Changes in Phytoplankton Communities in a Tropical Estuary in the Colombian Caribbean Sea



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Abstract

Phytoplankton community species composition, diversity, biomass, and distribution experience constant changes because of seasonal and temporal variations. This study was done with the aim of describing the response to environmental and seasonal changes of the phytoplankton communities of a tropical estuary in the south of the Colombian Caribbean Sea (Gulf of Urabá) with an emphasis on potential producers of toxin genera. To do these, 15 sites along the Gulf were studied during two cruises in the rainy season and one during the dry season. In each site, water samples from above and below the halocline were collected. Environmental factors such salinity, pH, dissolved oxygen, and nutrient concentration were correlated with diatoms, dinoflagellate, cyanobacteria, and biotoxin producer abundance. The results showed that the number of diatoms was at a maximum of 11,166 cell/L in the fluvial zone. Dinoflagellates were at a maximum of 4250 cell/L in the same zone during the dry season and cyanobacteria blooms during the rainy season. Four genera of potential biotoxin producers were found: *Dolichospermum, Prorocentrum, Dinophysis*, and *Pseudo-nitzschia*; this last genus represents 44% of the total diatom abundance during the rainy season with a detectable domoic acid production in a range between 25.54 and 1580.7 pg/mL; this substance can affect different trophic levels in the Gulf, especially mammals due to its non-reversible amnesic effect. Overall, this study shows that the phytoplankton community structure in this tropical stratified estuary presents environment conditions during the rainy season that increase the abundance of phytoplankton that may thrive into blooms.

Keywords Phytoplankton · Tropical estuaries · Harmful algae · Community dynamics · Caribbean Sea · Domoic acid

Introduction

Phytoplankton represent more than 45% of the net primary production of the planet and the first trophic level in marine food webs (Hernández 2014; Martínez et al. 2013; Vajravelu et al. 2018). Phytoplankton are the largest producers of oxygen on the planet and comprise approximately 5000 species in marine and estuarine waters, including chlorophytes, dinoflagellates, cyanobacteria, and diatoms (Cloern 1996; Hallegraeff 1993; Hernández 2014). Due to their importance,

² Biotechnology Research Group, Biology Institute, Universidad de Antioquia UdeA, Calle 70 No. 52-21, Medellín, Colombia small variations in the phytoplankton community structure produce large impacts on ecosystems (Dogliotti 2007).

The taxonomic composition of microalgal communities and their biomass usually change when the concentrations of essential nutrients (N, C, P, O, Fe, Si) increase (i.e., eutrophication); phytoplankton communities will increase their growing rates, which leads to blooms that can affect ecosystems (Anderson et al. 2002; Hallegraeff 1993; McCabe et al. 2016; Smayda 1997; Vajravelu et al. 2018; Wells et al. 2015). When blooms occur, the required quantity of oxygen becomes higher than the production rate because of decomposition of organic matter, and this can promote anoxia events and the death of organism of commercial interest (Carstensen et al. 2015; Zilius et al. 2014). During blooms, some biotoxin producer species can thrive, producing harmful algae blooms (HABs) that cause ecological and anthropogenic negative impacts (Hallegraeff 1993; Sar et al. 2002).

Knowledge on the variation of phytoplankton communities and on the presence of potential biotoxin producer genera such as *Pseudo-nitzschia*, *Dinophysis*, *Dolichospermum*, and *Prorocentrum* is a useful tool for establishing appropriate

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management strategies (Barton et al. 2016) in order to prevent the consequences of harmful blooms. The relative abundance of phytoplankton also responds to environmental changes for example, variation in the thickness of the mixed layer, runoff, water surface temperature, salinity, and resuspension induced by waves and winds (Carstensen et al. 2015; Gobler et al. 2012; Jakobsen et al. 2015; Muylaert and Sabbe 1999; Muylaert et al. 2009; Smayda 1990; Vajravelu et al. 2018; Wells et al. 2015).

In tropical ecosystems, particularly in estuaries, rainfall and mostly salinity are fundamental modulators of the abundance dynamics and distribution of the phytoplankton community species (de Affe et al. 2018; Flöder et al. 2010; Masmoudi et al. 2015; Oliver et al. 2010; Pednekar et al. 2018). Thus, salinity is a key variable to understand distribution of this community. Tropical estuaries are usually rich in nutrients due to river discharges, becoming mesotrophic to eutrophic systems (Huang et al. 2012; Mahoney and Bishop 2017), which influence phytoplankton growth rates and biomass production (Glibert 2016; Reynolds 2006), although they represent a small percentage of the Earth's surface (Cloern et al. 2014). The northern region of Colombia has two estuaries with high nutrient loads and high productivity in which algal blooms can occur (Blanco et al. 2006; Cloern et al. 2014; INVEMAR 2017a, b). However, the Gulf of Urabá, the largest estuary, has been poorly studied. Considering tropical estuary dynamics, we hypothesized that phytoplankton community genus composition, diversity, abundance, and distribution present changes linked to the nutrients and environmental conditions of each season.

Methods

Study Site

The Gulf of Urabá is a semi-closed estuarine area located in the southwestern Colombian Caribbean between $7^{\circ} 55'-8^{\circ} 40'$ N and $76^{\circ} 53'-77^{\circ} 23'$ W, with a length and width of ca. 80 km and ca. 25 km, respectively (Fig. 1). The Gulf has high sedimentation rates from the second largest river in Colombia (the Atrato River, rate close to 11 ton/year), and these freshwater flows are mixed with waters of the Caribbean Sea (Montoya 2010). Also, the gulf is recognized as a stratified estuary with salinity differences ranging from 7 to 35 psu (Montoya et al. 2017).

Additionally, the estuary has depths between 2 and 80 m, with an average depth of 34 m (François et al. 2007), and a regional precipitation around 2500 mm/year. The intertropical convergence zone (ITCZ) is the main physical mechanism that modulates the hydro-climatology of the region. As result, there are two main climatic seasons defined by the precipitation and circulation patterns during the year (Chevillot et al.

1993). The dry season occurs between December and April and is generally characterized by low precipitation levels influenced by high-intensity trade winds. In contrast, the rainy season is experienced between May and November, and is characterized by higher values of precipitation and weak winds from the southeast (Chevillot et al. 1993). Briefly, the Gulf is governed by three current systems: the Caribbean, the deep Caribbean, and the Panamá-Colombia countercurrents (Andrade et al. 2003). The Panamá-Colombia countercurrent is the system of currents that modulates the hydrodynamics of the Gulf in both seasons of the year. During the dry season, this current extends from the Gulf of Darien to the mouth of the Magdalena River, while in the rainy season, the pattern of this current extends to the peninsula of La Guajira (Bernal et al. 2006; Lozano-Duque et al. 2010; Pujos et al. 1986).

Field Sampling and Methodology

The regional climatology exhibited climatic seasons; therefore, to assess changes in phytoplankton communities in the estuary, three cruises were done, two during the rainy season (May 5 and October 20, 2018) and one in the dry season (March 25, 2019). The values of the two samplings during the rainy season were averaged. During these cruises, 11 sites were sampled in order to measure the biological parameters at 15 sites for physicochemical characteristics. Data of continuous vertical profiles were obtained through a CastAway CTD V 1.60 model updated and calibrated by the manufacturer. The data set of annual precipitation for the period 2000-2016 at the closer IDEAM meteorological stations was taken from the National Meteorological Information Center of Colombia (http://www.ideam.gov.co/). The locations of the meteorological stations and their measurements are given in Figs. 1 and 2.

