



## Short Communication

# Occurrence and distribution of *Scrippsiella* cf. *acuminata* (Dinophyta, Thoracospharaceae) in a tropical estuarine gradient

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### Abstract

*Scrippsiella acuminata* is a species complex that can cause anoxic conditions in the water column during blooms. This study aimed to analyze the occurrence and distribution of *Scrippsiella* cf. *acuminata* along the estuarine gradient of Paraguaçu River, from bimonthly sampling (March 2018 to March 2019) performed at 12 sampling points. Environmental variables were measured *in situ*, and water samples were collected for analysis of dissolved inorganic nutrients and phytoplankton cell density. At each point, samples were collected for analysis of species composition and preserved with formaldehyde at a final concentration of 4%. *S.* cf. *acuminata* occurred throughout the estuarine gradient (salinity 0.1–38.9), except in July, coinciding with the lowest concentrations of dissolved inorganic nutrients. Cell density varied between 20 cel L<sup>-1</sup> (January 2019) and 1.8 × 10<sup>6</sup> cell L<sup>-1</sup> (March 2018); in the latter, the species bloomed under conditions of low salinity (5.7) and the highest dissolved inorganic nutrient concentration recorded in the study. Cell density (1.7 × 10<sup>5</sup> L<sup>-1</sup>) increased in November 2018. Neither event had any detrimental effects on the estuarine system. This study allowed us to better understand the distribution patterns of *S.* cf. *acuminata* considering the environmental characteristics that can favor its development.

**Key words:** algal bloom, dinoflagellate, potentially harmful species.

### Resumo

*Scrippsiella acuminata* é um complexo de espécies que pode causar condições anóxicas na coluna d'água durante florações. Este estudo teve como objetivo analisar a ocorrência e distribuição de *Scrippsiella* cf. *acuminata* ao longo do gradiente estuarino do rio Paraguaçu, a partir de coletas bimestrais (março de 2018 a março de 2019) realizadas em 12 pontos amostrais. Variáveis ambientais foram medidas *in situ*, e amostras de água foram coletadas para análise de nutrientes inorgânicos dissolvidos e densidade de células fitoplanctônicas. Em cada ponto, as amostras foram coletadas para análise da composição das espécies e preservadas com formaldeído com concentração final de 4%. *S.* cf. *acuminata* ocorreu em todo o gradiente estuarino (salinidade 0,1–38,9), exceto em julho, coincidindo com as menores concentrações de nutrientes inorgânicos dissolvidos. A densidade celular variou entre 20 células L<sup>-1</sup> (janeiro de 2019) e 1,8 × 10<sup>6</sup> células L<sup>-1</sup> (março de 2018); neste último, ocorreu uma floração em condições de baixa salinidade (5,7) e a maior concentração de nutrientes inorgânicos dissolvidos registrada para o estudo. A densidade celular (1,7 × 10<sup>5</sup> L<sup>-1</sup>) aumentou novamente em novembro de 2018. Nenhum dos eventos teve efeitos prejudiciais no sistema estuarino. Este estudo permitiu-nos compreender melhor os padrões de distribuição de *S.* cf. *acuminata* considerando as características ambientais que podem favorecer seu desenvolvimento.

**Palavras-chave:** florações algais, dinoflagelados, espécies potencialmente nocivas.

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*Scrippsiella* is a genus of thecate dinoflagellates proposed by Balech (1959) based on samples from San Diego, California. The type species is *S. sweeneyae* Balech ex A. R. Loeblich III 1965. Members of the genus are characterized by having cells with 24–32.5 mm of total length and 19–24 mm of total width, conical epitheca, rounded hypotheca, without horns. Moreover, they have a wide, excavated, and descending cingulum, with marked overlapping and displaced two-thirds of its width. The cingulum has six plates, and five equals. The deep sulcus is slightly curved to the right. Plate formula is 4', 3a, 7'', 6c, 5''' 2''''', and 4s; plate 1' is narrow, rhomboid, and has an upward curved base, whereas 2a is usually pentagonal, sometimes hexagonal. Despite its peridinioid pattern, *Scrippsiella* differs from other peridinioid species (*i.e.*, *Pentapharsodinium*, *Peridinium*, and *Protoperidinium*) in having six cingular plates, with two cingular sutures in the dorsal view of the mobile cells (Dale 1977, 1978).

