

Temporal and spatial variation of phytoplankton in a tropical reef area of Brazil

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Abstract: In the reef ecosystem of São José da Coroa Grande (State of Pernambuco, Brazil), located in the Coral Coast Area of Environmental Protection, an analysis of environmental data (rainfall, water temperature, salinity, depth, water transparency, nitrite, nitrate, phosphate, silicate, ammonia, suspended solids, dissolved oxygen and pH), phytoplankton biomass and density was performed, on surface water samples during the rainy (May, June and July 2010) and dry season (November, December 2010 and January 2011). The results showed no significant seasonal and spatial variation in the analyzed parameters. The only exceptions were water transparency, nitrite, phosphate and silicate that were significantly influenced by the typical pattern of rainfall of the coast of Pernambuco. The phytoplankton exhibited low density and ranged from 7.0×10^3 to 9.5×10^4 cells L^{-1} , with diatoms and dinoflagellates as prominent groups. Low levels of nutrients and chlorophyll *a* were observed with the size fraction smaller than 20 μm (nano and picoplankton) being the main contributor to the overall phytoplankton biomass. The area was determined to be oligotrophic as would be expected for coastal ecosystems free of environmental impacts.

Resumen: En el ecosistema arrecifal de São José da Coroa Grande (estado de Pernambuco, Brasil), situado en la Zona de Protección Ambiental Costa de Coral, se llevó a cabo un análisis de datos ambientales (precipitación, temperatura del agua, salinidad, profundidad, transparencia del agua, nitritos, nitratos, fosfatos, silicatos, amoníaco, sólidos suspendidos, oxígeno disuelto y pH), y de la biomasa y la densidad del fitoplancton, en muestras de agua superficial durante la temporada de lluvias (mayo, junio y julio de 2010) y la temporada seca (noviembre, diciembre de 2010 y enero de 2011). Los resultados no mostraron variación estacional y espacial significativa en los parámetros analizados. Las únicas excepciones fueron la transparencia del agua, nitritos, fosfatos y silicatos, los cuales estuvieron influenciados significativamente por el típico patrón de lluvias de la costa de Pernambuco. El fitoplancton mostró una densidad baja que varió de 7.0×10^3 a 9.5×10^4 células L^{-1} , siendo las diatomeas y los dinoflagelados grupos prominentes. Se observaron niveles bajos de nutrientes y de clorofila *a*, siendo la fracción de tamaño inferior a 20 μm (nano y picoplancton) la que más contribuyó a la biomasa total del fitoplancton. Se determinó que el área es oligotrófica, como se espera para los ecosistemas costeros que no han sufrido impactos ambientales.

Resumo: No ecossistema recifal de São José da Coroa Grande (Pernambuco, Brasil),

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inserido na Área de Proteção Ambiental da Costa dos Corais, efetuou-se uma análise de dados ambientais (precipitação pluviométrica, temperatura da água, salinidade, profundidade, transparência da água, nitrito, nitrato, fosfato, silicato, amônia, material em suspensão, oxigênio dissolvido e pH), biomassa e densidade fitoplanctônica, realizada mediante coletas superficiais com garrafa durante o período chuvoso (maio, junho e julho / 2010) e de estiagem (novembro, dezembro / 2010 e janeiro / 2011). Os resultados obtidos indicaram ausência de variação espacial e sazonal significativa nos parâmetros analisados exceto transparência da água, nitrito, fosfato e silicato, influenciados significativamente pelo padrão típico de chuvas para o litoral pernambucano. O fitoplâncton apresentou baixos valores de densidade, variando de $7,0 \times 10^3$ a $9,5 \times 10^4$ cel L⁻¹, com diatomáceas e os dinoflagelados como grupos de destaque. Foram encontrados baixos teores de sais nutrientes e de clorofila a, no qual frações menores que 20 µm (nano e picoplâncton) foram os principais contribuintes para biomassa fitoplanctônica total. A área foi classificada como oligotrófica, dentro do esperado para ecossistemas costeiros livres da ação de fatores impactantes.

Key words: Fractionated chlorophyll *a*, phytoplankton density, reef ecosystem.

Introduction

Reefs stand out among the coastal ecosystems due to the species richness and the productivity they support (Barker & Roberts 2004), their role in protecting the coastline against erosion and favoring numerous human activities (Manso *et al.* 2003). Such activities result in changes in ecological conditions, such as modification of habitats, biodiversity loss and the appearance of toxic species. These changes in habitats, or even in the abundance of marine species, are observed mainly in coastal areas subject to multiple indiscriminate uses (Costa Jr. 2007).

Among the main human activities that generate changes in reef environments stand out fishing, tourism, aquariophily and housing occupation. However, the consequences of environmental damage on this ecosystem are still poorly studied (Viles & Spencer 1995). Among the studied areas, stands out the Great Barrier Reef of Australia (Ayukai 1995; Furnas *et al.* 1990), as well as other reefs located at French Polynesia (Charpy & Charpy-Roubaud 1990; Ferrier-Pagés & Furla 2001), Fiji Islands (Charpy & Blanchot 1999), Red Sea (Genin *et al.* 2009) and the coast of India (Banse *et al.* 1996; Sridhar *et al.* 2006; Sridhar *et al.* 2011).

Along the coast of northeastern Brazil, extensive reef formations are found, also known as beachrocks, consisting of sandstone and ferruginous sandstone basis, being covered by algae and corals. It constitutes, therefore, one of the main

areas of species diversity along the coast of Brazil (Pereira *et al.* 2002).

Despite being a very peculiar feature of the region, the studies performed on the phytoplankton community are still limited and mostly use primary productivity, biomass (chlorophyll *a*) and taxonomic groups, including: Sassi & Moura (1989) at Ponta do Seixas, State of Paraíba; Feitosa & Passavante (2004) at Rocas Atoll; Feitosa & Bastos (2007) and Mayal *et al.* (2009) at Parrachos de Maracajá, State of Rio Grande do Norte; Fonseca *et al.* (2002) and Machado *et al.* (2007) at Porto de Galinhas, Jales *et al.* (2009) at Serrambi and Bastos *et al.* (2011) at Maracáipe, State of Pernambuco.