Water samples (12 L using Niskin bottles) were collected above and below the halocline points and were used to determine the concentrations of macronutrients (PO_4^{-3} , SiO_2 , NO₃⁻), chlorophyll a, and phytoplankton abundance. To measure chlorophyll a, water samples of 1 L above and below the halocline were collected in duplicate, refrigerated, and analyzed 24 h after collection using the analytical colorimetric method SM-10200-H with 90% acetone for seawater, following the protocols proposed by Aguirre Gómez and Salmerón García (2015). Phosphate (PO_4^{-3}) levels were determined using the ascorbic acid method (SM-4500-P), opal levels were determined via atomic absorption (SM-3111-D, iCE 3300 Thermo scientific), and for nitrate levels, the cadmium reduction method (SM 4500-NO3-) was used. Measurements were done following the methodology proposed by the Standard Methods for Examination of Water and Wastewater (APHA 2012). A 100 mL sample of surface water was taken in order to measure domoic acid, and the samples were preserved at 4 °C in amber plastic bottles. An ELISA kit from Bioscience



Fig. 1 Location of sampling sites, defined in the study in the Gulf of Urabá, Colombian Caribbean Sea

was used to quantify total domoic acid, and the samples were sonicated and filtered (0.2 μ m) following manufacturer's instructions. This method has a detection limit between 0.16 pg/mL and 10,000 pg/mL of domoic acid (Trainer et al. 2002). Dissolved oxygen and pH were measured using a HANNA

HI-98194 multi-parameter, while transparency (m) was measured using a Secchi disk.

To quantify phytoplankton cell abundance (cells/L), three samples of 100 mL were taken from the surface of water below the halocline and preserved with 1% Lugol at 4 °C,



Fig. 2 Annual variability of precipitation in the Urabá region, Caribbean Colombian. Lines represent average precipitation values for San Juan de Urabá (), Acandí (), Tanela (), Titumate (), and Arboletes () IDEAM meteorological sites considering the experience of preliminary sampling done in the Gulf by the authors. In addition, three aliquots of 1 mL of each sample were quantified using Sedgewick Rafter counting chambers in an inverted microscope (Optika IM-3 fluorescence). The whole chamber was examined to obtain the concentration in 1 mL, and the results were reported in cell per liter. To determine the genus phytoplankton and its quantity in the community in each site (richness), samples were taken by horizontal and vertical trawls for 3 min with phytoplankton nets with pore sizes of 25 µm and 60 µm. The samples were preserved with Lugol (1%) at 4 °C, three aliquots of 1 mL for each sample were evaluated in a Sedgewick Rafter, and the whole chamber was examined. The observation and identification of the phytoplankton genera was carried out following the guides proposed by Tomas (1997) and Vidal Velásquez (2010) and confirmed using the AlgaeBase database resource (https://www.algaebase.org/). An additional sample of 500 mL of water from sampling sites located close to the Atrato River was sent to Dra. Vera Trainer, at the laboratories of the NOAA's Northwest Fisheries Science Center, in order to identify Pseudo-nitzschia species via scanning electron microscopy.

Data Processing and Statistical Analysis

To analyze the data, three zones were defined, considering the surface salinity of each site with the Ward's method using quadratic Euclidean distance as a grouping method with IBM SPSS Statistics v. 25 software. The values of the nutrients (except opal), physicochemical parameters, and phytoplankton counts obtained at each site were used to obtain an average measurement of the variables for describing each zone, and this average was the statistic mean of the data. Furthermore, standard deviation and the statistic differences of the variables at the sites per zone were calculated. Phytoplanktonic counts were also analyzed per site, due to the high deviation observed between sites inside the defined zones.

A descriptive analysis of the data was made in order to evaluate the main statistics. Additionally, we used the Kolmogorov-Smirnov test, the Levene's tests, and residual plots for verification of normality, homogeneity of variance, and independence, respectively, in order to determine if the variables were parametric and to define the statistical method to compare them. An ANOVA was carried out in order to verify the significant differences in the parametric variables. When non-compliance with assumptions of normality, homogeneity, or independence was found, a Kruskal-Wallis (KW) test was used to evaluate the possible statistically significant differences in the physicochemical properties, nutrients, and biological variables for the hydro-climatological zones. KW is shown as a sub-table in the ANOVA results. Abundance data were transformed with natural logarithm. Finally, a Spearman correlation test was done in order to evaluate and correlate the structure of the community with the concentration of nutrients and other physicochemical variables. The correlations were considered significant when they showed a rho (ρ) value higher than |0.7| and a *p* value for the spearman correlation lower than 0.05.

Results

Environmental Conditions

Differences in the salinity measured above the halocline were found between the sites in both seasons, with a range between 0.15 and 36.25 psu (p < 0.001, n = 42, KW). Taking as reference, the obtained salinity values, three zones were established per season. For the rainy season: (1) The fluvial zone from sites 1 to 10 $(4.45 \pm 3.00 \text{ psu})$, (2) the estuarine zone from sites 11 to 13 (11.19 ± 1.26 psu), and (3) the oceanic zone, with sites 14 and 15 (31.05 ± 0.89 psu). In the dry season, the fluvial zone was composed of sites 1 to 7 (6.51 \pm 1.49 psu), the estuarine zone was composed of sites 8 to 12 $(17.83 \pm 4.04 \text{ psu})$, and the oceanic zone of sites 13 to 15 $(31.50 \pm 0.89 \text{ psu})$. Salinity measurements above the halocline increased with proximity to the Caribbean Sea and showed higher average values during the rainy season (p = 0.01, n =42, KW) (Tables 1 and 2). Estuary stratification was noted with density and Brunt-Väisälä analyses (Figs. 3 and 4).

The defined zones exhibited similar salinity in the measurements done below the halocline during the rainy season (Tables 3 and 4, Fig. 5a), with an average of $34.58 \pm$ 1.40 psu (p = 0.41, n = 42, KW). During the dry season, the values of salinity below the halocline were similar between the fluvial and estuarine zones, with an average of $25.33 \pm$ 1.44 psu (p = 0.33, KW), but were different from the oceanic zone, with a salinity value below the halocline of $31.97 \pm$ 1.25 psu (p = 0.01, KW) (Tables 3 and 4, Fig. 5b). The temperature had higher values during the rainy season, ranging between 28.56 ± 0.24 °C and 28.77 ± 0.30 °C compared with the dry season, with values ranging between 27.69 ± 0.02 and 27.95 ± 0.07 °C (p < 0.001, n = 42, KW). Temperature values were similar in all the zones for both seasons (p = 0.829, KW) (Table 1, Fig. 5c, d), with small variations observed as the depth increased. During the dry season in the oceanic and estuarine zone, two currents with water cooler than the surface water where detected in the water column (Fig. 5d).

