedimentation chamber was standardized. The results were expressed in cells per liter (cell.L^{-1}), colonial organisms were considered as a counting unit, according to the formula proposed by Ferrario et al. (1995), for phytoplankton, while microplastics were expressed in the number of particles per liter.

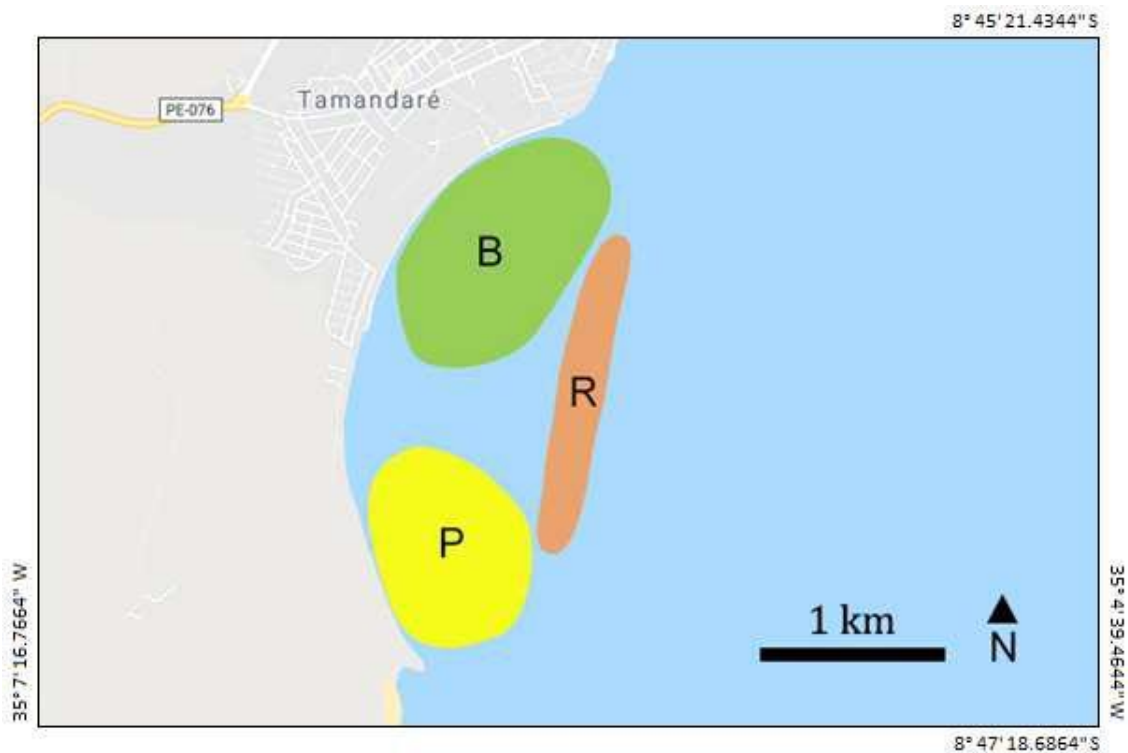


Figure 1 - Study Area, Tamandaré bay with representation of the sampling sites, where the green area (B) is the region within the Bay, the yellow area (P) is the region with influence of the river plumes and the orange area (R) is the reef region. Source: Google Maps modified by author, 2020.

Analysis of environmental data

Chlorophyll *a* concentration was measured using the spectrophotometry method (UNESCO 1997).

Environment data was assessed with a Secchi Disk and a multiparameter equipment (Horiba U52) with sensors for temperature, pH, salinity, transparency, turbidity and total dissolved solids. The rainfall during the samplings was provided by the Pernambuco Water and Climate Agency (APAC).

Numerical analysis

The specific richness of phytoplankton was considered as the number of taxa present in each sample. A similar methodology was applied for microplastics, considering the total number of particles found in each sample.

The relative abundance was calculated following Lobo and Leighton (1986), where the phytoplankton species were classified as Dominant (occurrence above 50%), Abundant (occurrence higher than the average value of individuals in the sample), and Rare (occurrence below the average value of individuals). It was applied the same methodology to microplastics.

The frequency of occurrence was calculated from the number of samples in which each taxon and plastic debris occurred, and the total number of samples analyzed (Mateucci and Colma 1982). Based on this calculation, species and microplastics were classified into four categories: Very Frequent ($FO > 70\%$), Frequent ($30 < FO \leq 70\%$), Infrequent ($10 < FO \leq 30\%$), and Sporadic ($FO \leq 10\%$).

Subsequently, the species diversity index was calculated for each sample, according to the Shannon-Weiner index (Shannon 1948), with the data expressed in $\text{bits} \cdot \text{cel}^{-1}$ and the evenness calculated according to Pielou (1967). Species diversity and evenness were calculated using the Microsoft Excel program and considering all identified taxa. This indicated the degree of complexity of the community structure and how distributed it was.

To structure the phytoplankton community, the ecological characteristics of each taxon identified at a specific level were considered. The classification followed the works of Honorato da Silva et al. 2009 and Sousa et al. 2008. The species were divided into Planktonic (they live suspended in the water column); Tichoplanktonic (benthic, but can be taken to the water column); Marine (present in the marine environment); Neritic (live on the continental shelf); Oceanic (live in the open sea); Estuarine (found in estuaries) and Freshwater (inhabit freshwater environments).

Simple mathematical ratios were calculated to evaluate the proportion between the amount of microplastics particles and cells from the phytoplankton in the study area.

Differences between phytoplankton species richness and the number of microplastics were compared through statistical tests, where the significance level was $p < 0.05$. The Kruskal-Wallis test is a nonparametric test used to determine whether there are significant differences between two or more groups of independent variables belonging to a set. Spearman Correlation is a nonparametric correlation measure that describes the relationship between two variables, without making any assumptions about the frequency distribution of the variables.

Results

Environmental data

The highest averages found in the rainy season were Tide and Dissolved oxygen, while the other parameters had a higher mean in the dry season. In the bay, the highest averages were in the Water temperature, pH, Dissolved oxygen, and Total suspended solids. In the plume, just Turbidity had the highest average, while in the reef area was the Salinity (Table I).

Table I - Mean values and standard deviation (Mean $\pm$ SD) of rainfall (mm), water temperature ($^{\circ}$ C), tide, pH, salinity (ppt), dissolved oxygen (mL.L $^{-1}$), total suspended solids (TSS), and turbidity (NTU), from November/2017 to October/2018, in Tamandaré Bay, Pernambuco, Brazil. Note: * = $p < 0.05$ (significant variation).