The study of phytoplankton on reef ecosystems is of great ecological importance, since they constitute the beginning of the trophic web, as well as they respond quickly to environmental impacts, being considered excellent indicators of water quality (Eskinazi-Leça *et al.* 2011). Although most of the primary production being attributed to benthic algae, to dinoflagellate symbionts (zooxanthellae) and seagrass meadows (Costa *et al.* 2007), an expressive development of phytoplankton has been found in these environments, though its waters have low concentrations of nutrient salts (Sassi *et al.* 1990). This phenomenon, attributed to the mechanisms of enrichment due mainly to local climatological conditions, demonstrates that these areas of oligotrophic characteristics may present specific phases of mesotrophy (Bastos *et al.* 2011; Sassi *et al.* 1990).

Some human activities are observed in the reefs of São José da Coroa Grande, on the southern coast of the state of Pernambuco, in which the tourism and fishing activities are mainly related to seasonal and tidal periods. Thus, they harm the reef area, once they result in trampling the reef, use of vessels nearby, deposition of solid waste and pollutants, and the use of fishing nets.

In view of what was said above, the main objective of the present study was to evaluate the quantitative changes in cell density and biomass of phytoplankton in the reef system of São José da Coroa Grande (Pernambuco, Brazil), resulting from the dynamic action of the environmental parameters and the human actions in the area. With this, we intend to answer the following question: "How phytoplankton and environmental parameters are related, due to seasonality, spatiality and tidal action in the reef system of São José da Coroa Grande-PE (Northeastern Brazil)?".

Materials and methods

Study area

The Municipality of São José da Coroa Grande is located at the southern end of the coast of Pernambuco, distant 123 km of the capital, Recife. It is included in the geographical microregion of the Pernambuco Meridional Forest, limited to the north and west by the Municipality of Barreiros; to the south by the Municipality of Maragogi, State of Alagoas, and to the east by the Atlantic Ocean (CPRH 1999). The name of the beach, São José da Coroa Grande, has as origin the sandbanks called crowns ('coroa' in portuguese) that emerge during low tide, between the sea and coral reefs, forming natural pools (Bernardino 2004).

The dominant climate is tropical humid with rainy autumn, i.e., the climate As' in Köppen classification (pseudo tropical). The total rainfall is above 2400 mm per year, caused mainly by the cyclones of the Atlantic Polar Front that reach the northeast coast of Brazil with greater intensity in autumn and winter (Aragão 2004). The highest rainfall occurs in the months of May, June and July, while the months of November, December and January are the driest ones. The average annual temperature in the area is of 24 °C, ranging between 18 and 32 °C, under strong influence of the Southeast and Northeast trade winds, prevailing winds in Pernambuco coast throughout the year (CPRH 1999).

The coastal region of the municipality is rich in

mangroves and estuaries, related to the watersheds of the Una and Persinunga Rivers and also the Meireles Creek. At the North of the studied area, it is located the Una river estuary, with a reasonable state of conservation regarding the flora and fauna, but its geomorphology has suffered great alterations due to changes in its mouth. The estuary of the Meireles creek is the less impacted by anthropic actions, being threatened by crop pesticides and housing occupation (in the areas of mangrove and riparian forest), being under investigation of the environmental organizations. At the South, it is located the Persinunga river, with serious pollution problems, arising from poor housing occupation on its margins (CPRH 2003).

The inner continental shelf (the area between the shoreline and the isobath of 20 m) presents a gentle slope, interrupted by irregularities related to the presence of channels and sandstone reefs (beachrocks) arranged parallel to the coast. This platform is narrow, since the break occurs around 32 km from the coast, between depths of 50 - 60 m (Camargo *et al.* 2007).

Reefs of the São José da Coroa Grande beach are inserted in the Environmental Protection Area (APA) of the Coral Coast, regulated by Federal Decree of October 23, 1997 and it is considered the first national conservation unit for coastal reefs of northeastern Brazil and the largest federal unit of marine conservation. Despite of its good condition, the lack of awareness about the importance of this ecosystem, agricultural and domestic pollution, degradation due to fishing (subsistence fishing, deep sea fishing and amateur commercial catch of shrimps, lobsters, reef and pelagic fishes), disorganized traffic of vessels, uncontrolled tourism and urban development are potential risks (Maida & Ferreira 1997; Wilkinson 2002).

Experimental strategy

Samples were collected in May, June and July 2010 (rainy season) and in November, December 2010 and January 2011 (dry season), during high and low tides of spring tide, on the same day, with the aid of boat (small motorboat). Three sampling points were established on the reef region (Fig. 1): Point 1 (P1) - 8° 53' 143" S, 35° 8' 065" W - situated near the Meireles creek, often visited by vessels (tourism) due to its shallow depth during low tide (natural pool formation), Point 2 (P2) - 8° 53' 981" S, 35° 8' 501" W - place widely used in subsistence fishing; Point 3 (P3) - 8° 54' 609" S, 35° 8' 917" W -