Transparency was lower in the rainy season, with values between 1.08 ± 0.27 m and 7.35 ± 2.05 m compared with the dry season, with values between 1.80 ± 0.44 m and 8.56 ± 1.57 m (p < 0.001, n = 27, AN, Table 1), with higher values in the oceanic zone and lower values in the fluvial zone for both seasons (p < 0.001, n = 42, AN, Table 1). The pH presented a minimum value of 7.83 ± 0.32 , corresponding to the measurement made below the halocline in the fluvial zone

| Table 1 A letters A and values under | verage value: 1 B correspor r detection lir | s of physicoche nd to measuren nit | emical variables _] nents above and | per season and below the hal | l zone. Transpa locline water n | arency (Trans, r espectively. Va | n), temperature (Te lues inside parentl | emp, °C), salin hesis represent | ity (Sal), dissol standard devi | lved oxygen (DO) ation of the avera |), pH, NO ³⁻ , a ige, * values ; | are PO_4^{-3} are slare starts for sites wi | hown. The th $n = 1$ or |
|---------------------------------------------|---------------------------------------------------|--------------------------------------------|--------------------------------------------------|----------------------------------|------------------------------------|--------------------------------------|--------------------------------------------|------------------------------------|------------------------------------|------------------------------------------|------------------------------------------------|-----------------------------------------------|------------------------------------------|
| | Trans (m) | Temp (°C) | Sal A (psu) | Sal B (psu) | Average Sal (psu) | DO A (mg/L |) DOB (mg/L) | pH A | pH B 1 | NO ₃ ⁻ A (mg/L) | NO ₃ ⁻ B (mg/L) | $PO_4^{3-}A = I$ (mg/L) (| PO ₄ ^{3–} B mg/L) |
| Rainy seaso | n 1 | | | | | | | | | | | | |
| Fluvial Estuarine | 1.08 (0.77) 2.21 (0.52) | 28.56 (0.24) 28.74 (0.09) | 3.65 (1.46) 11.19 (1.26) | 35.3 (1.46) 35.72 (0.07) | 29.59 (2.56) 33.4 (0.41) | 7.9 (0.09) | 7.73 (0.02) | 8.1 (0.09) 8.19 (*) | 8.07 (0.04) (8.15 (*) < |).18 (0.23) ≤ 0.01 (*) | 0.21 (0.22) < 0.01 (*) | 3.05 (0.26) 2 3.11 (*) 2 | 2.78 (0.03) 2.82 (*) |
| Oceanic | 7.35 (2.9) | 28.77 (0.3) | 30.49 (1.17) | 33.02 (4.1) | 32.13 (3.24) | 8.4 (0.66) | 8.15 (0.55) | 8.18 (0.04) | 8.15 (0) (0 |).45 (0.29) | < 0.01 (*) | 2.84 (*) | < 0.01 (*) |
| Dry season | | | | | | | | | | | | | |
| Fluvial | 1.8 (0.77) | 27.95 (0.07) | 6.51 (1.5) | 24.31 (8.58) | 22.69 (7.5) | 6.56 (0.43) | 4.74 (1.18) | 8.05 (0.08) | 7.83 (0.32) (| 0.04 (0.07) | 0.04~(0.06) | 0.23 (0.37) (| .07 (0.03) |
| Estuarine | 6.94 (2.46) | 27.75 (0.03) | 17.39 (2.74) | 26.35 (1.25) | 32.79 (0.87) | 6.69 (0.6) | 5.29 (0.53) | 8.11 (0.03) | 8.06 (0.02) < | < 0.01 (*) | < 0.01 (*) | 0.09 (0.03) (| 0.13 (0.09) |
| Oceanic | 8.56 (2.72) | 27.69 (0.02) | 29.43 (3.66) | 31.97 (1.25) | 32.95 (1.72) | 6.93 (1.54) | 5.41 (0.6) | 8.09 (0.03) | 8.07 (0.02) < | < 0.01 (*) | < 0.01 (*) | 0.07 (0.05) (| (*) 90.(|
| | | | | | | | | | | | | | |
| | | | | | | | | | | | | | |
| | | | | | | | | | | | | | |
| Table 2 The letters values und | Average valu A and B corr ler detection 1 | tes of biologica espond to meas imit | al variables for <i>es</i> surements above | ach variable pe and below the | er season and z halocline wat | sone. Chlorophy ter respectively. | /ll a (Chl-a), abunc Values inside pare | dance (Abd), a enthesis repres | nd genus of dia ent standard de | atom, dinoflagells viation of the ave | ates (Dino), ar rage, * values | nd cyanobacter are for sites wi | ia (Cyano) tith $n = 1$ or |
| | Abd Cy: A (cell/I | ano Al | bd Diatom (cell/L) | Abd Dir A (cell/I | no Ab L) B(| od Cyano (cell/L) | Abd Diatom B (cell/L) | Abd Dino B (cell/L) | Chl A (µg/L) | Chl B (µg/L) | Cyano richness | Diatoms richness | Dino richness |
| Rainy seas | uo | | | | | | | | | | | | |
| Fluvial | 666.5 (6 | (08.5) 43 | 360.2 (3989.6) | 333.2 (4 | 171.4) 16 | 6.8 (333.5) | 333.5 (385.09) | (0) (0) | 3.14 (1.44) | 0.94 (1.17) | 2 (1.7) | 19.3 (2.5) | 2.3 (1.5) |
| Estuarine | (*) 0 (*) | 99 | (*) (*) | (*) 0 | 0 (| (*) | 11111 (*) | (0) (0) | 1.87 (*) | 0.27 (*) | 2 (*) | 25 (*) | (*) 6 |
| Oceanic | 2333.5 (| (471.6) 38 | 388.5 (314.7) | (0) 0 | 0 (| (*) | 0 (0) | (0) (0) | 0.28 (0.06) | 0.32(0.11) | 1.5 (0.7) | 26.5 (6.4) | 4.5 (0.7) |
| Dry seasor | ſ | | | | | | | | | | | | |
| Fluvial | 1333.2 (| (1515.3) 11 | (,166.5 (14,221.8 | 8) 4250 (2) | 885.2) 0 (| (0) | 999.7 (666.5) | (0) (0) | 1.8 (0.68) | 0.24(0.13) | 5.6 (2.5) | 14.7 (2.5) | 4.7 (0.6) |
| Estuarine | 333.4 (5 | (77.6) 34 | 122.2 (3263.4) | 933.4 (1 | 0 (2.090.5) 0 (| (0) | 583 (500) | (0) (0) | 0.26 (0.30) | 0.02(0.04) | 1.8(0.4) | 18.4 (3.7) | 4(1) |
| Oceanic | 111 (192 | 2.3) 38 | 313.3 (2864.7) | 222 (38- | 4.5) 0 (| (0) | 999.7 (1452.66) | 0 (0) | 0.03 (0.04) | 0.04(0.04) | 2 (1) | 36.7 (7.5) | 7 (1) |

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Fig. 3 Vertical profile of estuary density. a Rainy season. b Dry season

during the dry season, and a maximum of 8.19 m in the estuarine zone above the halocline during the rainy season. The pH did not show significant variations between the zones or seasons in the measurements taken above the halocline (p =0.13, p = 0,17, n = 17, AN), but it was more alkaline during the rainy season compared with the dry season in measurements below the halocline (p = 0.02, n = 17, KW), with a maximum pH range between seasons of 0.24 in the fluvial zone, with a maximum of 8.07 ± 0.04 during the rainy season in measures below the halocline (Table 1).