With the advent of molecular biology, Gottschling *et al.* (2005) studied several individuals and DNA sequences of *Scrippsiella sensu lato* (including *Calcigonellum*, *Calciodinellum*, and *Pernambugia*) from four clades, one of which was *Scrippsiella sensu stricto* and mostly comprised the *S. trochoidea* (F.Stein) A.R.Loeblich III species complex. These authors showed that molecular data suggested the existence of cryptic species, but no morphological variation was observed (especially in *Scrippsiella sensu stricto*).

Kretschmann *et al.* (2015) collected, isolated, and cultivated individuals of *Scrippsiella acuminata* (Ehrenberg) Kretschmann, Elbrachter, Zinssmeister, Soehner, Kirsch, Kusber & Gottschling, previously identified as *Peridinium acuminatum* Ehrenberg, from the type locality (Kiel, Germany-Baltic Sea). They also barcoded species of the family Thoracosphaeraceae using rRNA sequences and evaluated the species' morphological aspects using optical and scanning electron microscopy. As a result, these authors showed that the analyzed individuals differed from *Peridinium cinctum* (O.F.Müller) Ehrenberg, but not from from *Scrippsiella trochoidea* (Stein) Loeblich III, thus considering this as a heterotypic synonym and proposing the combination *Scrippsiella acuminata*.

Among the 30 species belonging to the genus (Guiry & Guiry 2020), *S. acuminata* is a cosmopolitan, neritic, and estuarine species (as *S. trochoidea* in Steidinger & Tanger 1997) commonly

found in eutrophic environments or during nutrient input caused by increased precipitation, which carries nutrients from the continent to the aquatic body (Hodgkiss & Yang 2001; Okolodkov & Gárate-Lizárraga 2006; Tas & Yilmaz 2015). This species forms blooms around the world (*e.g.*, Hallegraeff 1992; Al-Azri *et al.* 2015; Hameed & Saburova 2015; Tas & Yilmaz 2015). It is a potentially harmful species that does not produce toxins but can cause the death of fish and marine invertebrates by generating anoxic conditions in the water when reaching high cell concentrations (Hallegraeff 1993; Hold *et al.* 2001).

Morphological studies have shown that *S. acuminata* is widely distributed along the Brazilian coast and is registered in several marine and coastal environments in the states of Pará, Alagoas, Bahia, Pernambuco, Rio Grande do Norte, Espírito Santo, Rio de Janeiro, Santa Catarina, and Rio Grande do Sul [Gottschling *et al.* 2005; Costa 2014; Flora do Brasil 2020 (continuously updated)]. The first record of a bloom of this species in Brazil was in Guanabara Bay (Faria 1914), when fish mortality was reported. Oliveira (1947, 1950) also reported blooms of *S. acuminata* (as *Glenodinium trochoideum*) in Guanabara Bay. Although the effects of these blooms were initially related to the presence of toxins, Odebrecht *et al.* (2002) argued that the reported mortality resulted from the condition of anoxia in the water, which was caused by high cell density ( $10^8$  cell L<sup>-1</sup>).