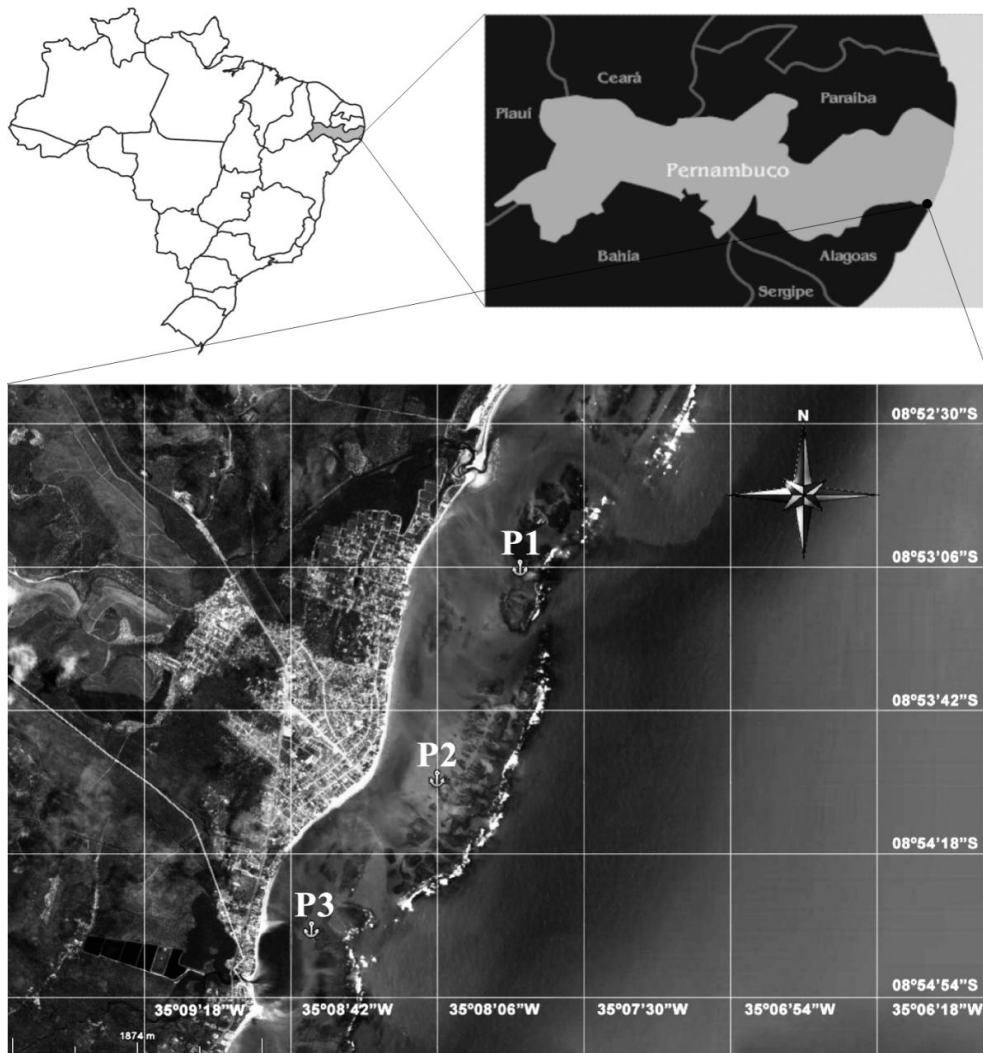


Fig. 1. Geographic location of São José da Coroa Grande, in the State of Pernambuco, Brazil. In the insert, the location of the sampling points in the reef ecosystem.

situated near the mouth of the Persinunga River, where a large sandbar connects the beach to the reefs and natural pools during low tide, facilitating access to touristic and recreational activity without the need of boats and yachts.

Environmental parameters

The height of the tides was obtained from the tide chart of the National Bank of Oceanographic Data of the Directorate of Hydrography and Navigation (DHN) of the Brazilian Navy, for the Port of Suape, Pernambuco, during the study period.

The rainfall data were obtained from the National Institute of Meteorology (INMET), corresponding to the rainfall during the months of study and the climatological normal for the last 30 years.

The local depth was verified with the aid of a manual probe, attached to a cable graduated in centimeters. From the depth of disappearance of the Secchi disk, the water transparency was determined, while using the Kitahara bottle, samples were collected for determination of physical, chemical and biological characteristics of the water.

The water temperature was measured *in situ* with the aid of a graduated thermometer. In the laboratory, the salinity was analyzed by the Mohr-Knudsen method and the dissolved oxygen by the Winkler method, both described by Strickland & Parsons (1972). The analysis of suspended particulate matter followed as recommended by Melo *et al.* (1975). The dissolved nutrient salts (nitrate, nitrite and phosphate) were determined

according to Strickland & Parsons (1972), while the concentrations of ammonia and silicate were obtained according to Grasshoff *et al.* (1983).

*Concentration of chlorophyll *a**

In laboratory, the total and fractioned phytoplankton biomass was quantified by measuring the concentration of chlorophyll *a*, with filtering, pigment extraction and spectrophotometric analysis, according to the method of UNESCO (1966). The fractioned phytoplankton biomass was obtained through the separation of fractions smaller than 20 μm (pico and nano-phytoplankton) and greater than 20 μm (micro-phytoplankton), according to Platt *et al.* (1983). For that, the sample was filtered with a PVC glass with nylon mesh of 20 μm . The system of trophic classification of the environment used was based on Håkanson (1994), in which the environments with average levels of chlorophyll < 1 are considered oligotrophic, between 1 - 3 mesotrophic, between 3 - 5 eutrophic, and > 5 hypereutrophic.

Phytoplankton density

The samples for the study of the phytoplankton cell density were placed in one litre polyethylene frosted bottles, fixed with 2 % acetic lugol, for later analysis under the Axiovert 40 C - Carl Zeiss inverted microscope at 450 x, according to the method of Utermöhl (1958). For that, 10 ml subsamples were used for the identification and counting of the organisms, with the results expressed as cells per liter (cells L^{-1}). When necessary, the 100 x objective lens was used, with immersion oil, in the optical microscope Axioskop 50 - Carl Zeiss, for accurate species identification. Such individuals were measured with the aid of ocular micrometer. For a better view of ornamentation of some species of diatoms and dinoflagellates in optical microscope, the method proposed by Carr *et al.* (1986) was used, as well as the use of phase contrast to show the number of chloroplasts. Whenever possible, the organisms were photographed by TCA - 1.31C digital camera and TSView software for image capture.

The identification and classification of taxa were determined through specialized literature: Hustedt (1930, 1959, 1961 - 66), Cupp (1943), Balech (1988), Silva-Cunha & Eskinazi-Leça (1990), Licea *et al.* (1995), Tomas (1997) and Gómez (2005). The criteria for refinement of identification and the scientific names of species were confirmed through the online database Algaebase (Guiry & Guiry

2014) and the List of species of Brazilian Flora (Eskinazi-Leça *et al.* 2014).

Statistical analysis

To check for significant differences ($P < 0.05$) among the sampling points (P1, P2 and P3) the nonparametric Kruskal-Wallis test was used, while to check for seasonal differences (dry and rainy period) and between tides (low tide and high tide), the nonparametric Mann-Whitney test was applied. All tests were performed using the software BioEstat 5.3.

The principal components analysis (PCA) was performed using the Primer® 6.1, correlating a data matrix in order to assess the relationship among environmental parameters, chlorophyll *a* and number of cells per liter (total density) through the Pearson coefficient.

Results

Environmental parameters

The monthly average rainfall obtained during the study period confirms the occurrence of a well-defined seasonal cycle and oscillated from 23 to 513 mm (Fig. 2). Comparing these values with the equivalent of the climatological normal (last 30 years), it was observed that there was no significant difference, which characterizes the studied months as normal, from the point of view of rainfall, in relation to the climatological normal for the coast of Pernambuco State.

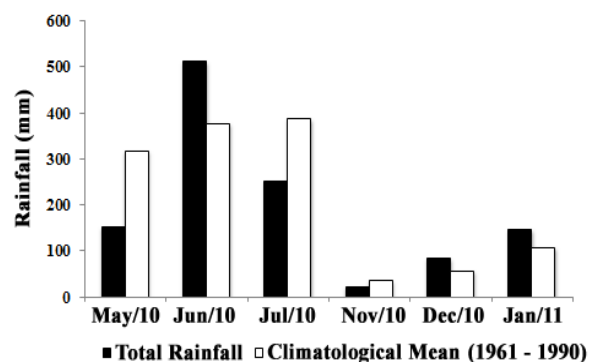


Fig. 2. Data on rainfall, with the total monthly rainfall and the climatological mean from 1961 to 1990.