Dissolved oxygen measurements above the halocline showed higher values during the rainy season, with a maximum of 8.4 ± 0.47 mg/L compared with the concentration obtained during the dry season of 6.56 mg/L in the fluvial zone (p < 0.001, n = 26, KW). Measurements of dissolved oxygen below the halocline showed the same seasonal pattern as those done above the halocline (p < 0.001, n = 26, KW), with a maximum in the oceanic zone of 8.15 ± 0.55 mg/L in the rainy season and a minimum of 4.74 ± 1.18 mg/L during the dry season in the fluvial zone. Oxygen values were similar



between the zones, showing a range during the rainy season of 0.99 mg/L above and 1.09 mg/L below the halocline, and 0.37 mg/L above and 0.67 mg/L below the halocline during dry season (p = 0.133, above and p = 0.51 below halocline, n = 26, KW) values for each zone are shown in Table 1.

Nitrate exhibited a higher concentration in the rainy season, with a maximum concentration in the oceanic zone of $0.45 \pm 0.29 \text{ mg/L}$ above the halocline, and in the fluvial zone, a minimum value in the fluvial zone of 0.18 mg/L compared with the dry season, with detectable levels only in the fluvial zone of $0.04 \pm 0.07 \text{ mg/L}$ (p < 0.001, n = 27 KW). Nitrate concentration below the halocline also exhibited a minimum in the dry season. Nitrate was only detectable in the fluvial zone, with a value of 0.04 ± 0.06 , compared with the rainy season, when the nitrate concentration reached $0.21 \pm 0.22 \text{ mg/L}$ on average, with a high deviation due to the higher values present at site 5 of 1.12 mg/L. Measurements of phosphate concentration above the halocline showed higher values during the rainy season, with an estimated maximum of $3.11 \pm 0.26 \text{ mg/L}$ at the estuarine zone compared with the dry season,



Fig. 4 Vertical stratification (or Brunt-Väisälä frequency) in the Gulf of Urabá, Colombian Caribbean Sea. High values indicate strong stratification. a Rainy season. b Dry season

Table 3 One-way ANOVA to check the differences in abundance of some phytoplankton groups between the seasons. A and B refer to values measured above and below the halocline water respectively, and R and D to rainy and dry season. p values < 0.05 indicate significant differences between variables. Ho = the distribution of variable is the same across the seasons

| Abundance (In transformed) | | Sum of squares | Df | Mean square | F | p value |
|------------------------------|-------------------|----------------|----|-------------|--------|---------|
| Diatoms A | Between groups | 24.619 | 2 | 12.309 | 15.387 | 0.000 |
| | Within groups | 16.000 | 20 | 0.800 | | |
| | Total | 40.618 | 22 | | | |
| Diatoms B | Between Groups | 0.484 | 9 | 0.242 | 16.656 | 0.000 |
| | Within groups | 0.290 | 2 | 0.015 | | |
| | Total | 0.774 | 20 | | | |
| Dinoflagellates A | Between groups | 7.430 | 2 | 3.715 | 2.915 | 0.093 |
| | Within groups | 15.294 | 12 | 1.274 | | |
| | Total | 22.724 | 14 | | | |
| Cyanobacteria A | Between groups | 36.356 | 2 | 18.178 | 16.009 | 0.000 |
| | Within groups | 15.896 | 14 | 1.135 | | |
| | Total | 52.252 | 16 | | | |
| Genus richness | | Sum of squares | Df | Mean square | F | p value |
| Dinoflagellates | Between groups | 39.138 | 2 | 19.569 | 8.861 | 0.002 |
| | Within groups | 44.167 | 20 | 2.208 | | |
| | Total | 83.304 | 22 | | | |
| Kruskal-Wallis for non-paran | netric variables | | | | | |
| Variable | | p value | | | | |
| Ln abundance of dinoflagella | tes B | 0.552 | | | | |
| Ln abundance of cyanobacter | ria B | 0.319 | | | | |
| Diatom richness | | 0.002 | | | | |
| Cyanobacteria richness | | 0.085 | | | | |

Table 4 One-way ANOVA to check the differences in abundance of some phytoplankton groups between zones. A and B refer to values measured above and below the halocline water respectively, F, E, and O to fluvial, estuarine, and oceanic zones. p values < 0.05 indicate significant differences between variables. Ho = the distribution of variable is the same across the zones

| Abundance (In transformed) | | Sum of squares | Df | Mean square | F | p value |
|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------|----------------|----|-------------|-------|---------|
| Diatom A | Between groups | 1.018 | 2 | 0.509 | 0.257 | 0.776 |
| | Within groups | 39.600 | 20 | 1.980 | | |
| | Total | 40.618 | 22 | | | |
| Diatom B | Between groups | 0.023 | 2 | 0.012 | 0.311 | 0.737 |
| | Within groups | 0.751 | 20 | 0.038 | | |
| | Total | 0.774 | 22 | | | |
| Dino A | Between groups | 2.555 | 2 | 1.277 | 0.760 | 0.489 |
| | Within groups | 20.169 | 12 | 1.681 | | |
| | Total | 22.724 | 14 | | | |
| Genus richness | | Sum of squares | Df | Mean square | F | p value |
| Diatom | Between groups | 1188.487 | 2 | 594.244 | 9.570 | 0.001 |
| | Within groups | 1241.948 | 20 | 62.097 | | |
| | Total | 2430.435 | 22 | | | |
| Dinoflagellates | Between groups | 29.704 | 2 | 14.852 | 5.542 | 0.012 |
| | Within groups | 53.600 | 20 | 2.680 | | |
| Diatom B Dino A Genus richness Diatom Dinoflagellates Kruskal-Wallis for non-paran Variable Abundance of dinoflagellate I Abundance of cyanobacteria Abundance of cyanobacteria | Total | 83.304 | 22 | | | |
| Kruskal-Wallis for non-parame | etric variables | | | | | |
| Variable | | p value | | | | |
| Abundance of dinoflagellate B | 3 | 0.522 | | | | |
| Abundance of cyanobacteria A | A | 0.178 | | | | |
| Abundance of cyanobacteria E | 3 | 0.552 | | | | |
| Diatom richness | | 0.002 | | | | |
| Cyanobacteria richness | | 0.993 | | | | |



Fig. 5 Salinity (psu) values during a rainy season and b dry season. Temperature (°C) during c rainy season and d dry season

during which the maximum concentration was 0.23 mg/L, also in the fluvial zone (p < 0.001, n = 22, KW). The zones exhibited a similar phosphate concentration of 2.8 ± 0.02 mg/L and 0.08 ± 0.03 mg/L during the rainy and the dry seasons, respectively, in measurements done below the halocline (Table 1). The highest concentration of opal was 43.0 mg/L in the fluvial zone during the rainy season, and the lowest in the same season in the estuarine zone, with a value below the detection limit of the method (10.7 mg/L) at measurements above the halocline (Table 1).

