Structure of phytoplankton functional groups in shallow lakes with different equilibrium states

Estrutura de grupos funcionais fitoplanctônicos em lagos rasos com diferentes estados de equilíbrio

Núbia da Silva¹; João Paulo de Oliveira Santos¹; Danielle Lima de Oliveira²; Kelly Dayane Pereira da Silva¹; Ênio Wocioli Dantas³ & Luciana Gomes Barbosa¹

ABSTRACT
Changes in equilibrium states in shallow lakes are associated with disturbances and resilience. The study evaluated the structure and composition of phytoplankton functional groups in two lakes, Lagoa do Paó (turbid waters) and Santa Lúcia (clear waters). Monthly collections of limnological variables and phytoplankton occurred between August 2014 and June 2015 in the pelagic region. The lakes shared five functional groups: K, S₁, X₁, W₂ and P, formed by coccoid and filamentous cyanobacteria, coccoid chlorophytes and euglenophytes, and desmids, respectively. The canonical correspondence analysis evidenced a clear separation, highlighting the groups T₄, N, W₂ and S₁ in Santa Lúcia and functional groups composed of potentially toxic cyanobacteria in Lagoa do Paó (S₅, L₀), indicating that the submerged macrophyte coverage can act promoting the phosphorous stabilization in the sediment in Santa Lúcia, reducing its contribution to the water column and inhibiting the dominance of the mentioned species. Temperature, pH, phosphorus and light attenuation coefficient influenced the occurrence of H₁, SN, S₁, W₂, X₁, D, and L₀, in the Lagoa do Paó. The composition and structure of the functional groups responded in the two lakes with some functional groups associated with toxic cyanobacteria occurring exclusively in Lagoa do Paó, having also been observed the sharing of some functional groups.

Keywords: eutrophication; bloom; macrophytes; nutrients; turbidity.

RESUMO
Mudanças nos estados de equilíbrio em lagos rasos estão associadas a perturbações e resiliência. O estudo avaliou a estrutura e composição de grupos funcionais fitoplanctônicos em dois lagos, Lagoa do Paó (água turbida) e Santa Lúcia (água clara). Coletas mensais de variáveis limnológicas e fitoplanctônicas ocorreram entre agosto de 2014 e junho de 2015 na região pelágica. Os lagos compartilharam cinco grupos funcionais: K, S₁, X₁, W₂ e P, formados por cianobactérias coccoides e filamentosas, clorofíceas e euglenofíceas coccoides e desmídeas, respectivamente. A análise de correspondência canônica evidenciou nítida separação, destacando os grupos T₄, N, W₂ e S₁ em Santa Lúcia e grupos funcionais compostos de cianobactérias potencialmente tóxicas na Lagoa do Paó (S₅, L₀), indicando que a cobertura de macrofitas submersas pode atuar promovendo a estabilização do fósforo no sedimento em Santa Lúcia, reduzindo seu aporte na coluna d’água e inibindo a dominância das espécies mencionadas. Temperatura, pH, fósforo e coeiciente de atenuação de luz influenciaram a ocorrência de H₁, SN, S₁, W₂, X₁, D, L₀, na Lagoa do Paó. A composição e estrutura dos grupos funcionais responderam nos dois lagos com alguns grupos funcionais associados a cianobactérias tóxicas ocorrendo exclusivamente na Lagoa do Paó, tendo sido também observado compartilhamento de alguns grupos funcionais.

Palavras-chave: eutrofização; floração; macrofitas; nutrientes; turbidez.

¹ Núcleo de Pesquisas em Limnologia (Nuliba), Programa de Pós-Graduação em Biodiversidade, Centro de Ciências Agrárias, Universidade Federal da Paraíba (UFPB), Campus II, Rodovia PB 079 – CEP 58397-000, Areia, PB, Brazil.
² Correspondence author: nubiaetnobio@gmail.com.
³ Correspondence author: nubiaetnobio@gmail.com.
INTRODUCTION

Shallow lakes are considered potentially unstable ecosystems, vulnerable to climatic conditions and anthropic pressures. It significantly contributes to change the characteristics of these water bodies, which, for being shallow, are constantly affected by the accumulation of phosphate and nitrogenous nutrients (PHILLIPS et al., 2016). This fact makes them more and more vulnerable to eutrophication, what affects not only water quality, but also the composition of macroscopic and microscopic species (JANSSSEN et al., 2014).