The salinity and water temperature variables showed no significant differences (Table 1) for the three studied treatments (spatial, seasonal and tidal). The salinity was characteristic of the coastal

Table 1. Minimum, mean and maximum values, standard deviation and statistical significance of abiotic and biotic variables in the reef ecosystem of São José da Coroa Grande, Pernambuco, Northeastern Brazil, in the rainy and dry season - (SPM) suspended particulate matter, (Chlor-a) chlorophyll a, (Min) minimum, (Max) maximum, (SD) standard deviation, (KW) Kruskal-Wallis test, (MW) Mann Whitney test.

Parameters	Rainy season			Dry season			KW (p)		MW (p)
	Min	Max	Mean \pm SD	Min	Max	Mean \pm SD	Spatial	Seasonal	Tidal
Temperature ($^{\circ}$ C)	26.00	29.00	27.67 \pm 1.19	27.50	29.00	28.47 \pm 0.54	0.57	0.07	0.24
Salinity	30.33	37.16	34.87 \pm 1.75	33.42	37.43	35.56 \pm 1.07	0.60	0.30	0.08
Depth (m)	0.90	3.70	2.08 \pm 0.87	0.90	3.85	2.18 \pm 0.99	0.40	0.81	*0.00
Transparency (m)	0.70	2.35	1.35 \pm 0.42	0.90	3.40	2.04 \pm 0.82	0.16	*0.02	*0.00
Oxygen (mL L ⁻¹)	3.40	6.29	5.36 \pm 0.68	4.44	6.39	5.36 \pm 0.51	0.42	0.76	0.08
Saturation (%)	76.45	145.20	120.90 \pm 15.36	100.00	145.20	120.70 \pm 12.24	0.27	0.57	0.16
SPM (mg L ⁻¹)	2.00	45.78	15.37 \pm 15.20	1.78	46.43	15.79 \pm 12.24	0.73	0.59	0.17
pH	8.10	8.46	8.25 \pm 0.09	7.91	8.42	8.18 \pm 0.14	0.54	0.09	0.30
Ammonia (μ M)	0.00	0.06	0.01 \pm 0.02	0.00	0.00	0.00 \pm 0.00	0.14	0.27	0.13
Nitrite (μ M)	0.00	0.23	0.11 \pm 0.07	0.00	0.00	0.00 \pm 0.00	0.91	*0.00	0.92
Nitrate (μ M)	0.40	3.10	1.4 \pm 0.79	0.20	18.20	3.61 \pm 5.08	0.72	0.21	0.27
Phosphate (μ M)	0.16	0.29	0.23 \pm 0.04	0.04	0.27	0.13 \pm 0.07	0.78	*0.00	0.99
Silicate (μ M)	3.60	42.50	12.41 \pm 10.00	4.09	40.50	20.24 \pm 11.40	0.68	*0.02	0.96
Chlor-a Total (mg m ⁻³)	0.14	8.79	3.32 \pm 2.29	0.00	6.24	2.71 \pm 2.09	0.98	0.67	0.90
Chlor-a < 20 μ m (mg m ⁻³)	0.00	5.50	2.36 \pm 1.71	0.00	5.70	2.43 \pm 1.92	0.87	0.63	0.91
Chlor-a > 20 μ m (mg m ⁻³)	0.00	3.42	0.87 \pm 1.19	0.00	2.06	0.28 \pm 0.56	0.85	0.12	0.16
Density (cells L ⁻¹)	8000	80000	25000 \pm 15940	7000	95000	37890 \pm 25390	0.99	0.11	*0.01

* Significant at $P < 0.05$.

area comprising values between 30.33 (P3, rainy period, low tide), and 37.43 (P1, dry period, high tide), while the temperature variation is slight, ranging from 26 $^{\circ}$ C to 29 $^{\circ}$ C (Fig. 3a).

There was a significant difference between tides for the variable depth ($p = 0.00$); during low tides the P2 was the most shallow (in both weather periods), with a minimum depth of 0.90 m (Table 1). On the other hand, the P1 during high tide (dry period) showed maximum depth of 3.85 m. Water transparency, in turn, showed a significant difference between seasons and tides ($p = 0.02$ and $p = 0.00$, respectively, Table 1), ranging from 0.70 m (P2, rainy period, high tide) and 3.40 m (P1, dry period, high tide) (Fig. 3b).

The other studied environmental parameters, such as suspended particulate matter (SPM), dissolved oxygen and hydrogen potential showed no significant differences for the studied treatments. The SPM had the minimum and maximum values recorded during the high tides of the dry period, ranging from 1.78 mg L⁻¹ to 46.43 mg L⁻¹ (P2 and P3, respectively, Fig. 4a). The dissolved oxygen

ranged between 3.40 mL L⁻¹ (P1, rainy period, high tide) to 6.39 mL L⁻¹ (P1, dry period, low tide, Fig. 3c) and being near or above the saturation point (maximum of 145 %, Table 1). The hydrogen potential remained alkaline, with a small variation from 7.91 (dry period) to 8.46 (rainy period, Fig. 3c), both during low tides, in P3.

Among the nitrogen compounds, ammonia and nitrate showed no significant differences, while nitrite was different only seasonally (Table 1). The concentration of ammonia ranged from values below the limit of detection to 0.06 μ M. The undetected limits were recorded at P1, during all low tides of the period studied (Fig. 4a). Nitrate in turn presented the minimum of 0.20 μ M (P3 and P2, dry period, low tide and high tide, respectively). Stand out peaks of 15.66 μ M and 18.20 μ M, which occurred during high tide, in the dry season and the points P1 and P2, respectively (Fig. 4b). Regarding the concentrations of nitrite, their values were from below the limit of detection to 0.23 μ M (P2, rainy period, low tide, Fig. 4b).