Chlorophyll *a* concentrations were higher during rainy season, with a maximum of $3.14 \pm 1.44 \ \mu g/L$ in the fluvial zone, compared with the dry season, with a minimum of $0.04 \pm 0.04 \ \mu g/L$ in the oceanic zone (p < 0.001, n = 53, KW). It was equally possible to observe that chlorophyll *a* had higher values in the fluvial zone, with differences between the concentrations of $1.27 \ \mu g/L$ and $2.86 \ \mu g/L$ during the rainy season, and $0.22 \ \mu g/L$ and $0.20 \ \mu g/L$ during the dry season at above the halocline for measurements of the estuarine and oceanic zones, respectively (p = 0.03, n = 53, KW). During the first cruise, a bloom was found in the fluvial zone, showing a rise in the chlorophyll *a* concentration that reached 4.1 $\mu g/L$.

Phytoplankton Community Structure and Spatial Toxin Distribution

Phytoplankton abundance in the Gulf of Urabá showed differences between seasons, with diatoms as the most abundant genus, followed by dinoflagellates, and cyanobacteria (Figs. 6, 7, 8, 9, and 10). Diatoms exhibited higher abundance values during the dry season of 11,166 cell/L, 3422 cell/L, and 3813 cell/L in the fluvial, estuarine, and oceanic zones, respectively, compared with the values of 4360 cell/L, 667 cell/L, and 3889 ± 105 cell/L for the same zones during the rainy season (p < 0.001, n = 52, AN) (Table 2, Fig. 8) with Coscinodiscus, Chaetoceros, Nitzschia, and Pseudo-nitzschia as dominant groups. Dinoflagellate maximum abundance occurred during the dry season with a value of 4250 ± 745 cell/L. Nonetheless, their abundance was similar between the zones or seasons with a range between 0 and 333 ± 471.4 cell/L during rainy season, and 222 ± 384.52 to 4250 ± 2885.24 cell/L (p = 0.093, n = 52, AN) during the dry season (Fig. 9) and it was not detectable in samples below the halocline. Dominant genera of dinoflagellates were Tripos, Dinophysis, and Protoperidinium. Cyanobacteria had high values above the halocline during the rainy season with a maximum of 2334 ± 472 cell/L during the rainy season at the oceanic zone in general and a maximum of $102,444 \pm 1025$ cell/L in the fluvial zone during a bloom found in the first cruise (Table 2, Fig. 10). Dominant genus of cyanobacteria included Dolichospermum and Oscillatoria.

Overall, a total of 39 orders of phytoplankton were found in this study (Table 3, Figs. 5 and 6). Diatoms were the most genus-rich group, with a maximum number of genera of $37 \pm$ 8 in the oceanic zone during the dry season, but with more genera during the rainy season in the fluvial and estuarine zones, with 19 ± 2 and 25 (n = 1) (Table 3), differentiating it from the dry season, with genus richness values of 15 ± 2 and 18 ± 3 , respectively (p = 0.001, n = 78, AN). Dinoflagellates exhibited a richness of 3 ± 2 genera during rainy season in the fluvial zone, 6 ± 1 genus in the estuarine zone, and 5 ± 1 genus in the oceanic zone. These values were surpassed during the dry season, with values of 5 ± 1 , 4 ± 1 , and 7 ± 1 genus in the fluvial, estuarine, and oceanic zones (p = 0.001, n = 78,



Fig. 6 Optical microscopy photos of some genera present in the Gulf of Urabá. a *Gyrosigma*, b *Thalassionema*, c *Proboscia*, d *Dithylum*, e *Asterolampra*, f *Leptocylindrus*, g *Biddulphia*, h *Coscinodiscus*, i

Rhabdonema, j Odontella, k Navicula, l Oscillatoria, m Triceratium, n Chaetoceros, o Tripos, p Ornithocercus

AN) respectively. Cyanobacteria exhibited a maximum of 6 ± 3 genera during the dry season in the fluvial zone, but in the other zones, regardless of the season, they showed a richness of 2 ± 1 genus (p > 0.05) (Tables 2 and 5).

The potential biotoxin producer genera found in this work were *Pseudo-nitzschia*, *Prorocentrum*, *Dinophysis*, and *Dolichospermum* (Fig. 7). During the first cruise, we found a *Dolichospermum* (Fig. 10c) bloom at all fluvial

and estuarine sites, with abundances higher than 100,000 cells/L, representing more than 97% of the total population of phytoplankton and affecting its richness (Figs. 8c and 9c). Our analyses of the conditions during the rainy season did not consider this phenomenon, as these values were out of range. During the second cruise, we found a high presence of the species *Pseudo-nitzschia pseudodelicatissima* around all the stations, representing

Fig. 7 Photos of the toxic genus and species found in the Gulf of Urabá. a Dinophysis acuminata, b D. caudata, c Prorocentrum sp., d Dolichospermum sp., e Pseudo-nitzschia pseudodelicatissima





Fig. 8 Abundance of diatoms during a rainy season, b dry season, c rainy season during a *Dolichospermum* sp. bloom in fluvial zone in the sites 3, 4, and 5

44% of the average abundance of diatoms found in the fluvial zone during rainy season.

The high quantity of *Pseudo-nitzschia* motivated the performance of domoic acid (DA) biotoxin measurements. DA concentration during the rainy season exhibited values between non-detectable levels and 1589.7 pg/L, and during the dry season between 25.5 and 164.9 pg/L (p = 0.017, KW). Maximum DA concentration of 1580.7 pg/L was reached at site 3 (fluvial zone) (Fig. 11a). The lowest values for concentration of DA were found during the dry season (Fig. 11b) in agreement with the low *Pseudo-nitzschia* abundance values found (< 100 cell/L). By contrast, there were no substantial changes in the DA concentration in the Gulf of Urabá (p = 0.212, KW).

Correlation Analyses

Spearman tests showed correlations of up to 70% with nitrate measurements above the halocline ($\rho = -0.829$, p = 0.042) in terms of diatom abundance above the halocline. Diatom



Fig. 9 Abundance of dinoflagellates during **a** rainy season, **b** dry season, **c** rainy season during a *Dolichospermum* sp. bloom in fluvial zone in the sites 3, 4, and 5

richness was correlated with nitrate levels above the halocline ($\rho = 0.841$, p = 0.036). Abundance of dinoflagellates above the halocline exhibited a correlation with salinity below the halocline ($\rho = -0.711$, p = 0.021). Domoic acid concentrations exhibited significant correlations with pH above the halocline ($\rho = 0.011$), but it was low ($\rho = 0.635$). Finally, chlorophyll *a* levels were correlated with salinity above the halocline ($\rho = -0.945$, p = 0.000), average salinity ($\rho = -0.723$, p = 0.003), transparency ($\rho = -0.925$, p = 0.000), and nitrate below the halocline ($\rho = 0.900$, p = 0.037). Other abundances

and richness did not show significant correlations with the variables.

Discussion

The Gulf of Urabá as an Estuarine Ecosystem

Coastal systems are important zones in which water and land interact, connecting continents with the ocean, due to the



Fig. 10 Abundance of cyanobacteria during a rainy season, b dry season, c rainy season during a *Dolichospermum* sp. bloom in fluvial zone in the sites 3, 4, and 5

physical, chemical, biological, and geological processes around them (Bauer et al. 2013; Dalrymple et al. 1992). Estuaries are major receptors of the terrigenous material and nutrients from rivers, making them an important environment for understanding global carbon flux and oceanic primary production (Mallin et al. 1993; Statham 2012; Twilley et al. 1992). Due to their nature as nutrient receptors and the spatiotemporal physiochemical characteristics of the water column, estuaries exhibit higher levels of primary production than other ecosystems (Costa et al. 2009; Lara-Rodríguez et al. 2015; Lucas et al. 1999a; Montoya et al. 2017; Pednekar et al. 2018).