	Enviroments			Seasonal periods		P-value	
	Bay	Plume	Reef	Dry	Rainy	Spatial	Seasonal
Rainfall	147.37			149.73	147.37	-	-
Tide	0.21			0.17	0.21	-	-
Temperature	27.91 $\pm$ 1.08	27.82 $\pm$ 0.76	27.58 $\pm$ 0.60	28.34 $\pm$ 0.76	27.54 $\pm$ 0.82	p=0.98	p=0.006*
pH	7.98 $\pm$ 0.64	7.66 $\pm$ 0.59	7.86 $\pm$ 0.55	8.20 $\pm$ 0.09	7.68 $\pm$ 0.67	p=0.48	p=0.002*
Salinity	31.05 $\pm$ 1.88	27.63 $\pm$ 2.21	32.93 $\pm$ 1.79	31.83 $\pm$ 3.95	30.02 $\pm$ 1.70	p=0.0001*	p=0.15
Dissolved Oxygen	5.65 $\pm$ 1.32	4.63 $\pm$ 1.31	5.52 $\pm$ 1.99	4.34 $\pm$ 0.99	5.72 $\pm$ 1.55	p=0.50	p=0.001*
TSS	57.18 $\pm$ 29.21	49.83 $\pm$ 17.56	31.36 $\pm$ 6.53	46.52 $\pm$ 12.47	45.86 $\pm$ 23.96	p=0.14	p=0.05
Turbidity	3.52 $\pm$ 2.85	6.40 $\pm$ 2.52	2.23 $\pm$ 1.39	3.31 $\pm$ 2.44	4.32 $\pm$ 3.01	p=0.006*	p=0.006*

Water temperature, pH, dissolved oxygen, and turbidity showed significant seasonal variation, while salinity and turbidity showed significant variations between the sampling stations ($p < 0.05$) (Table I).

Phytoplankton structure

The phytoplankton community presented a total of 113 taxa distributed in the phyla: Bacillariophyta (69.91%), Miozoa (21.24%), Cyanobacteria (4.42%), Chlorophyta (1.77%), Euglenozoa (1.77%) and Ochrophyta (0.88%). During the seasonal periods, the community was represented for 97 taxa (rainy season) and 91 taxa (dry season). In relation to the environment, in the bay area occurred 82 taxa, in the plume 97 taxa and in the reef 84 taxa. The presence of

all phyla was observed during the rainy season and in bay and plume area (Table II). All the taxa are listed in the Appendice A.

Table II – Distribution of phytoplankton community during the seasonal periods (Dry and Rainy seasons) in three different environments (Bay, Plume, and Reef) in Tamandaré bay, Pernambuco, Brazil.

	Environments			Seasonal periods	
	Bay	Plume	Reef	Dry	Rainy
Bacillariophyta	56	69	66	69	67
Miozoa	20	18	13	15	22
Cyanobacteria	3	5	3	5	4
Ochrophyta	1	1	1	0	1
Euglenophyta	1	2	1	1	2
Chlorophyta	1	2	0	1	1
Total	82	97	84	91	97

During the period studied, the phytoplankton mean richness fluctuated between 4 in November/17 to 41 in August/18, both registered at bay. In general, the rainy season in all environments (bay, plume, and reef) presented the highest values of richness (Figure 2). The plume showed the highest values of richness, followed by reef and bay.

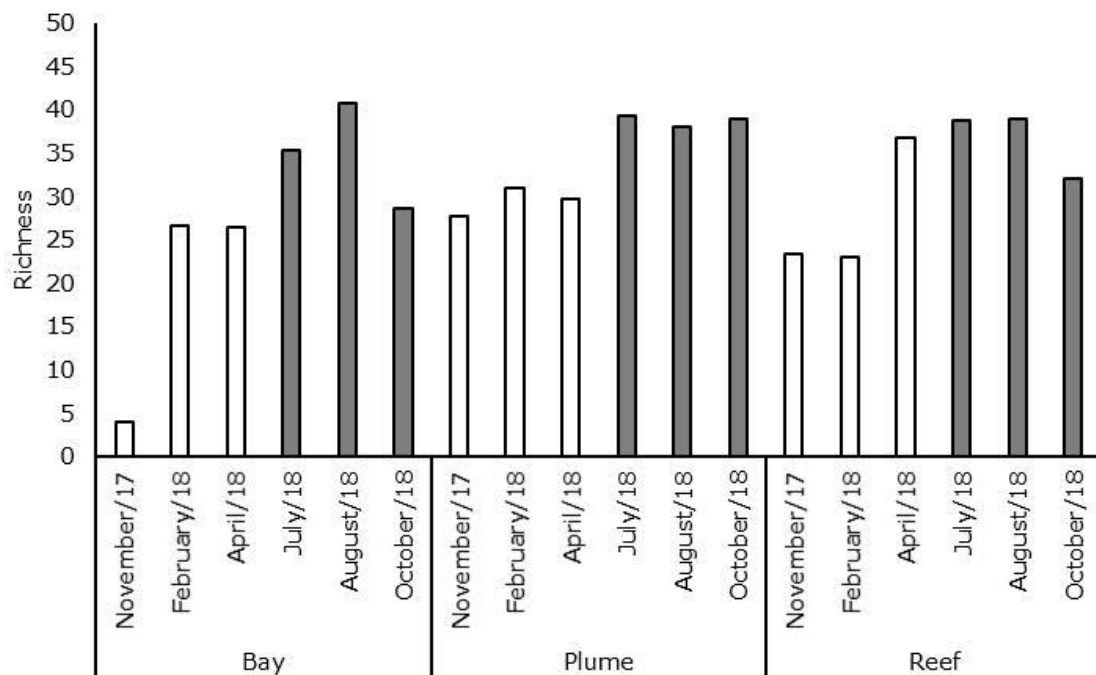


Figure 2 – Phytoplankton richness during the Dry season (White bars) and Rainy season (Gray Bars) in the three different environments (Bay, Plume, and Reef) in Tamandaré bay, Pernambuco, Brazil.

In the studied area was observed 12 taxa classified as Very Frequent, *Trichodesmium* spp. (88.89%), *Protoperidinium* spp. (75.93%), *Campyloneis grevillei* (W.Smith) Grunow &

Eulenstein (81.48%), *Climacosphenia moniligera* Ehrenberg (87.04%), *Coscinodiscus* spp. (94.44%), *Cylindrotheca closterium* (Ehrenberg) Reimann & J.C.Lewin (70.37%), *Licmophora* spp. (85.19%), *Navicula* spp. (94.44%), *Paralia sulcata* (Ehrenberg) Cleve (85.19%), *Pleuro/Gyrosigma* spp. (79.63%), *Rhabdonema adriaticum* Kützing (70.37%), and Euglenophyceae (75.93%). The other taxa were distributed as follows: 30 frequent, 19 infrequent and 52 sporadic.