The factors that contribute to the alteration of the characteristics of shallow lakes are associated with the size of the water body and its low volume and are due to either natural stressors (climate) or anthropic stressors (irrigation, sewage discharge), which favor the modification of structure and dynamics, making them more susceptible to changes in their equilibrium state when compared to deep lakes (NASSELI-FLORES, 2003; JANSSSEN et al., 2014).

Due to their great fragility, these ecosystems have drawn attention in scientific studies since the 1970s, when the hypothesis that shallow lakes can alter their equilibrium state, differing from the original, was raised (MAY, 1977). Those with predominance of submerged macrophytes tend to exhibit a clear water profile, with reduced concentrations of nutrients and phytoplankton biomass, because the submerged vegetation acts as a filter, absorbing the nutrients available in the water body and elevating sedimentation levels (SCHEFFER & VAN NES, 2007; BLINDOW et al., 2014).

In the absence of submerged vegetation or through processes of decomposition of these plants, the environment starts to be dominated by phytoplankton, especially cyanobacteria, which express optimal development under mixing conditions, high-nutrient concentrations and reduced euphotic zone (BLINDOW et al., 2014). Depending on the magnitude and duration of the entry of allochthonous sources, trophic network and its relationship with nutrient cycling, such abrupt changes of state, can be avoided (SCHEFFER, 1998).

Phytoplankton can be found in environments with varying levels of nutrients, pH, temperature, light and turbulence. Consequently, temporal and spatial fluctuations, especially in their composition (biomass), can be considered good indicators of changes in shallow lakes (PEREIRA, 2013).

Thus, the method of classification through polyphyletic groups sharing certain adaptive characteristics of different environments and environmental conditions (REYNOLDS et al., 2002; PADISÁK et al., 2009) presents itself as an effective instrument in ecosystem characterization, allowing the prediction of the associated abiotic factors, as well as those more specific in both clear- and turbid-water lakes (IZAGUIRRE et al., 2012).

In this context, this study aimed to evaluate the composition and structure of phytoplankton functional groups in lakes with different alternative stable states (clear water and turbid water). The main question—do the composition and structure of functional groups respond to differences in structure and equilibrium states of shallow lakes?—is associated with the hypothesis that functional groups formed by potentially toxic cyanobacteria are typical assemblies of turbid and eutrophicated lakes.

MATERIAL AND METHODS

Study area characterization

The study was conducted in two shallow lakes (Santa Lúcia and Lagoa do Paó), both belonging to the Mamanguape river basin (Paraíba, Northeast, Brazil), climatically classified as AW (ALVARES et al., 2013) (Figure 1).
Santa Lúcia (06º50’14.2” S; 35º19’51.4” W) is a small shallow lake (444 m long and 128 m wide) located in a rural area of the municipality of Araçagi, and it is used for fishing, domestic tasks, irrigation and leisure. Its ecosystem is considered oligo-mesotrophic, and it has dense cover of submerged macrophytes, mainly *Nitella cernua* A. Braun.

Lagoa do Paó (07º02’28.4” S; 35º37’55.0” W) is located in the urban area of Alagoa Grande (834 m long and 266 m wide) and it is intensively used for fishing. For decades, this lake was characterized by the massive presence of *Eichornia crassipes* (Mart.) Solms, which in 2009 was completely removed by anthropic action. In addition to the discharges of domestic sewage, the ecosystem currently has turbid waters and bloom of potentially toxic algae.

**Sampling**

Hydrometeorological information was obtained from the Executive Management of Monitoring and Hydrometry of the Executive Agency of Water Management of Paraíba State (Agência Executiva de Gestão das Águas do Estado da Paraíba—AESA, 2013).