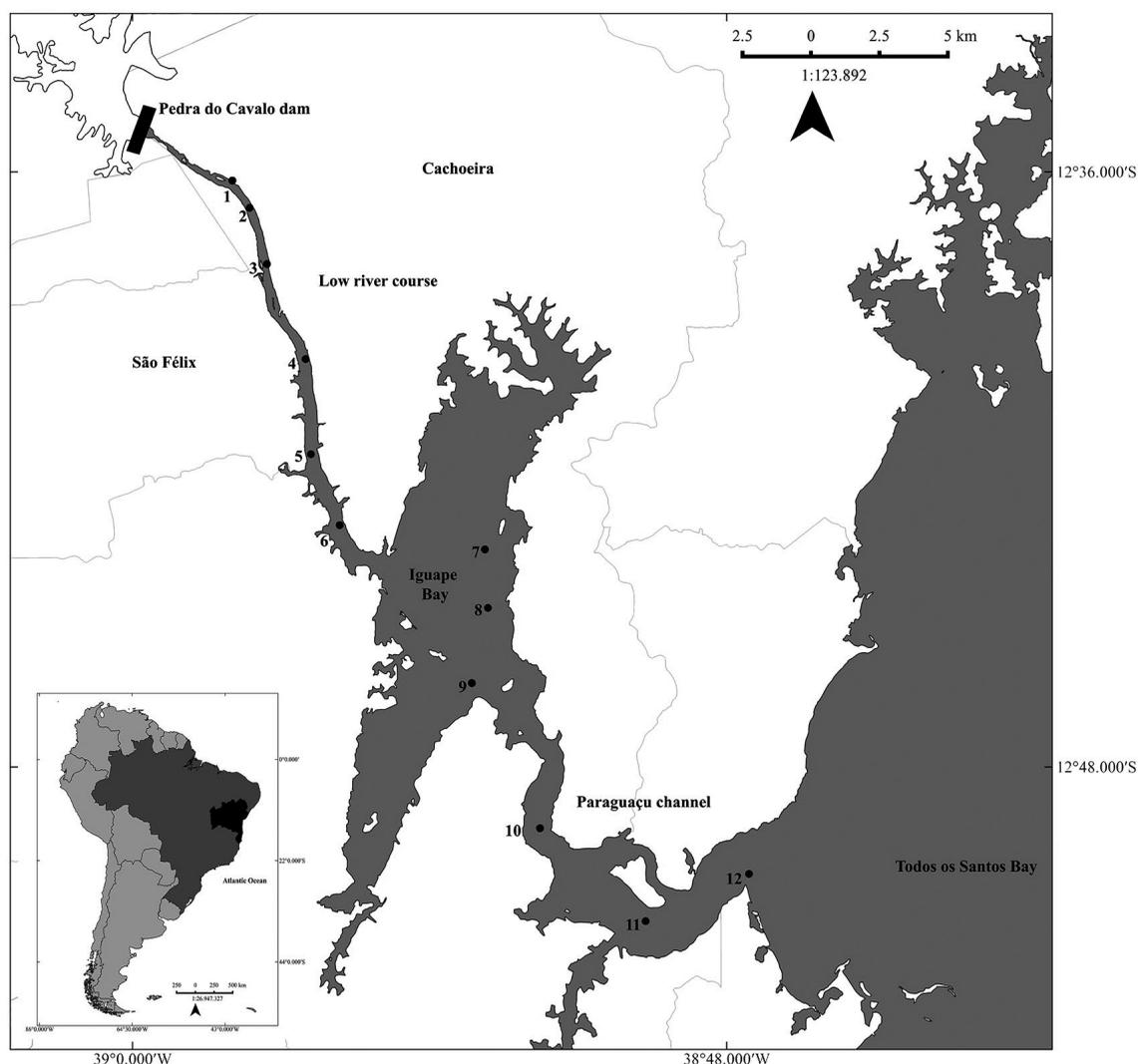
Algal blooms increase the primary biomass in the environment, providing more food to the upper levels of the food web (Smayda 1997). Some species produce cysts, bioactive compounds, toxic metabolites, or growth-inhibiting substances, in addition to causing shading in the water column or depletion of oxygen by forming large cell densities, characterizing the so-called harmful algal blooms (HABs), which negatively affect health, aquaculture, the environment, and recreational activities (Anderson *et al.* 2002; Reguera *et al.* 2011). For example, the harmful bloom of the species *Akashiwo sanguinea* (K. Hirasaka) G. Hansen & Moestrup, registered in 2007 in the Todos os Santos Bay, caused the death of 50 tons of fish (Azevedo 2007).

The Paraguaçu River estuary, one of the main tributaries of Todos os Santos Bay, is a largely unknown regarding the composition of dinoflagellate assemblages. The importance of this knowledge has become more evident since the *Akashiwo sanguinea* bloom. This phenomenon

was associated with the opening of the floodgates of Pedra do Cavalo dam (Reis-Filho *et al.* 2012) as since its installation in 1985, this dam has been altering the hydrological regime of the river (Genz 2006; Genz *et al.* 2008). Moreover, this dam directly or indirectly affects the water's physical and chemical characteristics, which in turn affect biological aspects such as species occurrence and distribution.

Thus, the present study aimed to analyze the occurrence and distribution of *S. cf. acuminata* considering abiotic conditions (*i.e.*, rainfall, water flow, water temperature, salinity, water transparency, and dissolved inorganic nutrients) along the estuarine gradient of Paraguaçu River.