For the other studied nutrients, there was signi-

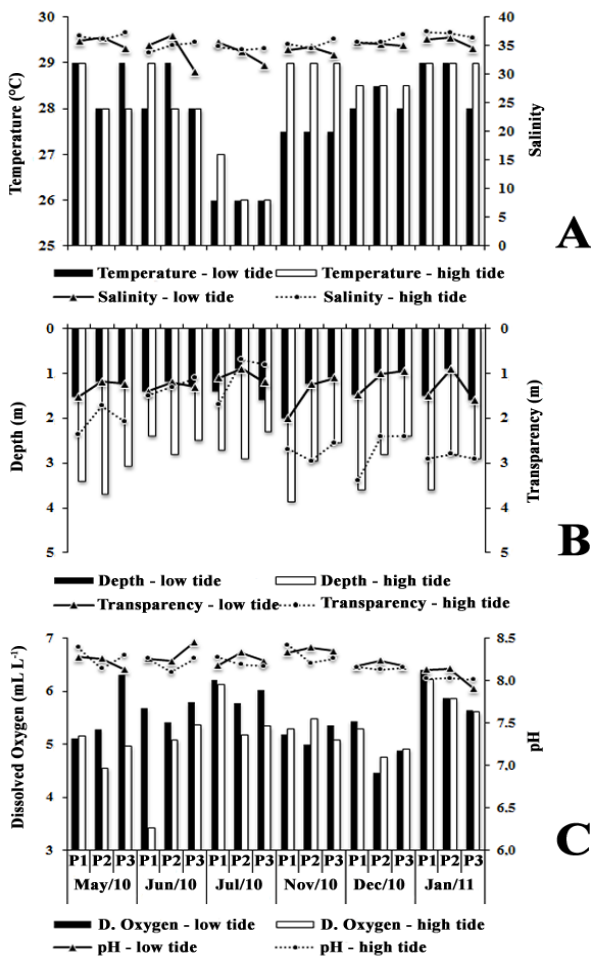


Fig. 3. Variation of temperature, salinity (A), depth, water transparency (B), dissolved oxygen, pH (C), during the rainy and dry periods, in the reef ecosystem of São José da Coroa Grande, Pernambuco, Northeastern Brazil.

significant seasonal difference for phosphate and silicate (Table 1). The minimum of $0.04 \mu\text{M}$ (P3, dry period, low tide) and the maximum of $0.29 \mu\text{M}$ (P1, P2 and P3, rainy period, high tide and low tide) were observed for the phosphate, whereas the concentration of silicate was distributed from $3.60 \mu\text{M}$ (P1, rainy period, low tide) to $42.50 \mu\text{M}$ (P3, rainy period, low tide) (Fig. 4c).

Concentration of chlorophyll a

The levels of total phytoplankton biomass (chlorophyll a) ranged from below the detection limit (P1, low tide, dry period) to 8.79 mg m^{-3} (P3, rainy period, high tide), with no significant differences (Table 1). Thus, the studied reef ecosystem ranged from oligotrophic to eutrophic, with a mesotrophic trend.

Regarding the contribution of the different fractions of the phytoplankton community in São José da Coroa Grande (Fig. 5), the fraction $< 20 \mu\text{m}$ (nano and picoplankton) was predominant and accounted for 81 % of total chlorophyll a, as indicated by high the correlation in the PCA (Fig. 6). Their concentrations ranged from values below the detection limit to 5.70 mg m^{-3} (P3, dry period, high tide). However, the fraction $> 20 \mu\text{m}$ (microphytoplankton) corresponded to 19 % of total chlorophyll a without significant differences. Their concentrations ranged from values below the detection limit until 3.42 mg m^{-3} (P2, rainy period, high tide).

Phytoplankton density

Quantitative analysis revealed a phytoplankton community consisting of 47 taxa (Table 2), among which the group that most contributed was Heterokontophyta / Bacillariophyceae, corresponding to 80.9 % of the total identified species. The phylum Dinophyta was represented by 8.5 % of the total, followed by Cyanobacteria (6.4 %), Euglenophyta and Chlorophyta (2.1 % for both).

The total number of cells per liter in the two annual periods remained between $8 \times 10^3 \text{ cells L}^{-1}$ to $9.5 \times 10^4 \text{ cells L}^{-1}$, it is noteworthy that the extreme values were obtained during the dry season, at low tide, P3. Significant difference was observed between periods of tides (Table 1).

Diatoms presented the main contribution to the density of phytoplankton (Table 3), as they were present in all sampling sites and seasons, reaching a maximum density of $8.2 \times 10^4 \text{ cells L}^{-1}$ in the dry period, high tide, P3, and a minimum density of $4 \times 10^3 \text{ cells L}^{-1}$ in the dry period, low tide, P1.

The dinoflagellates were most significant during the dry period, especially the species *Prorocentrum lima* (Ehr.) Dodge, in the month of January 2011, with $4.6 \times 10^4 \text{ cells L}^{-1}$, low tide, P3. The phylum Cyanobacteria had quantitative representation during rainy period, reaching $8 \times 10^3 \text{ cells L}^{-1}$ at high tide in P1. Other groups, such as Euglenophyta and Chlorophyta, little influenced the increase in the number of cells L^{-1} in the samples, as they occurred sporadically, not reaching values above $2 \times 10^3 \text{ cells L}^{-1}$.

Multivariate analysis

The factorial plan 1 - 2 explained 40.9 % of the variance of the data (Fig. 6). The component 1 explained 22.0 %, showing a direct correlation with