Samples were collected at monthly intervals, between August 2014 and June 2015 (except November), at a point in the central region of the environment. Data of temperature (ºC), electrical conductivity (μs/cm⁻¹), hydrogen potential (pH), dissolved oxygen (mg/L) and turbidity were measured *in situ* with a multiparametric probe, and water transparency (m) was determined based on the depth of visual disappearance of Secchi disc (ZSD). The vertical light attenuation coefficient (k) was calculated using the ratio k = 1.7 x ZSD⁻¹ (Poole, & Atkins, 1929). The euphotic zone (Zeu) was empirically calculated by multiplying the value obtained through Secchi disc by 2.7 (Cole, 1983). The value of depth was considered for the mixing zone (Zmix), since there were no differences between the temperature profiles during the entire study period, and the light availability index in the mixing layer was determined based on the ratio between Zeu/Zmix (Jensen et al., 1994).

Water samples were collected at the subsurface of the water column in each of the environments and kept frozen to avoid degradation of the compounds of interest. The concentrations of total phosphorus and soluble reactive phosphorus in the water were determined by the ascorbic acid method, according to the methodology described by American Public Health Association (APHA, 2005).
Biological parameters

Phytoplankton

Quantitative samples were collected by dipping a 250-mL polyethylene bottle into the subsurface and subsequently adding Lugol’s solution to it. These samples were used for identification and counting of species, respectively.

Phytoplankton was counted under an inverted microscope by the sedimentation method (UTERMÖHL, 1958). For the error to be lower than 5% and the confidence coefficient higher than 95%, a minimum of 100 individuals of at least the two most frequent species were counted, and the sedimentation time was 4 h for each centimeter of chamber height, as established by Lund et al. (1958). The results were expressed as density (ind.mL$^{-1}$) and calculated according to the formula described by Ros (1979).

The biovolume of each species was estimated based on the product between its cell volume and density. Cell volume was calculated based on the volume of the geometric solid that was most closely similar to the cellular form, isolated or combined, according to Hillebrand et al. (1999) and Sun & Liu (2003), and from the mean values of the measurements of 20 to 30 individuals, when possible. Biovolume was expressed in fresh weight unit, in which 1 mm$^3$ L$^{-1}$ = 1 mg/L$^{-1}$ (WETZEL & LIKENS, 2000).

Functional groups

The species found were classified according to the functional group approach, taking as reference the system of functional classification of freshwater phytoplankton, contained in Reynolds et al. (2002) and revised by Padisák et al. (2009), from the descriptor species, that is, those which reached a volume equal to or greater than 5% of the total biovolume of each sample.

Data analysis

A canonical correspondence analysis (CAA) was performed to verify the relationship between abiotic and biotic variables (with functional groups), using the R program, vegan package, Envif command, to identify the significance of the variables. Subsequently, analysis of variance (ANOVA) was carried out.

RESULTS

According to the monthly rainfall distribution, the corresponding region recorded peaks of similar precipitation, concentrated in the months of September, March and June (Figure 2). In this study, precipitation was relatively low and had little influence on the environments, considering the reduced rainfall occurring during the studied period. In Lagoa do Paô, high levels in physicochemical properties were found when compared to Santa Lúcia, for the corresponding months.
The environments had pH above 7 and remained alkaline in all months. Water temperature was above 27°C, reaching 34.6°C. The contents of total phosphorus and soluble reactive phosphorus in Santa Lúcia were low (< 40 µg/L⁻¹), classified as oligotrophic (with values below 44 µg/L⁻¹). Lagoa do Paó had total phosphorus contents above 86 µg/L⁻¹ and reactive phosphorus contents above 81 µg/L⁻¹, hence being characterized as eutrophic (TSI ≥ 54 µg/L⁻¹).

### Functional groups

The 36 phytoplankton species recorded in the Santa Lúcia lake were classified into eight functional groups: K, S₁, X₁, P, W₂, M, T, and N. In Lagoa do Paó, 22 species were identified, which were classified into ten groups: K, S₁, X₁, P, W₂, S₀, F, E₀, D, and H₁, as shown in Figures 3A and 3B. Both environments shared five functional groups: K, S₁, X₁, W₂, and P.
In Santa Lúcia, the groups N and S$_1$ occurred along the entire sampling period. N had more than 70% of biomass between January and February, reaching 96% in April, represented by the diatoms (Pennales sp., and Synedra sp.) and desmids (Cosmarium sp., C. biretum Brébisson ex Ralfs, C. regnellii Wille, C. quadrum P. Lundell, and Staurastrum taylori Gronblad). The group S$_1$—Planktolyngbya sp., Planktotorix agardhii (Gomont) Anagnostidis & Komárek, Phormidium sp., and Spirulina sp.—showed the highest biomass value in December (68% of the total).