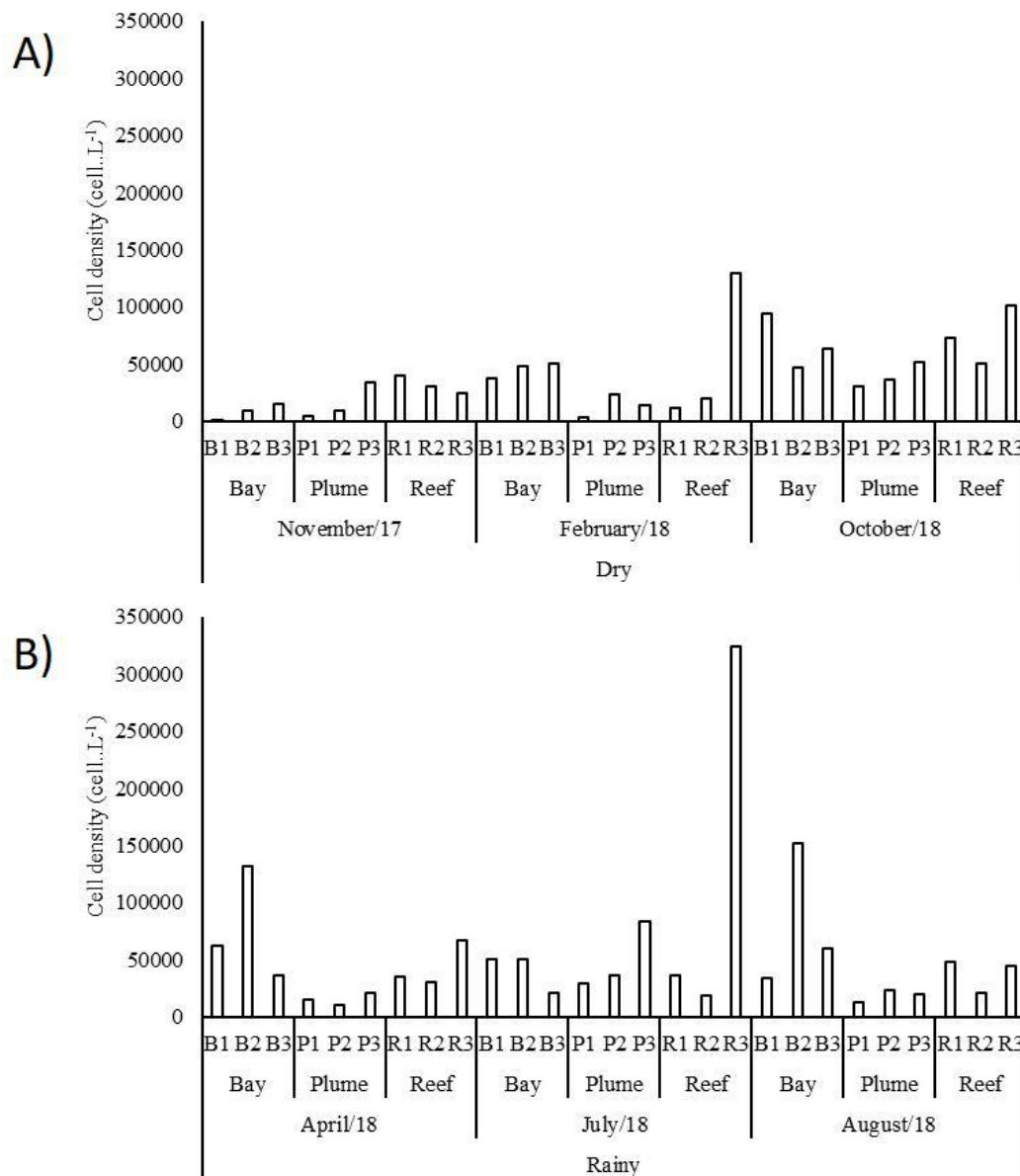


Figure 3 – Phytoplankton cell density (cell.L⁻¹) in each environment (Bay, Plume, and Reef) and seasonal period (Dry (A) and Rainy (B)) in Tamandaré Bay, Pernambuco, Brazil.

Were observed three dominant taxa, *Navicula* spp. at B1 (100%) in November/17, *Trichodesmium* sp. at R3 (64.31%) in July/18 and at B1 in October/18, and *Coscinodiscus* spp. at B1 (54.73%), B2 (80.70%), B3 (51.91%) and R2 (64.29%) all registered in February/18.

In general, the phytoplankton composition was represented by planktonic, marine, and neritic species. Most species identified are of planktonic neritic marine origin (35.00%), followed by tichoplanktonic neritic marine species (28.80%), planktonic oceanic marine species (30.00%), tichoplanktonic neritic estuarine (5.00%) and planktonic oceanic estuarine (1.20%) (Appendices A).

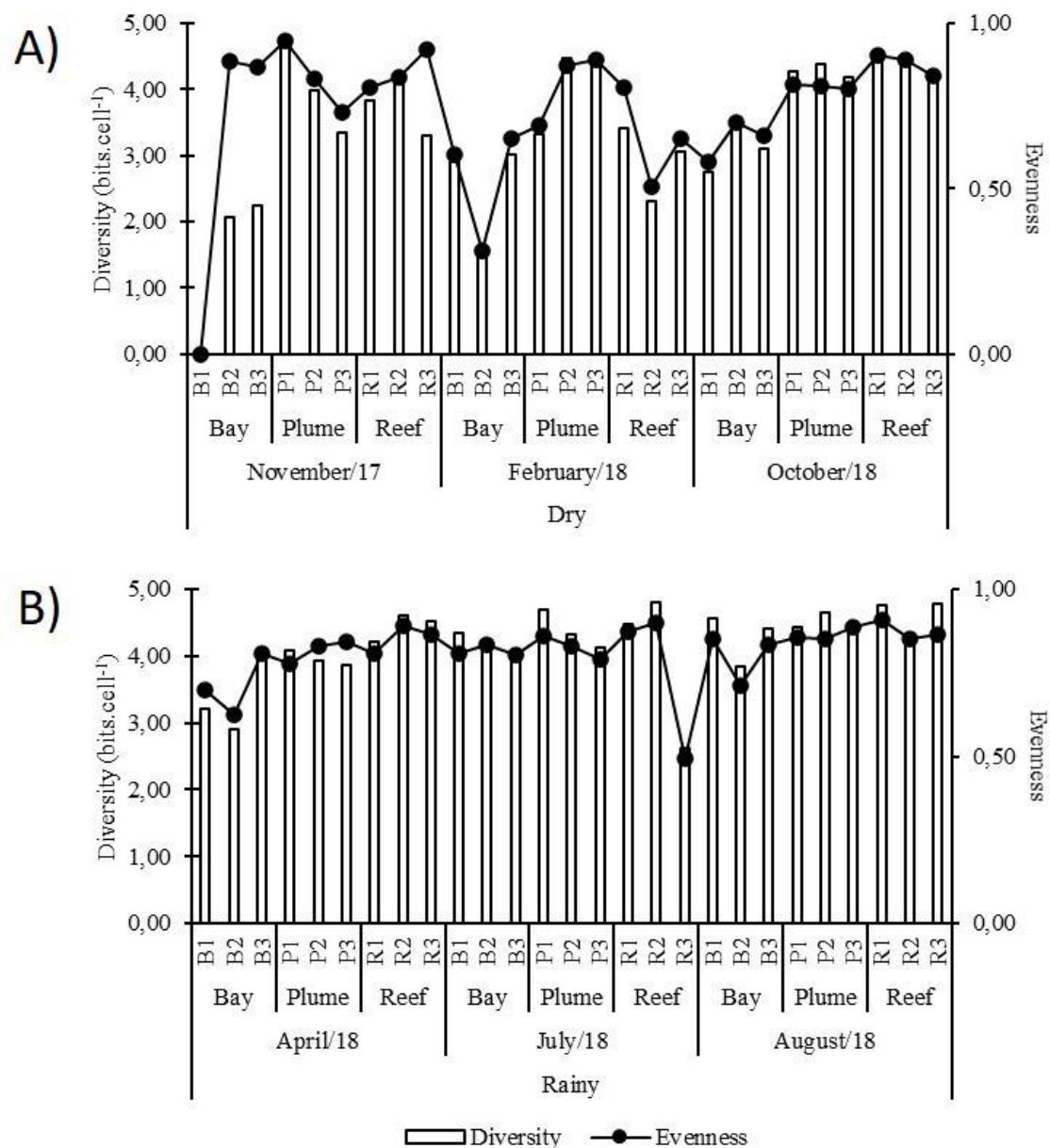


Figure 4 – Phytoplankton species diversity and evenness in each environment (Bay, Plume, and Reef) and seasonal period (Dry (A) and Rainy (B) in Tamandaré Bay, Pernambuco, Brazil.