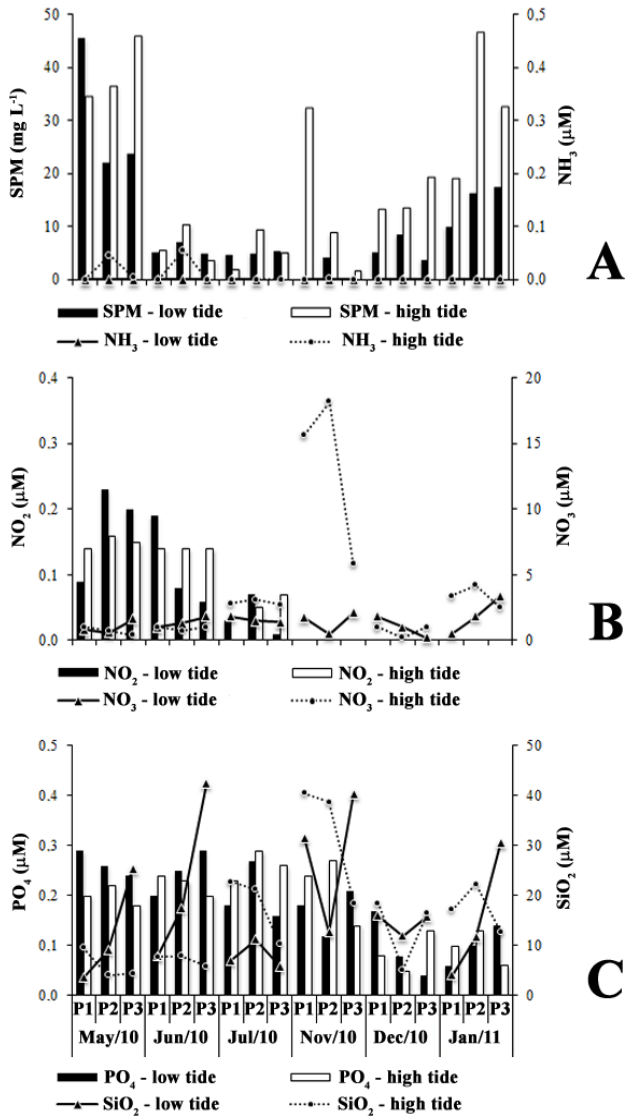


Fig. 4. Variation of suspended particulate matter (SPM), ammonia-NH₃ (A), nitrite-NO₂, nitrate-NO₃ (B), phosphate-PO₄³⁻ and silicate-SiO₂ (C) during the rainy and dry periods, in the reef ecosystem of São José da Coroa Grande, Pernambuco, Northeastern Brazil.

dissolved oxygen and its saturation rate, and inverse correlation with pH, total chlorophyll *a*, chlorophyll *a* < 20 μm and chlorophyll *a* > 20 μm. The component 2 explained 18.9 %, showing a direct correlation with phytoplankton density, temperature, salinity, depth, water transparency, suspended particulate matter, and inverse correlation with silicate. The component 3 explained 12.5 %, showing a direct correlation with nitrate and inverse correlation with ammonia, nitrite and phosphate.

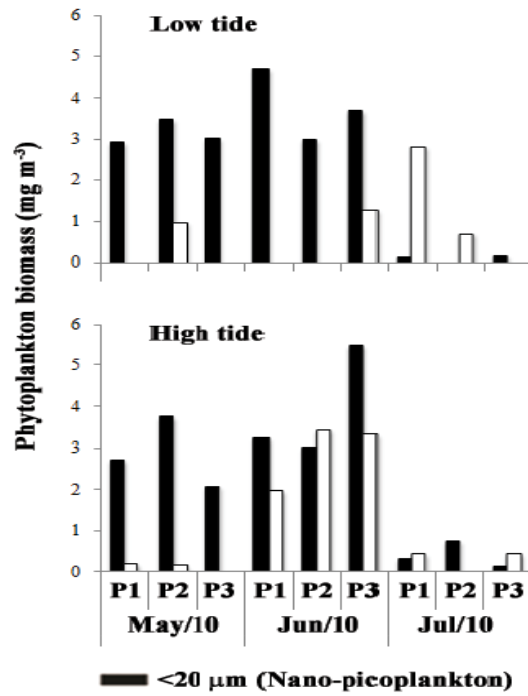


Fig. 5. Fractionated phytoplankton biomass during rainy and dry periods, in the reef ecosystem of São José da Coroa Grande, Pernambuco, Northeastern Brazil.

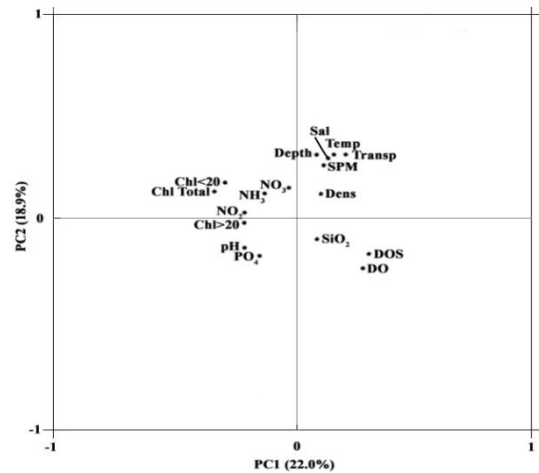


Fig. 6. Principal component analysis (PCA) of environmental variables analyzed in the reef ecosystem of São José da Coroa Grande, Pernambuco, Northeastern Brazil, for the two principal components - (Chl Total) total chlorophyll *a*, (Chl > 20) chlorophyll *a* > 20 μm, (Chl < 20) chlorophyll *a* < 20 μm, (Dens) phytoplankton density, (DOS) saturation rate of dissolved oxygen, (DO) dissolved oxygen, (NO₂) nitrite, (NO₃) nitrate, (NH₃) ammonia, (PO₄) phosphate, (Sal) salinity, (SiO₂) silicate, (SPM) suspended particulate matter, (Temp) temperature, (Transp) water transparency.

Table 2. List of phytoplankton species in the reef ecosystem of São José da Coroa Grande, Pernambuco, Northeastern Brazil.

	Cyanobacteria
<i>Oscillatoria</i> sp.	<i>Spirulina</i> sp.
<i>Pseudoanabaenopsis</i> sp.	
	Euglenophyta
<i>Trachelomonas</i> sp.	
	Dinophyta
<i>Gymnodinium</i> sp.	<i>Prorocentrum lima</i> (Ehr.) Dodge
<i>Podolampas spinifera</i> Okamura	<i>Protoperidinium</i> sp.
	Ochrophyta
<i>Amphora</i> sp.	<i>Licmophora</i> sp.
<i>Asterionellopsis glacialis</i> (Castracane) Round	<i>Melchersiella hexagonalis</i> C.Teixeira
<i>Asterionella</i> sp.	<i>Navicula</i> sp.
<i>Biddulphia biddulphiana</i> (J. E. Smith) Boyer	<i>Nitzschia</i> sp.
<i>Campyloneis grevillei</i> (W. Smith) Grunow & Eulenstein	<i>Odontella aurita</i> (Lyngbye) C. Agardh
<i>Catacombas gaillonii</i> (Bory) D.M. Williams & Round	<i>Paralia sulcata</i> (Ehrenberg) Cleve
<i>Cerataulina pelagica</i> (Cleve) Hendey	<i>Pleurosigma rigidum</i> W. Smith
<i>Cerataulus turgidus</i> (Ehr.) Ehr.	<i>Pleurosigma</i> sp.
<i>Chaetoceros</i> sp.	<i>Proboscia alata</i> (Brightwell) Sundström
<i>Climacosphenia moniligera</i> Ehr.	<i>Psammodictyon panduriforme</i> (W.Greg.) Mann
<i>Cocconeis scutellum</i> Ehr.	<i>Podocystis adriatica</i> (Kützing) Ralfs in Pritchard
<i>Coscinodiscus centralis</i> Ehr.	<i>Rhabdonema adriaticum</i> Kützing
<i>Coscinodiscus</i> sp.	<i>Rhaphoneis amphiceros</i> var. <i>tetragona</i> Grunow
<i>Diploneis</i> sp.	<i>Surirella fastuosa</i> (Ehr.) Ehr.
<i>Fragillaria</i> sp.	<i>Striatella striata</i> (Wigand) Rabenhorst
<i>Grammatophora marina</i> (Lyngbye) Kützing	<i>Surirella febigerii</i> F. W. Lewis
<i>Grammatophora</i> sp.	<i>Synedra</i> sp.
<i>Isthmia enervis</i> Ehr.	<i>Thalassionema nitzschioides</i> (Grunow) Mereschkowsky
<i>Licmophora abbreviata</i> C. Agardh	<i>Triceratium pentacrinus</i> (Ehr.) Wallich
	Chlorophyta
<i>Scenedesmus</i> sp.	