The estuarine system of Paraguaçu River ( $12^{\circ}00'S$  and  $13^{\circ}00'S$ ,  $38^{\circ}30'W$  and  $39^{\circ}30'W$ ) extends from the municipalities of Cachoeira and São Félix to Todos os Santos Bay (TSB) (Fig. 1). It comprises three segments: (1) low river course; (2) Iguape Bay, and (3) Paraguaçu channel; the latter establishes the connection between Iguape Bay and TSB (Mestrinho 1998; Genz *et al.* 2008). The climate is hot and humid, with autumn-winter rains. Annual rainfall in the region varies between 1,200 mm at Pedra do Cavalo dam, in Cachoeira, and 1,600 mm at the outlet of Paraguaçu Channel in TSB. The average annual temperature is  $24^{\circ}C$  ( $21-26^{\circ}C$ ) (Genz 2006).



**Figure 1** – Map of the Paraguaçu River estuary with the sampling stations in the Low river course (points 1-6), Iguape Bay (points 7-9), and Paraguaçu Channel (points 10-12).

Bimonthly collections were carried out between March 2018 and March 2019 at 12 points distributed along the estuary of the Paraguaçu River as follows: (1) points 1–6 in the low river course, in the nearest region upstream from the estuary and Pedra do Cavalo dam; (2) points 7–9 in the Iguape Bay; and (3) points 10–12 in Paraguaçu Channel, downstream of the estuary (Fig. 1).

Water transparency was estimated using a Secchi disk, and temperature and salinity were measured using a multiparameter meter (HANNA hi 9828, São Paulo, Brazil). Rainfall data from the city of Cachoeira, BA were obtained from the Center for Weather Forecasting and Climate Studies (CPTEC/INPE; <[http://proclima.cptec.inpe.br/balanco\\_hidrico/balancohidrico.shtml](http://proclima.cptec.inpe.br/balanco_hidrico/balancohidrico.shtml)>). Water flow data from the Pedra do Cavalo dam for the study period were obtained from Votorantim Energia, the company responsible for dam management.

Water samples (5 L) were collected from the surface with a graduated jar and stored in polyethylene bottles, previously washed with HCl and distilled water. The samples were filtered after each sample using a vacuum pump with glass fiber filters (Whatman GFF - 0.7  $\mu\text{m}$  pore, Sigma-Aldrich, Missouri, USA) for analysis of dissolved inorganic nutrients (nitrite, nitrate, phosphate, silicate, and ammonia). Aliquots (500 mL) were kept frozen until laboratory analysis. The dissolved inorganic nutrients were analyzed at their respective wavelengths using the spectrophotometric method, following Grasshoff *et al.* (1983).

Water samples (250 mL) for species identification were collected in horizontal subsurface trawls using a plankton net (20  $\mu\text{m}$  mesh size) for 5 min at each sampling station. Samples of 1 L were collected in a graduated flask (subsurface samples) for the determination of cell density. All samples were stored in polyethylene flasks and fixed with formaldehyde at a final concentration of 4%.

Taxonomic identification was performed using light microscopy (LM) and scanning electron microscopy (SEM). For the LM analysis, fast slides were mounted and observed under an optical microscope (Olympus® Trinocular CX31RTS5, Tokyo, Japan). Microphotographs were then obtained using the image capture program (QCapture Pro) and a digital camera (QImaging GO-3) attached to the microscope.

For the SEM analyses, cells of *Scrippsiella cf. acuminata* were isolated by capillarity, fixed on round coverslips containing poly-L-Lysine (Sigma-