Phytoplankton density showed a variation between $1,000 \text{ cell.L}^{-1}$ (B1 at November/17) to $324,000 \text{ cell.L}^{-1}$ (R3 at April/18). During the seasonal period, the density fluctuated between $39,037.03 \text{ cell.L}^{-1}$ in the dry season and $54,629.62 \text{ cell.L}^{-1}$ in the rainy (Figure 3). The most

representative area was the reef (61555.55 cell.L⁻¹), followed by bay (53,611.11 cell.L⁻¹) and plume (25,333.33 cell.L⁻¹).

The most representative group in density was Euglenozoa (905,000 cell.L⁻¹), followed by Cyanobacteria (670,000 cell.L⁻¹), Bacillariophyta (595,000 cell.L⁻¹), Miozoa (355,000 cell.L⁻¹) and Chlorophyta (4,000 cell.L⁻¹). Euglenophyceae and *Trichodesmium* sp. contribute to the increase in cell density of phytoplankton in the environments studied.

The species diversity and evenness were high and with well-distributed individuals. The values of species diversity varied from 0.00 bits cell.⁻¹ (due to the presence of only one taxon in the sample) at B1 in November/17 to 4.81 bits cell.⁻¹ at R2 in July/18. Evenness ranged from 0.00 at B1 in November/17 to 0.94 at P1 in November/17 (Figure 4).

The concentration of chlorophyll a varied from 0.13 µg/l in the reef in February/18 to 5.76 µg/l in the plume in April/18. The plume region had the highest mean values of chlorophyll (2.51µg/l), followed by the bay (2.25µg/l) and reef (0.76µg/l). The rainy season was the most productive period (2.13 µg/l) in relation to the dry season (1.41 µg/l) (Figure 5). The chlorophyll a showed significant spatial variation ($p=0.0001$).

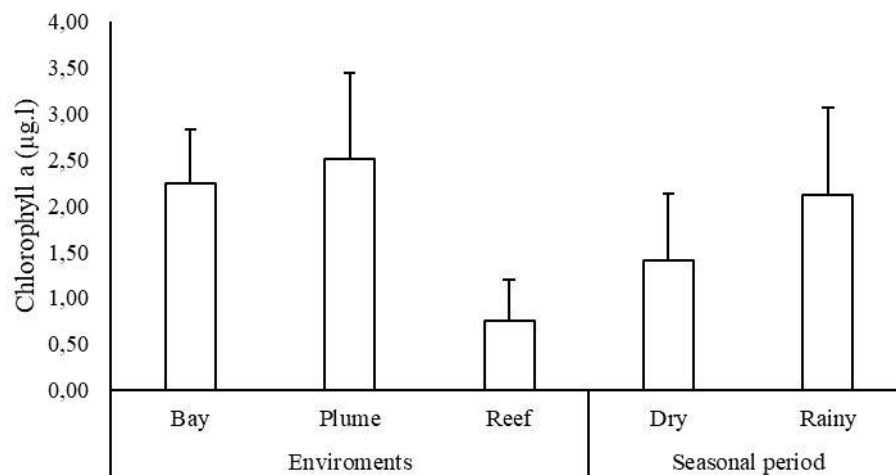


Figure 5 – Chlorophyll *a* average in each environment (Bay, Plume, and Reef) and seasonal period (Dry and Rainy) in Tamandaré Bay, Pernambuco, Brazil.

Microplastics

Qualitative analysis

A total of 845 particles of microplastics presenting filamentous shape were found, divided into four colors: transparent, blue, red, and black. In the bay, plume and reef were found a total of 261, 292 and 292 microplastics, respectively. During the dry season, 325 plastic particles occurred, while in the rainy season 520 particles were observed (Table III).

Table III – Distribution by quantity, color, and average length (μm) of microplastics at the Qualitative analysis recorded in Tamandaré Bay, Pernambuco, Brazil.

Color	Quantity					Length				
	Bay	Plume	Reef	Dry	Rainy	Bay	Plume	Reef	Dry	Rainy
Transparent	228	236	235	258	441	565.68	604.57	613.47	528.78	653.13
Blue	27	43	47	57	60	963.35	912.23	533.23	709.58	882.64
Red	1	9	6	6	10	1,173.40	912.33	1,290.50	1,785.75	670.33
Black	5	4	4	4	9	630.02	1,161.18	1,136.25	1,044.31	928.50
Total	261	292	292	325	520	833.11	897.58	893.36	1,077.10	783.65

The total number of microplastics had an average length of $890.96\mu\text{m}$. In the bay, plume, and reef, they had average lengths of $833.1\mu\text{m}$, $897.57\mu\text{m}$ and $890.86\mu\text{m}$, respectively. Seasonally, the lengths were $1044.31\mu\text{m}$ at dry season and $783.65\mu\text{m}$ at the rainy season. The transparent particles presented shorter length, and the red particles were higher in the entire study area and in the dry season, while the black particles were larger in the reef area and the plume (Table III).

The particles of the microplastics of transparent and blue color were present in all samples during the period studied. Microplastics of other coloration showed a lower frequency of occurrence, except for black particles that were frequent in the plume area.

Quantitative analysis

Were found 902 microplastic filaments in the four colors. In the bay, plume, and reef were found a total of 342, 292 and 268 microplastics, respectively. In the rainy and dry period, there were 338 and 564 particles, respectively. The particles in transparent color were the most representative (Table IV).

The microplastics present throughout the study area showed an average length of $824.76\mu\text{m}$. In the bay, plume, and reef, the lengths were $904.26\mu\text{m}$, $855.4\mu\text{m}$ and $732.18\mu\text{m}$, respectively. In the dry and rainy period, the lengths were $796.56\mu\text{m}$ and $837.04\mu\text{m}$, respectively. The red particles showed longer length, while the transparent particles were smaller, except for the black particles in the reef area and in the rainy season (Table IV).

Table IV – Distribution by quantity, color, and average length (μm) of microplastics at the Quantitative analysis recorded in Tamandaré Bay, Pernambuco, Brazil.

Color	Quantity					Length				
	Bay	Plume	Reef	Dry	Rainy	Bay	Plume	Reef	Dry	Rainy
Transparent	279	225	234	441	297	680.13	603.49	660.05	688.6	607.18
Blue	39	58	19	99	17	740.03	685.30	865.97	883.84	612.34
Red	5	3	3	5	6	1,352.45	1,507.00	1,119.58	1,014.28	1,662.02
Black	19	6	12	19	18	844.44	626.01	283.12	599.53	466.66
Total	342	292	268	564	338	904.26	855.45	732.18	796.56	837.04

Transparent microplastics were present in all samples analyzed, both throughout the study area and in each environment. Microplastics of other color were less frequent, with black microplastics being the least frequent.