In the Gulf of Urabá, as an estuarine ecosystem (Figs. 1 and 12), physicochemical parameters varied between the rainy and dry seasons. There was stratification during both seasons, as reported in other studies (Montoya and Toro 2006; Montoya 2010; Montoya et al. 2017), with a halocline closer to the surface during the rainy season. These results are similar to those obtained by Chevillot et al. (1993), in which changes in salinity were observed in response to seasonal wind speed and

Table 5Main phytoplankton groups and species in the Urabá Gulf, the letters F, E, and O refer to fluvial, estuarine, and oceanic zones respectively. Xmeans the presence of the genera/specie in the zone. Data of early rainy season refers to the richness found during a *Dolichospermum* bloom

| Таха | Rainy | season | | | | | Dry sea | ason | |
|------------------------------|-------|--------|---|------|---|---|---------|------|---|
| | Early | | | Late | | | | | |
| | F | Е | 0 | F | Е | 0 | F | Е | 0 |
| Phylum: Bacillariophyta | X | х | X | X | X | X | X | X | X |
| Order: Asterolamprales | | | | Х | Х | Х | | Х | Х |
| Asterolampra sp. | | | | Х | Х | Х | | Х | Х |
| Asteromphalus sp. | | | | Х | Х | Х | | Х | Х |
| Order: Aulacoseirales | | | | Х | Х | | Х | | |
| Aulacoseira cf. sp. | | | | Х | Х | | Х | | |
| Order: Bacillariales | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| <i>Bacillaria</i> sp. | | | | | | | Х | | Х |
| Cylindrotheca closterium | Х | Х | | Х | Х | Х | Х | Х | Х |
| Nitzschia sp. | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Pseudo-nitzschia sp. | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Order: Biddulphiales | | | | | | Х | Х | Х | Х |
| Biddulphia spp. | | | | | | Х | | Х | Х |
| Eucampia sp. | | | | | | | Х | Х | Х |
| Lamprisccus shadboltianum | | | | | | | Х | Х | Х |
| Order: Chaetocerotales | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Bacteriastrum spp. | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Chaetoceros affinis | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Chaetoceros borgei | Х | | | Х | | Х | Х | Х | Х |
| Chaetoceros coarctatus | | | | Х | | Х | Х | Х | Х |
| Chaetoceros curvisetus | | | | Х | Х | Х | Х | Х | Х |
| Chaetoceros danicus | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Chaetoceros decipiens | Х | | | Х | | | | | |
| Chaetoceros didymus | Х | | | Х | Х | Х | | | |
| Chaetoceros distans | | | | Х | Х | Х | Х | Х | |
| Chaetoceros diversus | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Chaetoceros lorenzianus | Х | | | Х | Х | Х | Х | Х | Х |
| Chaetoceros muelleri | | | | Х | | | | Х | |
| Chaetoceros peruvianus | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Chaetoceros pseudocurvisetus | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Chaetoceros subtilis | | | | Х | Х | Х | Х | Х | Х |
| Leptocylindrus danicus | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Leptocylindrus minimus | | | | Х | | | Х | | |
| Order: Coscinodiscales | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Coscinodiscus spp. | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Order: Cymbellales | | | | | | Х | | Х | Х |
| Gomphonema sp. | | | | | | Х | | Х | Х |
| Order: Eupodiscales | X | | | X | X | X | Х | Х | X |
| Odontella spp. | Х | | | Х | Х | Х | Х | Х | Х |
| Amphitetras cf. sp. | | | | | | Х | | Х | Х |
| Order: Fragilariales | | | | | | X | Х | X | Х |
| Fragilaria sp. | | | | | | | Х | Х | Х |
| Podocystis adriática | | | | | | | | | Х |
| Synedra sp. | | | | | | Х | | | Х |
| Order: Hemiaulales | Х | | | X | X | X | Х | X | Х |
| | - | | | | | | | | |

Table 5 (continued)

| Таха | Rainy | season | | | | | Dry sea | ason | |
|---------------------------|-------|--------|---|------|---|---|---------|------|---|
| | Early | | | Late | | | | | |
| | F | Е | 0 | F | Е | 0 | F | Е | 0 |
| Cerataulina pelágica | | | | Х | | | | | |
| Hemiaulus sp. | Х | | | Х | Х | Х | Х | Х | Х |
| Isthmia enervis | | | | | | | | Х | |
| Order: Licmophoraceae | | | | | | Х | Х | Х | Х |
| Licmophora sp. | | | | | | Х | Х | Х | Х |
| Order: Lithodesmiales | Х | | | Х | Х | Х | Х | Х | Х |
| Ditylum sp. | Х | | | Х | Х | Х | Х | Х | Х |
| Lithodesmium undulatum | | | | | | | Х | Х | Х |
| Order: Lyrellales | | | | | | Х | | | Х |
| Lyrella sp. | | | | | | Х | | | Х |
| Order: Mastogloiales | | | | | | Х | | | Х |
| Achnanthes sp. | | | | | | Х | | | Х |
| Order: Melosirales | | | | | | | Х | Х | Х |
| Melosia spp. | | | | | | | Х | Х | Х |
| Order: Naviculales | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Diploneis sp. | | | | | Х | Х | | Х | Х |
| Gyrosigma sp. | | Х | Х | Х | Х | Х | | | Х |
| Navicula sp. | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Pinnularia sp. | | | | | | | Х | | |
| Pleurosigma sp. | | | | Х | Х | Х | | | Х |
| Order: Plagiogrammales | | | Х | | Х | Х | | | |
| Plagiogramma sp. | | | Х | | Х | Х | | | |
| Order: Rhabdonematales | | | Х | Х | Х | Х | | Х | Х |
| Grammatophora marina | | | | | | Х | | | Х |
| Rhabdonema adriaticum | | | | | | Х | | | Х |
| Asterionellopsis sp. | | | Х | Х | Х | Х | | Х | Х |
| Delphineis sp. | | | | | | Х | | | Х |
| Order: Rhizosoleniales | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Dactyliosolen sp. | | | | | Х | | | | |
| Proboscia sp. | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Rhizosolenia sp. | Х | | | Х | Х | Х | | Х | Х |
| Guinardia fláccida | | | | Х | Х | Х | Х | Х | Х |
| Guinardia striata | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Neocalyptrella robusta | | | | Х | Х | Х | | Х | Х |
| Pseudosolenia sp. | | | | | | | Х | Х | |
| Order: Surirellales | | | | | | Х | Х | Х | Х |
| Entomoneis sp. | | | | | | Х | Х | Х | Х |
| Surirella sp. | | | | | | Х | Х | Х | Х |
| Order: Tabellariales | | | | | | Х | | Х | Х |
| Asterionella formosa | | | | | | Х | | Х | Х |
| Order: Thalassionematales | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Thalassionema spp. | Х | Х | Х | Х | Х | Х | Х | Х | Х |
| Order: Thalassiophysales | | | | | Х | Х | Х | | Х |
| Amphora sp. | | | | | Х | Х | Х | | Х |
| Order: Thalassiosirales | Х | Х | Х | Х | Х | Х | Х | Х | Х |