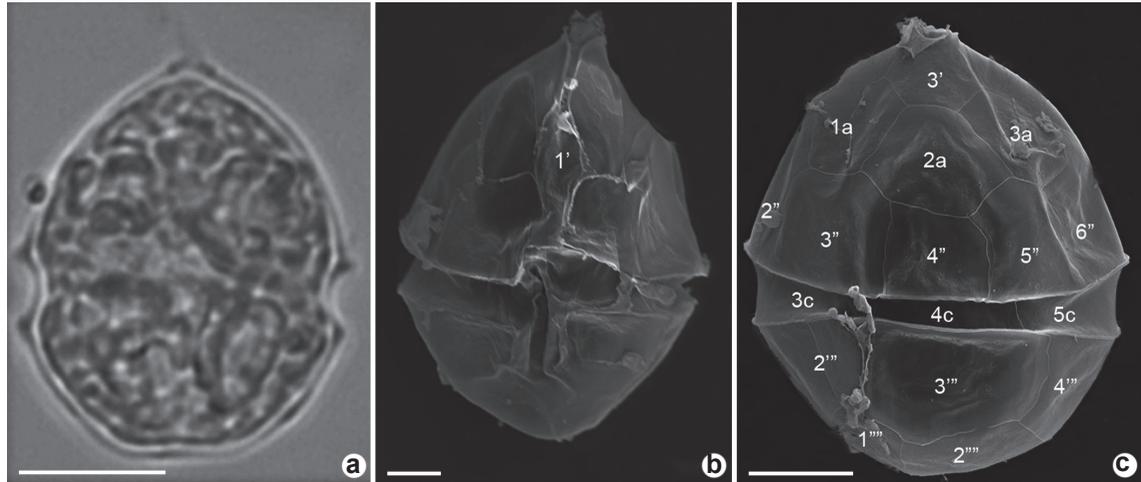
Aldrich, Germany), and then dehydrated in an ethanol series (30%, 50%, 70%, 90%, and 100%) for 10 min at each concentration, with three baths in ethanol at a concentration of 100%. The samples were then critical-point-dried in a Leica EM CPD030 apparatus (Mannheim, Germany), and the slide was subsequently fixed on stubs. The material was ion-sputtered with gold in a Desk IV sputter coater (Denton Vacuum IV, Moorestown, NJ, USA) and analyzed with a JEOL JSM 6390LV (JEOL Ltd., Tokyo, Japan) scanning electron microscope.

Cell density (cells  $\text{L}^{-1}$ ) was evaluated according to Utermöhl (1958) using 2-, 10-, or 50-mL sedimentation chambers; the entire floor of the chamber was counted using an inverted microscope (Motic AE 2000, Hong Kong, China) at 200 $\times$  magnification.

Principal component analysis (PCA) was performed to verify the ordering of the sample points according to the environmental variables (rainfall, water flow, water temperature, water transparency, salinity, nitrite, nitrate, ammonia, silicate, and phosphate) and the density of *Scrippsiella cf. acuminata*. The data for each sample were used separately, using standardized data because of their different measurement units, and the Hellinger transformation was used for the cell density data (Legendre & Gallagher 2001). This analysis was performed in the R software environment (R Core Team 2020) using the ‘Vegan’ package.

The morphological and ultrastructural analyses of the obtained specimens showed that they belonged to *Scrippsiella cf. acuminata*. The cells had an oval cell shape (Fig. 2a), with total length of 15–28.1  $\mu\text{m}$  ( $22.8 \pm 4.7 \mu\text{m}$ ,  $n = 16$ ) and total width of 11.3–25.0  $\mu\text{m}$  ( $17.5 \pm 4.0 \mu\text{m}$ ,  $n = 16$ ). The epitheca was longer than the hypotheca, conical, with convex margins, and a short and hyaline apical process. The cingulum was circular, excavated, and slightly descending, displaced by approximately half the cingular width. The hypotheca was rounded with an excavated sulcus stretching from the cingulum until close to bottom of the hypotheca (Fig. 2b). The SEM analysis showed that the tabulation of the epitheca was ortho-hexa, with plate 1' being narrow and slightly asymmetric (Fig. 2b) and three intercalary plates (a) with almost the same size. Plate 2a was hexagonal, while plates 1a and 3a were pentagonal and unequal precingular plates (Fig. 2c).

According to the current classification, *S. acuminata* is a species complex within the



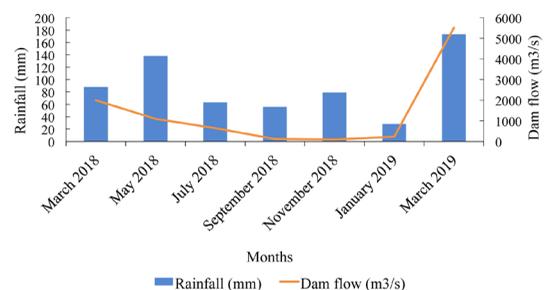
**Figure 2** – a-c. *Scrippsiella* cf. *acuminata* – a. cell under light microscopy; b-c. cell under scanning electron microscopy – b. ventral view of the cell, indicating plate 1'; c. dorsal view of the cell, indicating the plates. Scale bars: a = 10  $\mu\text{m}$ ; b = 3  $\mu\text{m}$ ; c = 3.5  $\mu\text{m}$ .