Phytoplankton x Microplastic

It was observed that particles of the microplastics was capable to adhere on phytoplankton cells during the actual study (Figure 6 and 7). Damaged cells were present in the samples, but it wasn't possible to identify the cause of the damage.

The ratio found between the number of living organisms and microplastic particles is approximately 6:1. In the bay, plume and reef, the proportions were approximately 7:1, 5:1 and 8:1, respectively.

In the qualitative analysis, in the bay, plume and reef the proportion was approximately 6:1, coinciding with the total proportion. In the quantitative analysis, the bay, plume, and reefs had approximate proportions at 5:1, 2:1 and 7:1, respectively.

The number of phytoplankton cells was higher than that of microplastic in the mean and maximum number, but the microplastic value exceeded that of phytoplankton organisms in the minimum amount. Significant values ($p < 0.05$) were observed seasonally for microplastic and spatially for phytoplankton (Table V).

Table V – Number of microplastics and phytoplankton recorded in Tamandaré Bay, Pernambuco, Brazil. Note: * = $p < 0.05$ (significant variation).

	Mean	Minimum	Maximum	Standard deviation	p-value	
					Seasonal	Spatial
Microplastic	1,690.741	400	5,900	1,100.47	p=0.01	p=0.65
Phytoplankton	4,970.63	100	32,400	5,027.223	p=0.39	p=0.02

Discussion

The studied area presented a high phytoplankton richness, with 113 inventoried taxa, of which 62 taxa were common in the three environments studied (bay, plume, and reef).

The structure of phytoplankton populations is related to the physicochemical characteristics of water and environmental factors, which acting together or separately, establish populations adapted to these variations (Phlips et al. 2002). On the coast of

Pernambuco, changes in phytoplankton distribution and abundance are mainly regulated by eutrophication, river discharge, tidal cycle, and rainfall (Eskinazi-Leça et al. 1997).

The species composition found in the current work is like that found in previous research conducted in the same area. The presence of several groups of microalgae was observed, with diatoms being the most diverse group. Diatoms are considered the most important group of phytoplankton in coastal environments (Rosevel da Silva et al. 2005). The phytoplankton observed in Tamandaré Bay was like registered in other areas of the coastal region of Pernambuco (Aquino et al. 2014; Aquino et al. 2015; Ferreira et al. 2010). This similarity is expected for the coastal region of northeastern Brazil (Santiago 2004).

One of the common taxa found in Tamandaré bay is the genus *Trichosdesmium* (Satô et al. 1963/64; Rosevel da Silva et al. 2005). This taxon contributed more than 90% of the total cyanobacteria identified in the area and was dominant at two different times, one in the rainy season on the reef and another in the dry season in the bay.

These cyanobacteria are adapted to survive in warm ($>20^{\circ}\text{C}$) and nutrient-poor environments, being able to fix atmospheric nitrogen (Marumo and Asaoka 1974). It can form *blooms*, a phenomenon known as "Red Tide", where there is a change in watercolor when there is a high density of these organisms. This *bloom* can be toxic and affect the local population and cause several symptoms, with the most common being fever (Satô et al. 1963/64) and the phenomenon became known as "Tamandaré fever" at the area.

The presence of species belonging to Euglenophyceae may indicate that the environment shows eutrophication (Alves-da-Silva et al. 2013). The highest values of cell density of this class in February/18 at the reefs indicate the human presence because is a vacation season with the increase of tourist at the area.

This phytoplankton diversity can be attributed to the environmental heterogeneity of Tamandaré Bay. The presence of reefs, the mixture of marine flows with the river plume, and the physical and chemical characteristics of the estuaries contributed to this diversity. The dinoflagellates were predominant in oceanic areas, while chlorophytes and euglenophytes were widely distributed in freshwater environments (Boney 1989).

The highest number of cells in the rainy season is related to the entry of nutrients into the environment through leaching. While the highest number in the reef environment may be related to the resuspension of organic matter in the reef sediment. The greatest richness in the plume occurs due to the entry of nutrients that favored the development of phytoplankton from the estuary.

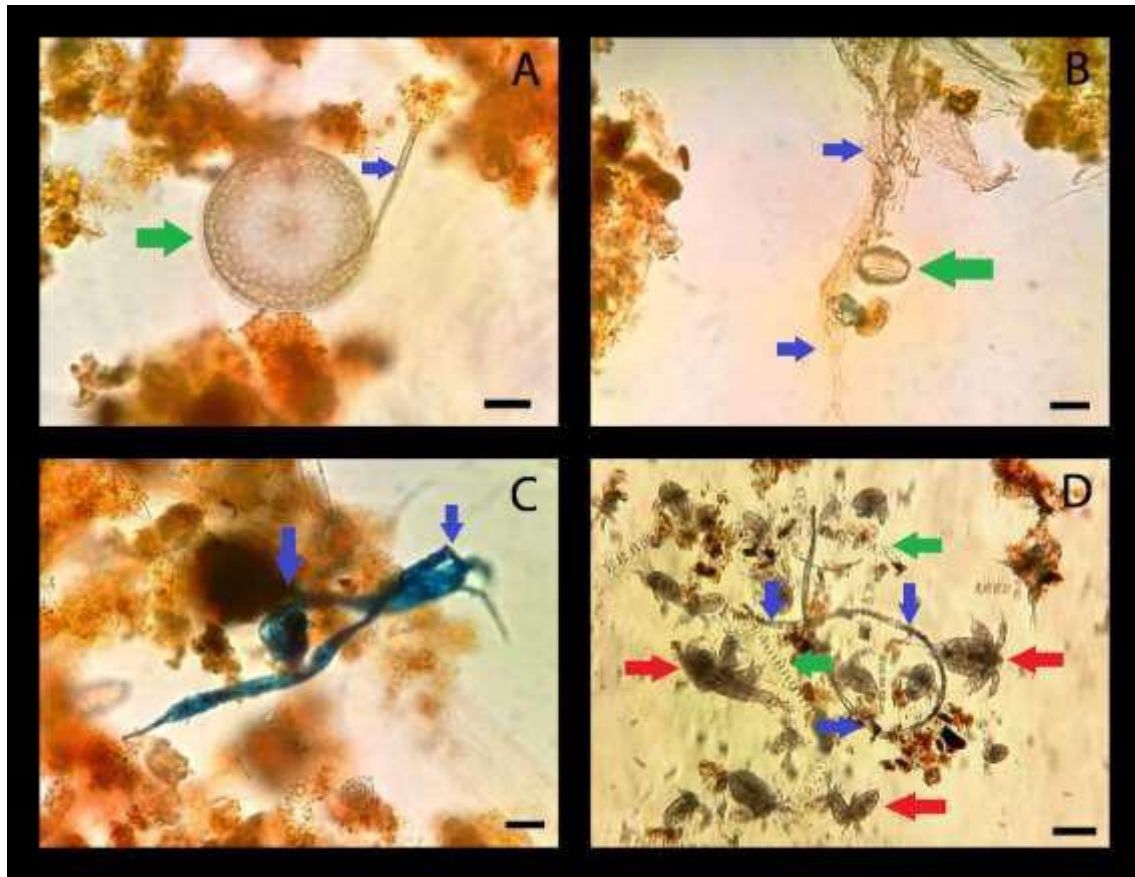


Figure 6 - Particles of the microplastics of transparent (A, B, D) and blue (C) color adhered to phytoplankton cells and in aggregates of organic matter during the period studied in Tamandaré Bay, Pernambuco, Brazil. Blue Arrow = Microplastic; Green Arrow = Phytoplankton; Red Arrow = Zooplankton. Scales are 20 μm (A, B, C), and 100 μm (D).