In Lagoa do Paó, the group S$_N$—Cylindrospermopsis raciborskii (Woloszynska) Seenayya & Subba Raju, and Raphidiopsis curvata F. E. Fritsch & M. F. Rich—showed 50% of biomass in September, S$_1$ showed 70% of biomass in October, and W$_2$ (Trachelomonas sp., Trachelomonas volvocina Ehrenberg, and Trachelomonas volvocinopsis Svirenko) stood out with 53% of total biomass in May. Among the functional groups exclusive to Lagoa do Paó, most were composed of potentially toxic cyanobacteria: S$_N$, H$_1$ (Dolichospermum sp., Anabaenopsis sp., and Cuspidothrix sp.), L$_o$ (Chroococcus sp., and Merismopedia sp.), F (Sphaerocystis sp.), and D (Tabellaria sp.), groups that were not found in Santa Lúcia (Figure 3B).

The CCA, performed with eight variables and 12 functional groups of the two environments, explained 73.87% of the data variability (axis 1 with 48.79% and axis 2 with 26.08%), according to the analysis of variance. The first two axes, corresponding to pH and water temperature, were the most significant, followed by total phosphorus, P-ortho, $Z_{mix}$, $Z_{eu} = Z_{mix}$ and K (table 2). There was a clear separation between the two environments, in which $Z_{eu} = Z_{mix}$ and $Z_{mix}$ were related to axis 2 (sampling units of Santa Lúcia), which influenced the occurrence of the functional groups N, T$_D$, S$_1$, W$_2$ in almost every month, except August/14, September/14 and May/15.
The axis 1 (negative side), for the sampling units of Lagoa do Paó, indicated the occurrence of groups common to environments with high-nutrient content, and the most important variables in their ordering were pH and water temperature, influencing the functional groups S, and H1 in almost all studied months, except April, May and June. Another grouping corresponding to the sampling units of Lagoa do Paó (positive side of axis 1) associated the total phosphorus, P-ortho and K (light attenuation coefficient) with the groups L, D, X1, W2 and F, predominant in the months of April, May and June (Figure 4).

Figure 4 – Canonical correspondence analysis of the sampling units of the lakes Santa Lúcia and Lagoa do Paó generated from eight abiotic variables and the descriptor functional groups of the phytoplankton community of both sites. Source: primary.

Table 2 – Summary of the CCA indicating the correlation coefficients between the phytoplankton functional groups and environmental variables of the Santa Lúcia lake in the period from August 2014 to June 2015*.

<table>
<thead>
<tr>
<th>Variables</th>
<th>CCA 1</th>
<th>CCA 2</th>
<th>r²</th>
<th>P ( &gt; r)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hydrogen potential (pH)</td>
<td>0.98591</td>
<td>-16728</td>
<td>0.6620</td>
<td>0.001***</td>
</tr>
<tr>
<td>Water temperature (°C) (Twater)</td>
<td>0.97836</td>
<td>-20692</td>
<td>0.1296</td>
<td>0.312</td>
</tr>
<tr>
<td>Total phosphorus (mg L⁻¹) (Pt water)</td>
<td>0.98450</td>
<td>0.17541</td>
<td>0.6182</td>
<td>0.001***</td>
</tr>
<tr>
<td>Soluble reactive phosphorus (μg.L⁻¹) (P-ortho)</td>
<td>0.99604</td>
<td>0.08890</td>
<td>0.6514</td>
<td>0.001***</td>
</tr>
<tr>
<td>Mixing zone (m) (Zmix)</td>
<td>-0.91387</td>
<td>-0.40600</td>
<td>0.6911</td>
<td>0.002**</td>
</tr>
<tr>
<td>Euphotic zone / Mixing zone (Zeu,Zmix)</td>
<td>-0.98009</td>
<td>-0.19853</td>
<td>0.9252</td>
<td>0.001***</td>
</tr>
<tr>
<td>Electrical conductivity (µs.cm⁻¹) (Cond)</td>
<td>0.96040</td>
<td>0.27864</td>
<td>0.0886</td>
<td></td>
</tr>
<tr>
<td>Light attenuation coefficient (K)</td>
<td>0.99644</td>
<td>0.08427</td>
<td>0.7420</td>
<td>0.001***</td>
</tr>
</tbody>
</table>