Discussion

The tropical waters that bathe the coastal reefs are crystal clear, warm and poor in nutrient salts (Richmond 1993), with small variations in temperature and salinity, with little influence on the quantitative variation of phytoplankton (Passavante & Feitosa 2004; Rosevel da Silva *et al.* 2005). This pattern is usually found in coastal reefs of northeastern Brazil.

The reef ecosystem of São José da Coroa Grande showed significant seasonal variation in water transparency and nutrient salts (nitrite, phosphate and silicate) influenced by rainfall, which acts in the transport of these nutrients to the coastal zone. The highest levels of nutrient salts and the reduction of water transparency were recorded during the months of highest rainfall, related to the river and marine input (higher

intensity of turbulence and waves) and revolving of the sediment. This pattern is also observed in surf zones (Ferreira *et al.* 2010) and reef environment (Fonseca *et al.* 2002; Machado *et al.* 2007) on the South coast of Pernambuco.

For our study, this observation is valid, proved through the PCA, in which the SPM was directly associated with depth and water transparency, besides temperature, salinity and phytoplankton density.

According to Diehl *et al.* (2002), in aquatic ecosystems a direct relationship between phytoplankton biomass and extremely shallow environments occurs. Water transparency has, therefore, an important role for reef organisms with high light requirement, as numerous species of macroalgae, coral symbiotic dinoflagellates (zooxanthellae) and the community of planktonic and benthic microalgae (Dubinsky 1990).

Table 3. Phytoplankton density (cells L⁻¹) in the reef ecosystem of São José da Coroa Grande, Pernambuco, Northeastern Brazil - (P1, P2 and P3) sampling points, (LT) low tide, (HT) high tide.

May/2010	P 1		P 2		P 3	
	LT	HT	LT	HT	LT	HT
Cyanobacteria	2000	-	-	2000	4000	-
Euglenoids	-	-	-	-	-	-
Dinoflagellates	-	8000	-	6000	-	6000
Diatoms	14000	22000	20000	16000	16000	18000
Chlorophytes	-	-	-	-	-	-
TOTAL	16000	30000	20000	24000	20000	24000
June/2010						
Cyanobacteria	-	8000	-	2000	-	2000
Euglenoids	-	-	-	-	-	-
Dinoflagellates	-	6000	-	-	-	-
Diatoms	18000	66000	10000	18000	8000	16000
Chlorophytes	-	-	-	-	-	-
TOTAL	18000	80000	10000	20000	8000	18000
July/2010						
Cyanobacteria	-	2000	2000	2000	-	-
Euglenoids	-	-	-	-	-	-
Dinoflagellates	-	-	-	-	-	-
Diatoms	28000	34000	26000	18000	12000	38000
Chlorophytes	-	-	-	-	-	-
TOTAL	28000	36000	28000	20000	12000	38000
November/2010						
Cyanobacteria	-	-	-	-	-	-
Euglenoids	-	-	2000	2000	-	-
Dinoflagellates	8000	-	2000	8000	6000	12000
Diatoms	10000	82000	18000	68000	38000	26000
Chlorophytes	-	-	-	-	-	-
TOTAL	18000	82000	22000	78000	44000	38000
December/2010						
Cyanobacteria	-	2000	-	-	-	-
Euglenoids	-	-	-	-	-	-
Dinoflagellates	12000	2000	12000	2000	-	-
Diatoms	4000	12000	18000	34000	24000	18000
Chlorophytes	-	2000	2000	-	-	-
TOTAL	16000	18000	32000	36000	24000	18000
January/2011						
Cyanobacteria	3000	-	-	2000	4000	-
Euglenoids	-	-	-	-	-	-
Dinoflagellates	2000	1000	1000	19000	46000	-
Diatoms	15000	39000	6000	40000	45000	30000
Chlorophytes	-	-	-	2000	-	1000
TOTAL	20000	40000	7000	63000	95000	31000

The oxygenation of the water is favored by the occurrence of autotrophic organisms on reefs (phytoplankton, phytobenthos, macroalgae and zooxanthellae), associated with local hydrodynamics and the effect of waves, allowing greater ocean-atmosphere interaction, contributing to a

saturated condition in dissolved oxygen. The saturation rate of dissolved oxygen, above 100 %, shows an area of low contribution of anthropogenic organic matter, taking into account the classification of Macêdo & Costa (1978). This fact is also observed in other reef areas in the northeast of

Brazil, as in Ponta do Seixas, State of Paraíba and Maracajaú, State of Rio Grande do Norte (Mayal *et al.* 2009; Sassi *et al.* 1990).

Regarding the variation of the pH, this variable is influenced by the chemical tidal cycle, photosynthesis and respiration rate (Flores-Montes *et al.* 1998), where the consumption of carbon dioxide and the release of oxygen by phytoplankton increase these values. Therefore, the pH data found for the site under study, besides confirming the strong marine influence, also suggest a high photosynthetic activity and low contribution of organic matter.

The concentrations of nitrite, nitrate, ammonia, phosphate and silicate, have high importance in the aquatic environment, because, along with the light, they act as a major limiting factors to the production and density of chlorophyllous organisms, interfering across the trophic web (Macêdo *et al.* 2004). In oligotrophic coastal areas, the input of dissolved compounds via continental drainage, associated to rainfall, result in temporary changes in the concentrations of these components in the sea water, especially nutrient salts (Braga *et al.* 2000). During the rainy period, our study showed higher concentrations of nitrite, phosphate and silicate as a result of greater continental discharge, due to the proximity of the Persinunga river.