*Scrippsiella sensu stricto* clade. The morphological variation within this clade is insufficient for the separation of cryptic species (Gottschling *et al.* 2005; Tang *et al.* 2010; Zinssmeister *et al.* 2011); Attaran-Fariman & Bolch (2012) showed by culturing cells of different species of the *Scrippsiella acuminata* complex, that strains of the same ribotype have significant morphological variability, and the species are genetically, but not morphologically, different. The plate pattern morphology of the specimens from the Paraguaçu River estuary is similar to that described for the taxon (Balech 1988; Steidinger & Tanger 1997; Gottschling *et al.* 2005; Zinssmeister *et al.* 2011; Hameed & Saburova 2015; Nunes *et al.* 2019). As species from this complex cannot be distinguished based only on morphological characteristics, and as no molecular analyses were carried out in the present study, we chose to treat the species as *Scrippsiella* cf. *acuminata*.

*Scrippsiella* cf. *acuminata* showed a density variation of 20 cells  $\text{L}^{-1}$  (January 2019) to  $1.8 \times 10^6$  cells  $\text{L}^{-1}$  (March 2018) ( $2.5 \times 10^4 \pm 2 \times 10^5$  cells  $\text{L}^{-1}$ , from March 2018 to March 2019). The highest abundance was registered at the lower course of the river, at an average density of  $5.1 \times 10^4$  ( $\pm 2.9 \times 10^5$  cells  $\text{L}^{-1}$ ). A bloom ( $1.8 \times 10^6$  cells  $\text{L}^{-1}$ ) was recorded in the innermost portion of the lower river course (P1), with lower salinity values (5.7), water transparency (0.3), and higher concentrations of dissolved inorganic nutrients (nitrite, nitrate, phosphate, ammonia and silicate) in March 2018

(Tab. S1, available on supplementary material <<https://doi.org/10.6084/m9.figshare.19763530.v1>>). A previous monitoring performed in 2007–2008 had already registered a *S. cf. acuminata* bloom in this area in December 2007 (Affe *et al.* 2021). These two bloom events did not impact the system, different from the bloom events in which fish deaths (Faria 1914; Yan *et al.* 2002), changes in water color (Gárate-Lizárraga *et al.* 2009, Park *et al.* 2013a, b), and lethal effects in bivalve larvae (Tang & Glober 2012) were registered.

During the study period, rainfall accumulation varied between 28.1 and 173.2 mm in January 2019 and March 2019, respectively (Tab. S1, available on supplementary material <<https://doi.org/10.6084/m9.figshare.19763530.v1>>; Fig. 3). This corresponds to the seasonal pattern in the Paraguaçu River estuary, which comprises a



**Figure 3** – Distribution and relationship between rainfall and dam flow over the months.

dry period (November to February) and a rainy period (March to October), as delimited by Genz (2006) and Genz *et al.* (2008). Variations in rainfall directly interfered with the flow of the Pedra do Cavalo dam (88.6–5,510 m<sup>3</sup>/s) (Tab. S1, available on supplementary material <<https://doi.org/10.6084/m9.figshare.19763530.v1>>; Fig. 3), which as a rule has the highest volumes in the rainy season.

The water temperature ranged from 27.2–32.2 °C along the Paraguaçu River estuary. Salinity ranged from 0.1 to 38.9, with the lowest values in the lower course of the river (points 1–6) and the highest values in the Paraguaçu Channel (points 10–12). Water transparency was of 0.3–3.7 m (Tab. S1, available on supplementary material <<https://doi.org/10.6084/m9.figshare.19763530.v1>>), following the same spatial pattern as salinity. These variation patterns this estuarine gradient have already been well characterized in other studies (Barros *et al.* 2008; Reis-Filho *et al.* 2010).