Table 5 (continued)

| Taxa | Rainy | season | | | | | Dry sea | ason | |
|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|--------|--------|--------|--------|--------|--------|---------|--------|--------|
| | Early | | | Late | | | | | |
| | F | Е | 0 | F | Е | 0 | F | Е | 0 |
| Detonula pumila | | | | Х | Х | Х | | | Х |
| Lauderia annulate | | | | | | Х | | Х | Х |
| Skeletonema sp. | Х | Х | Х | Х | Х | | Х | Х | Х |
| Order: Toxariales | | | | | | | Х | Х | Х |
| Climacosphenia monoligera | | | | | | | Х | Х | Х |
| Order: Triceratiales | | | | Х | | | | | Х |
| Triceratium pentacrinus | | | | Х | | | | | Х |
| Phylum: Cercozoa | Х | Х | Х | Х | | | Х | Х | Х |
| Order: Ebriida | Х | Х | Х | Х | | | Х | Х | Х |
| Hermesinum sp. | Х | Х | Х | Х | | | Х | Х | Х |
| Phylum: Charophyta | | | | Х | | | Х | | |
| Order: Desmidiales | | | | Х | | | Х | | |
| Staurastrum subgen eustaurastrum | | | | Х | | | Х | | |
| Phylum: Chlorophyta | | | | Х | Х | | | Х | |
| Order: Sphaeropleales | | | | Х | | Х | | Х | |
| Desmodesmus spp. | | | | Х | | | | Х | |
| Coelastrum sp. | | | | Х | | Х | | Х | Х |
| Pediastrum simplex | | | | Х | | | | Х | |
| Pediastrum duplex | | | | Х | | | | Х | |
| Order: Synechococcales | | | | X | | | X | X | |
| <i>Cvanothrix</i> cf. sp. | | | | Х | | | Х | Х | |
| Synechocystis cf. sp. | | | | | | | Х | | |
| Phylum: Cyanobacteria | | | | х | х | | X | | |
| Order: Chroococcales | | | | x | x | | x | | |
| Microcystis sp | | | | x | X | | x | | |
| Inhanneshantistia sp | | | | 24 | 24 | | x | x | |
| Order: Nostocales | x | x | x | x | x | x | x | x | x |
| Dolichospermum sp | X X | X X | X X | X X | X | x x | x x | v | 1 |
| Order: Oscillatorialas | Λ | X V | X V | X V | N V | N V | x v | N V | v |
| Planttothrin of sp | | A V | A V | A V | A V | A V | A V | A V | A V |
| Phylippi Muozoo | v | л V | л V | л V | л V | л V | л v | л V | л V |
| Order Dinenhyriolog | A V | A V | A V |
| Diversional and the second sec | Л | А | А | A V | A V | A V | | Λ | |
| Dinophysis acuminate | 37 | v | 37 | X | A V | A V | A V | 37 | X |
| Dinophysis caudata | Х | Х | Х | Х | Х | X | Х | X | X |
| Ornithocercus magnificus | | | | | | Х | | X | X |
| Phalacroma mitra | | | | | | | | X | X |
| Phalacroma rotundatum ct. | | | | | | | | X | X |
| Order: Gonyaucales | Х | | | Х | Х | X | X | Х | X |
| <i>Gambierdiscus</i> sp. | | | | | | | Х | | Х |
| Tripos hircus | | | | Х | Х | Х | Х | Х | Х |
| Tripos furca | | | | Х | Х | | | Х | Х |
| Tripos fusus | Х | | | Х | Х | Х | Х | Х | Х |
| Tripos kofoidii | Х | | | | Х | Х | | Х | Х |
| Tripos mollis | | | | | | Х | Х | | Х |
| Tripos macroceros | | | | Х | Х | Х | Х | Х | Х |

$Table \; 5 \; \; (continued)$

| Taxa | Rainy | season | | | | | Dry season | | |
|-----------------------|-------|--------|---|------|---|---|------------|---|---|
| | Early | | | Late | | | | | |
| | F | Е | 0 | F | Е | 0 | F | Е | 0 |
| Tripos trichoceros | | | | | | х | Х | Х | X |
| Order: Peridiniales | | Х | Х | Х | Х | Х | Х | Х | Х |
| Corythodinium sp. | | | | | | | | Х | |
| Protoperidinium sp. | | Х | Х | Х | Х | Х | Х | Х | Х |
| Order: Porocentrales | | | | Х | Х | Х | | | Х |
| Prorocenctrum sp. | | | | Х | Х | Х | | | Х |
| Phylum: Ochrophyta | | | | Х | Х | Х | | | |
| Order: Dictyochales | | | | Х | Х | Х | Х | | |
| Dyctiocha sp. | | | | Х | Х | Х | Х | | |
| Order: Mischococcales | | | | Х | | | | | |
| Centritractus sp. | | | | Х | | | | | |

its direct effect on circulation. Since winds are stronger in the dry season, the fresh water from the riverine inputs was confined to the interior of the Gulf. Conversely, during the rainy season, winds blow from the south, forming a surface current that allows the river to evacuate the Gulf and increase the influence of the river to the north (Escobar 2011), extending the estuarine zone.

In both seasons, riverine water is drained through the eastern side of the Gulf (Chevillot et al. 1993; Escobar 2011). This explains why it is possible to find water with a surface salinity higher than 28 psu on the northwest side of the Gulf (sites 13, 14, and 15). Therefore, Atrato River and wind forcing can lead to the variability of circulation observed in the estuary. As expected, density and salinity values in the oceanic zone are non-estuarine, but they were lower than the values found in the surface waters of the Caribbean Sea, especially during the rainy season. This can be explained by the high precipitation levels on the western side of the gulf (Figs. 2 and 3). For example, the Acandí meteorological station is close to the oceanic zone, and its historical data show an average precipitation of 278 mm per month (Fig. 2). In addition, density and Brunt-Väisälä frequency analyses show that the estuary is



Fig. 11 Domoic acid levels in a rainy season and b dry season



Fig. 12 Conceptual diagram the physical-chemical-biological factors that modulate the communities in estuary

stratified due to large volumes of fresh water (from the Atrato River), where there are horizontal layers of low surface density with high density in the deep layers (Figs. 3 and 4).

Phosphate and nitrate are also related to the river and to precipitation: the concentrations of phosphate presented a tendency to increase towards the estuarine zone during the rainy season; this is similar to various REDCAM reports (INVEMAR 2017a, b), where it was shown that the Gulf has the highest concentrations of nutrients in the rainy season.

Results from the present study also showed an inverse correlation between the oceanic influence (or salinity) and nutrient availability. For example, during the dry season, the phosphate and opal concentrations were higher in the fluvial zone, probably because of the decrease of the riverine inputs. Nitrate at the surface during the rainy season was higher in the oceanic zone, perhaps because the primary production (linked to chlorophyll a levels) is lower in this zone than in other zones. The differences between phosphate, opal, and nitrate concentrations lead us to believe that nitrate is the limiting nutrient for phytoplankton communities. Nitrate levels also showed an inverse correlation with the abundance of diatoms and a positive correlation with the richness of phytoplankton. Also, an oxygen concentration of 4.74 ± 1.18 mg/L was found in the fluvial zone during the dry season under halocline; we hypothesize that oxygen will show low concentration values in a great part of the estuary, and that eventually the concentration will decrease to hypoxic values (Table 1).