Regarding the silicate concentrations in the water, this nutrient, mainly of continental origin, is of great importance in the primary productivity in certain algal groups (Wu & Chou 2003), such as diatomaceous, dominant group in the study area, presenting higher genus diversity with wide distribution.

Eskinazi-Leça *et al.* (1997) argue that temporal changes present significant differences in the distribution and abundance of phytoplankton, mainly regulated by the discharge of rivers, tidal cycle and rainfall. In the studied reef ecosystem, the seasonal and tidal driving forces induce changes in chemical and biological properties, primarily with regard to phytoplankton cell density. Other studies have reported the effect of seasonality and tidal periods for the distribution and composition of coastal planktonic communities (Ferreira *et al.* 2010; Mayal *et al.* 2009; Totti *et al.* 2000).

Little is known about the phytoplankton community in reef areas and studies on this subject commonly discuss how their composition differs from adjacent waters (Ferrier-Pagés & Gattuso 1998; van Duyl *et al.* 2002; Yahel *et al.*

1998). In northeastern Brazil, some studies have used chlorophyll a as the main indicator in reef waters (Bastos *et al.* 2011; Feitosa & Bastos 2007; Jales *et al.* 2012; Machado *et al.* 2013), with the taxonomic groups, the quantification of cell and the algal sizes being rarely addressed in this environment.

Studies show that water components (such as nutrient salts and planktonic organisms) on reefs are subjected to the trophodynamics action of the ecosystem, in addition to human activities in adjacent waters. In this regard, van Duyl *et al.* (2002) in their comparative study of the waters on coral reefs and adjacent waters (open ocean and coastal area) in the Caribbean Sea, found that phytoplankton composition undergoes changes between passages from the shore to the reef and from the reef to the open ocean, following a decreasing gradient (toward shore-reef-ocean) in cell size of centric diatoms, as well as in the dominance of diatoms and dinoflagellates. Thus, the microphytoplankton (> 20 µm) was more representative in the reef area, when compared to ocean and coastal waters.

When compared to other fractions of the community, according to Ferrier-Pagés & Gattuso (1998), in their experiments on reef waters in Japan, microphytoplankton appears in lower contribution to the autotroph biomass, with only 0 to 45 % of the total, with the diatoms *Nitzschia* sp., *Rhizosolenia* sp., *Skeletonema* sp. and *Coscinodiscus* sp. being only recorded at densities above 3×10^4 cells L⁻¹. As demonstrated in the literature, the main components of marine phytoplankton correspond to the pico and nanoplankton, commonly exceeding 70 or 90 % of total chlorophyll a (Detmer & Bathmann 1997; Jochem & Zeitzschel 1993a,b; Olson *et al.* 1990) and can reach a maximum of 6×10^5 cells L⁻¹ (Ferrier-Pagés & Gattuso 1998).

In the reef area of São José da Coroa Grande, we showed that the contribution of the fraction < 20 µm (nano and picoplankton) was predominant and accounted for 81 % of total chlorophyll a, showing a direct correlation with the same, as indicated in the PCA, and a maximum density of 9.5×10^4 cells L⁻¹.

van Duyl *et al.* (2002) reported an eventual increase in algal biomass in some points on the reefs and explained the accumulation of diatoms, by decreasing the time of water renewal between the reefs and adjacent waters, prolonging the retention time of water on the reef. In our study, cell density in São José da Coroa Grande

presented significant differences between tides, registered in greater proportion during low tide at the sampling point near the Persinunga river.

However, at other points, a reduction in the number of smaller algal cells was noticed, especially of the cyanobacterium *Synechococcus* sp., i.e., there was a selective removal of cell size, explained by the grazing caused by experts benthic filter feeders, which keeps the density of phytoplankton at critical levels. This phenomenon is supported by Yahel *et al.* (1998), who found marked reductions, about 15 to 65 % of decline in the abundance of phytoplankton and total chlorophyll a, compared to the adjacent oceanic waters, while studying the distribution of phytoplankton and grazing near coral reefs located in the Red Sea.

The occurrence of low concentrations of chlorophyll a and phytoplankton cells on the reefs, as in our study area, can be explained by the removal of phytoplankton by benthic community, especially in reefs inhabited by colonies of soft corals, herbivorous organisms responsible for the high removal of phytoplankton in these environments.

Yahel *et al.* (1998) also reported that the reefs they studied are composed of hermatypic corals, which are not known as phytoplankton filters. Therefore, the authors state that not only reef organisms may have contributed to the grazing, but also inhabitants of sandy sediment, as well as the water column herbivores (zooplankton). Likewise, such possibilities are applied to our study, in which chlorophyll a cell density were low for both weather periods, at all points, with a significant difference only between tides for the number of cells per liter.

The study of van Duyl *et al.* (2002) discusses the influence of human activities in waters adjacent to reefs. These authors recorded at some points of the reef an algal biomass more concentrated due to the nutrients arising from the impacted coast. However, our study is focused on human actions on the reefs, and no anthropic effect was observed on phytoplankton, since the algal biomass and density were low, together with the low nutrient concentrations, suspended particulate matter and other environmental parameters analyzed. Additionally, such a statement is affirmed based on the definitions of Eskinazi-Leça *et al.* (2011), demonstrating that marine ecosystems with density values below 5×10^5 cells L⁻¹ are considered free of anthropogenic environments.

The low density of phytoplankton cells in the reef area of São José da Coroa Grande can be explained by the trophodynamic action, through the grazing on the phytoplankton (Cloern 1982; Ferrier-Pagés & Gattuso 1998; Yahel 1998).

In addition, Ferreira *et al.* (2010) in their study of the temporal variability of phytoplankton along the surf zone of urban beaches in northeastern Brazil, reported algal blooms during the dry period, reaching 2.3×10^6 cells L⁻¹, without patches. It is worth mentioning the occurrence of several bands of sandstone reefs on these shores, heavily impacted by human activities (tourism, fishing, housing speculation, etc.). These activities were also observed in São José da Coroa Grande, but with a lower intensity.

Conclusions

Therefore, our study demonstrates that human activities in the studied reef environment are not enough to change the local phytoplankton community. Thus, the discrete and eventual changes observed in some variables (such as phytoplankton biomass and density) can result from the action of trophodynamics and of water adjacent to the reefs.

Studies in this environment can be used as a subsidy for current and future conservation concern in the study area, with monitoring of human activities, so that they do not increase enough to reach the carrying capacity of the ecosystem.

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