The concentrations of dissolved inorganic nutrients ranged from values below their detection limits to a maximum of 9.8 µM, 11.4 µM, 23.9 µM, 79.9 µM, and 3.6 µM for nitrite, nitrate, ammonia, silicate, and phosphate, respectively (Tab. S1, available on supplementary material <<https://doi.org/10.6084/m9.figshare.19763530.v1>>). The highest concentrations of these nutrients were found in the lower course of the river, whereas the lowest concentrations were found in Iguape Bay and Paraguaçu Channel. Throughout the area downstream of the Pedra do Cavalo dam, the Paraguaçu River is influenced by the saline wedge of the Todos os Santos Bay, which accompanies the semi-day tidal cycle (Mestrinho 1998). According to Genz *et al.* (2008) waters with salinity above 30 penetrate almost the entire Paraguaçu channel, decreasing as it approaches the low course of the river, where it can reach values below 4.

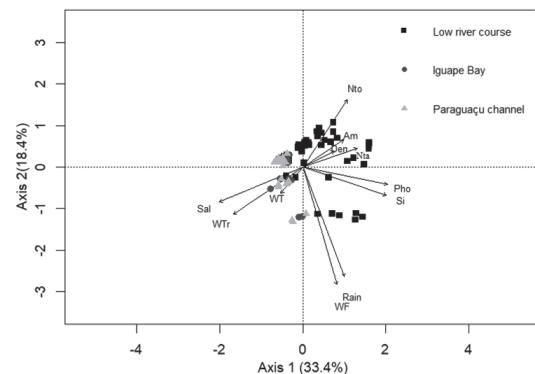
The increase in rainfall and the consequent water flow to the estuary, increased nutrient concentrations, which were probably from the water previously retained in the Pedra do Cavalo dam, A monitoring program by the Institute of Environment and Water Resources - INEMA (SEIA 2020) classified waters from areas that directly influence the Paraguaçu River estuary as mesotrophic and hypereutrophic for our study period (March 2018 to March 2019).

The highest abundance of *S. cf. acuminata* occurred in rainy periods, with higher flows and higher concentrations of dissolved inorganic

nutrients. In July 2018, the species was not recorded at any of the points of the estuarine gradient. During this period, low concentrations of phosphate, silicate, nitrite, and nitrate were observed. These concentrations may be related to a decrease in rainfall associated with the beginning of the dry season and the consequent decrease in the flow and opening of the Pedra do Cavalo dam floodgates (Tab. S1, available on supplementary material <<https://doi.org/10.6084/m9.figshare.19763530.v1>>).

Lower concentrations of dissolved nutrients are characteristic of oligotrophic tropical estuarine environments that commonly have an increase in nutrients during rainy periods, which can alter the phytoplankton community (Bastos *et al.* 2011; Affe & Santana 2016; Affe *et al.* 2018). Some species of dinoflagellates such as *Scrippsiella acuminata* take advantage of this increase in nutrients and originate large cell densities, which can generate blooms (Hallegraeff 1993; Hameed & Saburova 2015; Tas & Yilmaz 2015).

The PCA explained 51.8% of the data variability through two axes (Fig. 4; Tab. 1). The first axis explained 33.4% of the total variance and represented the spatial variation of the system, with less saline waters, lower values of water temperature and water transparency, and higher concentrations of nutrients on the positive side of the axis, representing the region of the lower course of the river; the negative side of the axis



**Figure 4** – Ordering of sampling points according to environmental variables and the density of *Scrippsiella cf. acuminata* in the Paraguaçu River estuary, in the factorial plane 1–2 of the Principal Component Analysis (Rain = rainfall; WF = water flow; WT = water temperature; WTr = water transparency; Salt = salinity; Nto = nitrite; Nta = nitrate; Am = ammonia; Si = silicate; Pho = phosphate; Den = density of *Scrippsiella cf. acuminata*).

**Table 1** – Linear correlation coefficients with the first two axes of Principal Component Analysis (Rain = rainfall; WF = water flow; WT = water temperature, WTr = water transparency; Salt = salinity; Nto = nitrite; Nta = nitrate; Am = ammonia; Si = silicate; Pho = phosphate; Den = density of *Scrippsiella cf. acuminata*).

|      | Axis 1  | Axis 2  |
|------|---------|---------|
| Rain | 0.6854  | -1.3295 |
| WF   | 0.5614  | -1.4252 |
| WT   | -0.3566 | -0.3195 |
| WTr  | -1.1367 | -0.5749 |
| Sal  | -1.3685 | -0.4250 |
| Nto  | 0.7262  | 0.8166  |
| Nta  | 0.8903  | 0.2312  |
| Am   | 0.6688  | 0.3304  |
| Si   | 1.3676  | -0.3413 |
| Pho  | 1.3918  | -0.2101 |
| Den  | 0.5161  | 0.1971  |

comprised waters with lower concentrations of nutrients and higher values of water transparency, water temperature, and salinity, representing the region of Paraguaçu Channel.