The number of cells was higher than the number of particles, in almost all samples analyzed, corroborating with one of the hypotheses. The ratio between microplastic and phytoplankton showed that the number of cells was higher than that of pollutants, with approximately six times more units. Even in smaller amounts, the microplastics were present in aggregates and adhered to algae (Figure 6, 7).

The presence of microplastics in plankton samples were observed in other works done in the Northeast of Brazil (Ivar do Sul et al. 2014; Silva-Cunha et al. 2019). The plastic particles found in this study were filamentous and can be classified as fibers (Zhou et al. 2018). They are secondary type microplastics and are considered the most harmful form because they have a large surface area and can interact with a larger number of cells (Wright et al. 2013).

The higher concentration of microplastics in the plume and bay region can occur due to the intake of these pollutants through the river, fishery and tourism activity and the fragmentation of plastic material present at the local.

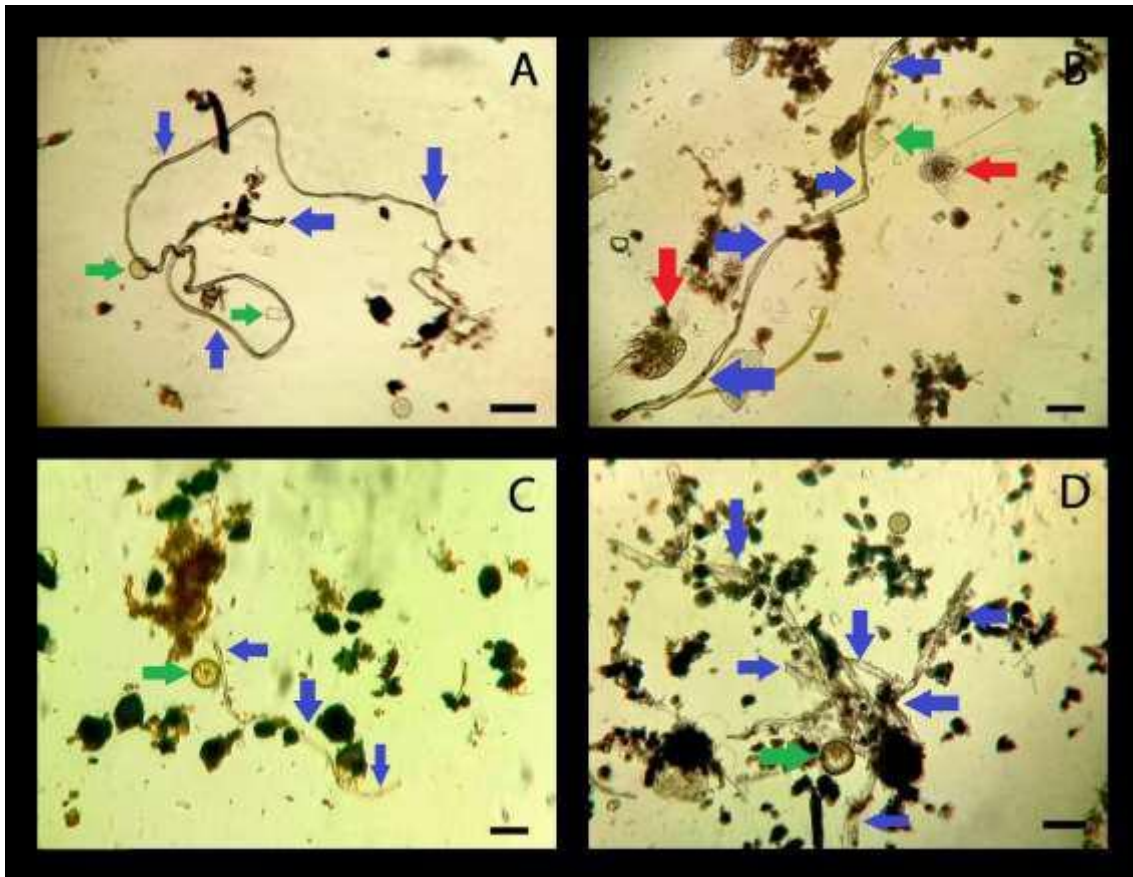


Figure 7 - Particles of the microplastics of transparent color adhered to phytoplankton cells and in aggregates of organic matter during the period studied in Tamandaré Bay, Pernambuco, Brazil. Blue Arrow = Microplastic; Green Arrow = Phytoplankton; Red Arrow = Zooplankton. Scales are 100 μm .

Contamination by microplastics in collecting equipment and in the handling of the material might have occurred, and the number of particles observed in this work can be slightly higher than in the natural habitat.

It is not yet completely understood how the properties of pollutants and the different species of phytoplankton interact with each other, and how the adaptations of organisms make them more resistant or susceptible to the toxicity of pollutants.

It is known that those particles can accumulate on the surface of the microalgae, causing damage to the organisms, and is capable to limit the interaction with light and air, creating oxidative stress in the cell, causing deficiencies in the photosynthesis and reproduction, higher respiration rate and abnormal cell growth (Besseling et al. 2014; Bhattacharya et al. 2010; Sjollema et al. 2016).

Some species of microalgae excrete polysaccharides, when they are under luminous and nutritional stress (Staats et al. 2000; Underwood et al. 2004). Those polysaccharides can coagulate and become adherent and is classified as Transparent exopolymer particles (TEP).

The TEP can form aggregates of cells and organic matter. Microplastic can integrate on those aggregates, and in the TEP produced by microalgae and modify the morphology and the function of the polysaccharides (Chen et al. 2011), changing the rate of precipitation of those aggregates (Long et al. 2015).

The microplastics adhered on the surface of the microalgae or in the aggregates can be transferred to higher rank in the trophic net. The consumers ingest those particles and get contaminated by additives and other toxic substances in the microplastics. Over time, the accumulation of those substances can happen in the organisms (Wright et al. 2013; Setälä et al., 2014).

Many of these studies use a much higher concentration of microplastics than the concentrations found in nature (Prata et al. 2019). It is necessary to understand the mechanism of action and toxic properties of microplastic, and the susceptibilities of phytoplankton, with particle concentrations equal to those found in the natural environment.

The current concentration of these plastic particles in the natural environment may be able to cause changes at the population level, such as the reduction of available nutrients that attach to the surface of the pollutant (Galloway et al. 2017), inhibition of consumer action (Cole et al. 2013) and by serving as substrate and vector of sessile organisms (Oberbeckmann and Labrenz 2020).

All these effects and damage caused by microplastics depend on their specific properties, such as their concentration in the environment, chemical composition, type of polymer, size, presence of additives, and the presence and type of load.