Phytoplankton Communities

The results of this study show that changes in the structure of the phytoplankton communities are modulated over both time and space in this estuary. However, the richness of cyanobacteria only exhibits differences over space. High phytoplankton abundance and richness were found during the dry season at the surface (e.g., Ciénaga Grande de Santa Marta) (De La Hoz Aristizábal 1996). With diatoms, this difference in the abundances of phytoplankton was related to riverine inputs (sites located at the mouths of the Atrato River), but there was no seasonal difference between the other zones. These phenomena are related to the low transparency value: transparency affects the availability of light radiation required for photosynthesis, but riverine inputs can also promote the phytoplankton communities (Cloern et al. 2014; Glibert 2016; Reynolds 2006). Of course, light and temperature play a key role in community development. Nevertheless, other authors have suggested that adaptations to turbidity, currents, and the mixing between superficial water layers strongly influence phytoplankton communities (Glibert 2016; Reynolds 2006). Therefore, runoff can be considered a crucial factor in the area of this study, since agricultural activities nearby utilize fertilizers rich in nitrogen, which alters the structure of the phytoplankton communities.

Although the seasons exert strong effects on the abundance, differences between zones in the different seasons were not observed. However, richness exhibited differences among the zones, with a greater number of genera during the rainy season in sites less influenced by the river (oceanic zone). The inverse correlation between abundance and richness of diatoms supports the hypothesis that the diversity and the rate of renovation of phytoplankton communities are inversely correlated (Glibert 2016; Margalef 1972, 1978).

For the fact that river mouths are sites with lower transparency, *Chaetocerales*, *Coscinodiscales*, *Thalassiosirales* orders, and some cyanobacteria can better adapt to river mouth conditions. Dinoflagellates have less capacity to adapt to high transparency conditions (Cullen and MacIntyre 1998; Glibert 2016; Margalef 1972, 1978; Reynolds 2006). This ecological phenomenon is evidenced in the composition of the community during the various seasons: in the rainy season, dinoflagellates were present in the estuarine and oceanic zones, with lower abundances in river discharge areas, possibly due to low transparency in the fluvial zone, linked with high turbidity in this area (Blanco-Libreros 2009). However, during the dry season, dinoflagellates exhibited a higher abundance in the fluvial zone, perhaps because of a water discharge reduction linked to the decrease in precipitation.

Concentration of chlorophyll *a* found in both seasons in this study seems to respond to the changes in the abundance of the phytoplankton organisms in these estuary variations. Although the rainy season exhibited higher values of chlorophyll *a* in the estuarine and fluvial zones (Table 2), in the oceanic zone, there were no seasonal differences compared with the dry season (p = 0.698, KW, Table 2), in opposition with the phytoplankton abundance that was high during dry season. This could be a consequence of adaptations that phytoplankton species develop in order to improve their capacity to use the light when the turbidity increases. Adaptations to low light availability can include producing more chlorophyll. The high concentrations of chlorophyll *a* during the rainy season could also be a consequence of the blooms found during the sample collection (Reynolds 2006).

By contrast, dry season chlorophyll a concentration values could be related to the ones found during rainy season because of the increased light availability during this season. This condition can change the requirements of chlorophyll a of the diatoms and result in a decrease of the pigment inside the cells, so the abundance could be higher during the season, but the chlorophyll a content per cell was lower.

It is important to point out that during the rainy season, the levels of nitrate and phosphate were higher than in the dry season (opal showed high values during all the year, so it is not a limiting resource). However, Lucas et al. (1999a, b) argued that local conditions for phytoplankton growth rates in an estuary may vary significantly over space and time, but also hydrodynamic forcing, such as wind or freshwater flow (in this case the Atrato River), could be important (Jennerjahn 2012; Lucas et al. 1999a, b). During the first and second cruise, we found *Dolichospermum* and high concentrations of *Pseudo-nitzschia pseudodelicatissima*, respectively. This may be related to agricultural runoff prevailing in the study

area (Blanco-Libreros 2009). Some studies have demonstrated that these coastal ecosystems are vulnerable to exhibiting harmful blooms due to agricultural runoff (Gruber and Sarmiento 1997; Beman et al. 2005). By contrast, during the dry season, the community contained a higher quantity of dinoflagellates (ca. 20%). These changes could be related to the increase of transparency during the dry season.

The higher abundance of toxic species during the rainy season, specifically the DA producer Pseudo-nitzschia pseudodelicatissima, could be a possible warning sign, indicating that the toxin could build up in the estuary. High levels of DA can produce intoxications that can affect not only all marine trophic levels but also humans due to its non-reversible amnesic effect (McCabe et al. 2016; Scholin et al. 2000; Trainer et al. 2007, 2012). In general, major levels of DA were measured in bivalves, with values higher than 610 mg DA/g (Trainer et al. 2012). Also, total DA concentrations found in other studies showed concentrations between 2 and 220 pg/ mL, with a toxicity risk at values higher than 10,000 pg/mL (Silver et al. 2010; Trainer et al. 2012). These values are higher than the maximum of 1580 pg/mL found in the gulf. Our measurements showed the presence of the biotoxin around all the zones in the Gulf during all seasons. However, sites at the front of the river exhibited higher concentrations, because conditions of high levels of nitrogen and low levels of salinity increase the population of Pseudonitzschia (Mckibben et al. 2017; Pednekar et al. 2018; Trainer et al. 2009). Moreover, some studies have shown the influence of factors such as salinity on the growth of Pseudo*nitzschia*, taking place over a salinity range of 1 to > 35 psu (Rachman and Thoha 2015; Thessen et al. 2005).

Further studies are required to determine if the domoic acid production per cell is also influenced by riverine inputs in the Gulf of Urabá, but it is important to note that the rainy season exhibits higher temperatures than the dry season, and this can modulate the production of DA by increasing it (Mckibben et al. 2017), also to take more replications of the samples per site will be very helpful to use tests such Permanova, for a better understanding of each zone characteristics and changes that can be related with the domoic acid production and phytoplankton community dynamics.

There are reports of the presence of *Pseudo-nitzschia* in the Caribbean Sea (Franco-Herrera et al. 2006; Mancera-Pineda et al. 2009) and in the Colombian Pacific basin (Chavarría and Sogamoso 2010). However, there is no clear information about the abundance of *Pseudo-nitzschia pseudodelicatissima* and domoic acid production in the Colombian Caribbean Sea. In order to confirm the occurrence of blooms of *Pseudo-nitzschia pseudodelicatissima* in the zone, a review of water samples from 2016 in the fluvial zone preserved by Florez-Leiva (unpublished data, 2019) showed abundances of diatoms higher than 10⁸ cells/L during the rainy season of that year. The evidence found in 2016 samples and the high

abundance of *Pseudo-nitzschia* during rainy season suggest a potential risk of blooms. In conclusion, the estuary exhibits changes in the phytoplankton community structure related to seasonal changes and nitrate concentration, especially during the rainy season, the phytoplankton communities will show a higher abundance and environment conditions that may thrive into blooms. In addition, we found potentially toxic algae that also are producing biotoxins such domoic acid, which can impact public health. Therefore, it is important to continue monitoring the estuary to prevent and predict the local effects associated with toxic algae.

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