Axis 2 explained 18.4% of the data variability, and it had a strong correlation with rainfall, water flow, water transparency, water temperature, salinity, phosphate, and silicate on the negative side of the axis and with nitrogen compounds on the positive side of the axis. The density of *S. cf. acuminata* cells is linked to the highest concentrations of inorganic nutrients dissolved in the environment.

Important ecological traits of the species include short dormancy periods of cysts and repeated cycles throughout the year, in addition to the efficiency in the production of temporary cysts, which provide high adaptability and tolerance to variations in environmental conditions in aquatic systems (Cloern & Dufford 2005). Mobile cells and cysts of *Scrippsiella cf. acuminata* were likely present in the entire estuary of Paraguaçu River; with the discharge of nutrient-rich water from the opening of the floodgates of the Pedra do Cavalo dam, these cells and cysts possibly found favorable conditions for their reestablishment in the estuary, resulting in the *S. cf. acuminata* bloom. *Scrippsiella cf. acuminata* blooms are generally associated with high concentrations of nutrients in eutrophic

environments (Licea *et al.* 2004; Wang *et al.* 2007; Gárate-Lizárraga *et al.* 2009; Okolodkov *et al.* 2014; Hameed & Saburova 2015; Conceição 2016). However, an experiment carried out from a bloom recorded in Port Shelter Bay (Hong Kong) showed that an increase in species biomass could occur even at low concentrations of inorganic nutrients in the environment as long as there is a high concentration of dissolved organic nitrogen (organic matter) (Yin *et al.* 2008). The highest cell density values recorded in the estuary of Paraguaçu River ( $1.8 \times 10^6$  cell L<sup>-1</sup> and  $1.7 \times 10^5$  cell L<sup>-1</sup> in march 2018 and november 2018, respectively) were similar to those found in the Cachoeira River estuary (Ilhéus-BA), with densities between  $4.5 \times 10^4$  and  $1.8 \times 10^5$  cells L<sup>-1</sup> in 2005 (Souza *et al.* 2005), and  $7.3 \times 10^5$  cell L<sup>-1</sup> in May 2016 (Conceição 2016), at a salinity of 7.1–35.9. In this system, a sewage treatment plant (STP) is located near the river; the authors reported that the dumping of waste into the river led to an increase in dissolved nutrients, providing conditions for the increase of the *S. acuminata* population.

Organic nutrient content was not measured in the present study. Nevertheless, domestic sewage inputs from riverside populations in the Paraguaçu River estuary probably influenced our results as we observed the presence of sewage in the vicinity of the area where algae proliferation was recorded in March; moreover, this was the only area in which ammonia was detected in the estuary in March. Furthermore, in March, the highest flow of the dam was observed and margins of *Eichhornia crassipes* (Mart.) Solms were found close to the area where the *S. cf. acuminata* bloom occurred. This species is commonly found in environments impacted by domestic dumps and characterized by absorbing large amounts of nitrogen and phosphorus (Esteves 1982). When decomposed, these components are released into the environment. The sum of these factors may explain the increased concentrations of phosphate, nitrate, and ammonia during this period, promoting conditions favorable to the high cell density of *S. cf. acuminata* in the estuarine system.

In 2019 there were no HAB events of *Scrippsiella cf. acuminata*. Among the months covered in this study, March 2019 presented higher values of rainfall and flow, and a low density of *S. acuminata* cells, recorded only for the region of the lower river course. This shows that, probably, the *Scrippsiella* cells may have been carried away by the flow of the river, not allowing the establishment of a large biomass of these organisms.

*Scrippsiella* cf. *acuminata* occurred along the entire Paraguaçu River estuary, with higher densities in regions with lower salinity and higher concentrations of nutrients. Factors such as the increase in nutrients from the opening of the Pedra do Cavalo dam or from sewage can lead to new HABs. This study allowed us to determine the distribution patterns of *S.* cf. *acuminata* and the environmental characteristics related to the species' occurrence, thus serving as a basis for future studies and monitoring programs of potentially harmful species to avoid the occurrence of new algal blooms.

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