This study confirms a composition and distribution of phytoplankton species is characteristic of tropical regions, and the microplastics observed were fibrous, indicating that they are the result of fragmentation. With the number of plastic particles observed and the ratio between these particles and phytoplankton, it was noticed that the current number of microplastics despite is smaller than that of organisms and they are capable to interact with the microalgae in the formation of aggregates. This interaction should be an alert, as they may become more recurrent if the rate of pollution increases.

More research needs to be carried out on this area of study both in Tamandaré Bay, and elsewhere, to assess whether there is any pattern of distribution of the microplastic, and fill gaps in knowledge due to the lack of information on *in situ* risks that affect the phytoplankton community.

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5 CONSIDERAÇÕES FINAIS

A caracterização do fitoplâncton no presente estudo, indicou a ocorrência de mudança sazonal significativa com a riqueza de espécie mais expressiva no período chuvoso do que no período de estiagem. A riqueza média durante o período chuvoso foi de 18,2 enquanto no período de estiagem foi de 16,7, demonstrando uma homogeneidade no número de espécies ao longo dos períodos sazonais.

Neste estudo, as características hidroclimáticas favoreceram o desenvolvimento de uma comunidade fitoplanctônica diversificada. As diatomáceas apresentaram uma maior riqueza de espécies, destacando-se *Coscinodiscus* spp. que foi mais abundante. A cianobactéria *Trichodesmium* sp. foi a principal espécie que contribuiu para número de células. A presença do filo Euglenophyta no local de estudo pode indicar que ocorre uma leve eutrofização.

Os microplásticos registrados na área são fibrosos, indicando possuem origem secundário e são resultados de fragmentação. A quantidade de microplástico observado no local de estudo, e a razão entre essas partículas e o fitoplâncton, deve ser vista como um alerta, apesar do número menor de partículas, ocorre a apresenta interação com as microalgas na formação de agregados.

O conhecimento acerca dos efeitos dos microplásticos sobre as microalgas é escasso, e existe a necessidade de entender como as propriedades e o mecanismo de ação dos poluentes afetam as diferentes espécies de organismos.

Mais trabalhos precisam ser realizados nessa área de conhecimento tanto no local de estudo atual, a Baía de Tamandaré, como em outros locais, para avaliar se existe algum padrão, além de preencher as lacunas no conhecimento devido à falta de informações dos riscos *in situ* que afetam a comunidade fitoplanctônica.

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**APÊNDICE A – LISTA DE TÁXONS, FREQUÊNCIA DE OCORRÊNCIA E
ECOLOGIA DAS ESPÉCIES DO FITOPLÂNCTON NA BAÍA DE TAMANDARÉ,
PE, BRASIL**

Lista de Táxons, Frequência de ocorrência e Ecologia das espécies do fitoplâncton na Baía de Tamandaré, PE, Brasil. Frequência: E = esporádica, PF = pouco frequente, F = frequente e MF = muito frequente. Ecologia: Plan = planctônica; Tico = ticoplanctônica, Mar = Marinha; Oc = oceânica; Ner = nerítica, Est = estuarina.

Lista de Táxons	Frequência de ocorrência	Ecologia
CYANOBACTERIA		
<i>Lyngbya</i> sp.	PF	-
<i>Merismopedia</i> sp.	E	-
<i>Oscillatoria</i> sp.	E	-
<i>Spirulina</i> sp.	E	-
<i>Trichodesmium</i> sp.	MF	-
MIOZOA		
<i>Ceratium</i> spp.	E	-
<i>Dinophysis caudata</i> W.S.Kent	E	Plan. Mar. Oc.
<i>Gonyaulax fusiformis</i> H.W. Graham	E	Plan. Mar. Oc.
<i>Oxytoxum</i> sp.	E	-
<i>Podolampas</i> sp.	E	-
<i>Prorocentrum micans</i> Ehrenberg	PF	Plan. Mar. Oc.
<i>Prorocentrum gracile</i> F.Schütt	E	Plan. Est. Ner.
<i>Prorocentrum</i> spp.	F	-
<i>Protoperidinium latissimum</i> (Kofoid) Balech	E	Plan. Mar. Oc.
<i>Protoperidinium ovum</i> (J.Schiller) Balech	E	Plan. Mar. Oc.
<i>Protoperidinium</i> spp.	MF	-
<i>Pyrocystis lunula</i> (Schütt)Schütt	E	Plan. Mar. Ner.
<i>Tripes arietinus</i> (Cleve) F.Gómez	PF	Plan. Mar. Oc.
<i>Tripes belone</i> (Cleve) F.Gómez	E	Plan. Mar. Ner.
<i>Tripes brevis</i> (Ostenfeld & Johannes Schmidt) F.Gómez	E	Plan. Mar. Oc.
<i>Tripes declinatus</i> (G.Karsten) F.Gómez	E	Plan. Mar. Oc.
<i>Tripes falcatus</i> (Kofoid) F.Gómez	E	Plan. Mar. Oc.
<i>Tripes furca</i> (Ehrenberg) F.Gómez	PF	Plan. Mar. Ner.
<i>Tripes fusus</i> (Ehrenberg) F.Gómez	PF	Plan. Mar. Ner.
<i>Tripes horridus</i> (Cleve) F.Gómez	PF	Plan. Mar. Ner.
<i>Tripes longirostris</i> (Gourret) F.Gómez	E	Plan. Mar. Oc.
<i>Tripes muelleri</i> Bory	E	Plan. Mar. Oc.
<i>Tripes pentagonus</i> (Gourret) F.Gómez	PF	Plan. Mar. Oc.
<i>Tripes teres</i> (Kofoid) F.Gómez	PF	Plan. Mar. Ner.
BACILLARIOPHYTA		
<i>Achnanthes</i> sp.	E	-
<i>Amphipentacrinus</i> Ehrenberg	PF	Tico. Mar. Ner.
<i>Amphiprora</i> sp.	F	-
<i>Amphitetras antediluviana</i> Ehrenberg	PF	Tico. Mar. Ner.
<i>Amphora</i> sp.	E	-

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Lista de Táxons	Frequência de ocorrência	Ecologia
<i>Anaulus mediterraneus</i> Grunow	E	Plan. Mar. Ner
<i>Asterionellopsis glacialis</i> (Castracane) Round	F	Plan. Mar. Ner
<i>Aulacodiscus</i> sp.	E	-
<i>Auricula</i> sp.	F	-
<i>Bacillaria paxillifera</i> (O.F.Müller)	F	Tico. Mar. Ner
<i>Bacteriastrum delicatulum</i> Cleve	E	Plan. Mar. Oc
<i>Bacteriastrum hyalinum</i> Lauder	E	Plan. Mar. Oc
<i>Bellerochea malleus</i> (Brightwell) Van Heurck	PF	Plan. Mar. Ner
<i>Biddulphia biddulphiana</i> (J.E.Smith) Boyer	F	Plan. Mar. Ner
<i>Biddulphia</i> sp.	E	-
<i>Biddulphia tridens</i> (Ehrenberg) Ehrenberg	PF	Plan. Mar. Ner
<i>Bleakeleya notata</i> (Grunow) Round	F	Plan. Mar. Ner
<i>Campylodiscus</i> sp.	F	-
<i>Campyloneis grevillei</i> (W.Smith) Grunow & Eulenstein	MF	Tico. Mar. Ner
<i>Cerataulina pelagica</i> (Cleve) Hendey	E	Plan. Mar. Oc
<i>Chaetoceros affinis</i> Lauder	E	Plan. Mar. Ner
<i>Chaetoceros brevis</i> F.Schütt	E	Plan. Mar. Ner
<i>Chaetoceros coarctatus</i> Lauder	E	Plan. Mar. Oc
<i>Chaetoceros curvisetus</i> Cleve	PF	Plan. Mar. Ner
<i>Chaetoceros lorenzianus</i> Grunow	F	Plan. Mar. Ner
<i>Chaetoceros</i> spp.	F	-
<i>Chaetoceros teres</i> Cleve	E	Plan. Mar. Ner
<i>Chrysanthemodiscus floriatus</i> A.Mann	F	Plan. Mar. Ner
<i>Climacosphenia moniligera</i> Ehrenberg	MF	Tico. Mar. Ner
<i>Corethron hystrix</i> Hensen	E	Plan. Mar. Oc.
<i>Coscinodiscus centralis</i> Ehrenberg.	F	Plan. Mar. Oc.
<i>Coscinodiscus oculus-iridis</i> (Ehrenberg) Ehrenberg	E	Plan. Mar. Ner.
<i>Coscinodiscus</i> spp.	MF	-
<i>Cylindrotheca closterium</i> (Ehrenberg) Reimann & J.C.Lewin	MF	Plan. Mar. Ner
<i>Entomoneis alata</i> (Ehrenberg) Ehrenberg	F	Plan. Mar. Ner
<i>Fragillaria</i> sp.	F	-
<i>Grammatophora marina</i> (Lyngbye) Kützing	F	Tico. Mar. Ner
<i>Grammatophora oceanica</i> Ehrenberg	F	Tico. Mar. Ner
<i>Guinardia striata</i> (Stolterfoth) Hasle	PF	Plan. Mar. Oc.
<i>Halamphora turgida</i> (Gregory) Levkov	E	Tico. Est. Ner
<i>Helicotheca thamensis</i> (Shrubsole) M.Ricard	F	Plan. Mar. Ner
<i>Hemiaulus</i> sp.	E	-
<i>Isthmia enervis</i> Ehrenberg	F	Plan. Mar. Ner
<i>Lampriscus orbiculatus</i> (Shadbolt) Peragallo & Peragallo	F	Plan. Mar. Ner
<i>Licmophora flabellata</i> (Greville) C.Agardh <i>Licmophora</i> spp.	PF	Tico. Mar. Ner
	MF	-

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Lista de Táxons	Frequência de ocorrência	Ecologia*
<i>Lithodesmium undulatum</i> Ehrenberg	F	Plan. Mar. Ner.
<i>Mastogloia splendida</i> (Gregory) H.Pergallo	F	Tico. Mar. Ner.
<i>Navicula</i> spp.	MF	-
<i>Neocalyptrella robusta</i> (G.Norman ex Ralfs) Hernández-Becerril & Meave del Castillo	PF	Plan. Mar. Oc.
<i>Nitzschia commutata</i> Grunow	E	Tico. Est. Ner.
<i>Nitzschia longissima</i> (Brébisson) Ralfs	E	Tico. Mar. Ner.
<i>Nitzschia</i> spp.	E	-
<i>Odontella aurita</i> (Lyngbye) C.Agardh	F	Tico. Mar. Ner.
<i>Odontella longicruris</i> (Greville) M.A.Hoban	F	Plan. Mar. Ner.
<i>Odontella turgida</i> (Ehrenberg) Kützing	F	Tico. Mar. Ner.
<i>Palmerina hardmaniana</i> G.R.Hasle	E	Plan. Mar. Ner.
<i>Paralia sulcata</i> (Ehrenberg) Cleve	MF	Tico. Mar. Ner.
<i>Petroneis humerosa</i> (Brébisson ex W.Smith) Stickle & D.G.Mann <i>Pleuro/Gyrosigma</i> spp.	PF MF	Tico. Mar. Ner. -
<i>Podocystis adriatica</i> (Kützing) Ralfs	F	Tico. Mar. Ner.
<i>Rhabdonema adriaticum</i> Kützing	MF	Tico. Mar. Ner.
<i>Rhabdonema punctatum</i> (Harvey & Bailey) Stodder	F	Tico. Mar. Ner.
<i>Rhizosolenia</i> sp.	E	-
<i>Rhizosolenia styliformis</i> T.Brightwell	E	Plan. Mar. Oc.
<i>Stephanopyxis</i> sp.	E	-
<i>Surirella</i> sp.	E	-
<i>Terpsinoë musica</i> Ehrenberg	E	Tico. Est. Ner
<i>Thalassionema nitzschioides</i> (Grunow) Mereschkowsky	F	Plan. Mar. Oc
<i>Thalassiosira leptopus</i> (Grunow) Hasle & G.Fryxell	E	Plan. Mar. Oc
<i>Toxarium undulatum</i> Bailey	E	Tico. Est. Ner
<i>Triceratium balearicum</i> Cleve & Grunow	E	Tico. Mar. Ner
<i>Triceratium biquadratum</i> Janisch	F	Tico. Mar. Ner
<i>Triceratium dubium</i> Brightwell	E	Tico. Mar. Ner
<i>Triceratium favus</i> Ehrenberg	PF	Tico. Mar. Ner
<i>Triceratium</i> spp.	PF	-
<i>Trieres mobiliensis</i> (Bailey) Ashworth & Theriot	F	Plan. Mar. Oc..
<i>Trieres regia</i> (M.Schultze) M.P.Ashworth & E.C.Theriot	F	Plan. Mar. Ner.
<i>Tryblionella compressa</i> (Bailey) Poulin	E	Plan. Mar. Ner.
OCHROPHYTA		
<i>Dictyocha fibula</i> Ehrenberg	E	Plan. Mar. Oc
CHLOROPHYTA		
Chlorophyceae	E	-
<i>Micrasteria</i> sp.	E	-
EUGLENOZOA		
Euglenophyceae	MF	-
<i>Trachelomonas</i> sp.	E	-

ANEXO A - NORMAS DA REVISTA REFERENTE AO ARTIGO

Normas da Revista referente ao Capítulo 1

Marine Biology Research (ISSN: 1745-1000)

Marine Biology Research welcomes the submission of research reports on all aspects of marine biology (ecology, biodiversity, evolution, physiology, and behavior). The Journal will consider applied aspects (i.e. environmental or fisheries management) insofar as they contribute to more general biological knowledge. See also the Marine Biology Research homepage (<http://www.tandfonline.com/smar>).

The following categories of reports will be considered:

1. *Original articles*
2. *Invited reviews*
3. *Short reports*
4. *Book reviews*

Invitations for reviews (category 2) may come directly from the editors. Authors who wish to publish an Invited Review should contact the editors to reach agreement on the topic and maximum length. Short reports should be no longer than six printed pages and may either represent short research articles or, published under separate subcategories, Mini-reviews, or Comments on recent articles in Marine Biology Research. Online samples of the latter are available for free download. Please visit the online author instructions at the journal homepage (www.tandfonline.com/smar) for further information.

Findings on the range extension of marine organisms must be accompanied by:

1. detailed biogeographic information,
2. systematically oriented comparative data, or
3. ecological data revealing impacts or interactions.

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