Level of significance (%); CCA: canonical correspondence analysis; ANOVA: analysis of variance; **P ≤ 0.01; ***P ≤ 0.001.
DISCUSSION

In tropical lakes and lagoons, the primary productivity of phytoplankton depends on factors such as radiation availability, nutrient concentrations, temperature and pH (REYNOLDS, 2006). The synergistic interaction of some of these factors, as indicated in the CCA through the variables associated with the underwater light climate and nutrients (e.g., \( Z_{\text{eu}} = Z_{\text{mix}}, Z_{\text{mix}}, \) light attenuation coefficient and orthophosphate), was determinant in the limnological characterization of the lakes and in the understanding of the dynamics of their phytoplankton functional groups, leading to monthly fluctuation of functional groups both in Santa Lúcia and Lagoa do Paô.

Besides these factors, the CAA indicated that the composition and structure of the functional groups were closely related to the peculiar characteristics of both aquatic ecosystems and their equilibrium states. The influence of light availability on the mixing layer revealed how much water transparency and the level of infestation of submerged macrophytes exert a strong effect on the variation of algal biomass (COSTA & DANTAS, 2011) and on the composition of the functional groups, as observed for the groups T and N, with affinity for permanently oligo-mesotrophic waters with slow flow and the presence of submerged macrophytes (PADISÁK et al., 2009). Also in this context, the depth of an ecosystem can exert a strong influence on the behavior of phytoplankton associations in tropical eutrophic reservoirs, due to the propensity to water column mixing (DANTAS et al., 2012).

Santa Lúcia and Lagoa do Paô were different in relation to limnological characteristics (underwater light climate and nutrient concentrations, for example), evidencing a spatial-temporal variation in their functional groups, and macrophytes were a preponderant factor in the inhibition of phytoplankton in Santa Lúcia.

Submerged aquatic vegetation, as observed in Santa Lúcia, plays a regulating role in the composition and structure of shallow lake microalgae, and the occurrence of desmids (group N), for example, was related to their affinity for meso-oligotrophic environments adhered to the periphyton and present in the metaphyton (CROSSETTI et al., 2013). Thus, the group N, represented in Santa Lúcia by diatoms and desmids, is composed of species susceptible to high concentrations of phosphorus and high pH, which explains the absence of this group in Lagoa do Paô (REYNOLDS et al., 2002; PADISÁK et al., 2009).

In Santa Lúcia, the group S\(_1\), formed by species considered strong competitors, typical of shallow and well-mixed lakes, had high biomass (REYNOLDS et al., 2002). The shading facilitated by submerged macrophytes, creating a dense cover on the water column surface and preventing light from reaching the bottom, may have favored the emergence of cyanobacteria, as well as diatoms, euglenophytes and cryptophytes (PINTO & O’FARRELL, 2014).

The irrigation use in Santa Lúcia and the load of urban effluents (sewage) discharged in Lagoa do Paô are indicative of interferences of anthropic actions in these lakes and, consequently, in the alternation of their equilibrium states. Such alternation exhibits a contrasting scenario of functional groups consisting of potentially toxic cyanobacteria (S\(_2\), L\(_1\), H\(_1\), F, D), exclusive to Lagoa do Paô, indicating that the submerged macrophyte cover can function as an inhibitor of the dominance of these groups (CROSSETTI et al., 2013).

Due to the low depth in the two environments, isothermal profile with high temperatures in Lagoa do Paô, the temperature and alkaline pH were influencing factors on the occurrence of the groups S\(_1\), H\(_1\), S\(_3\) and W\(_2\), whose success of occurrence is associated with high temperatures and pH, scarcity of rains and excess of nutrients (PINTO & O’FARRELL, 2014).

The group S\(_1\) (exclusive to Lagoa do Paô), whose dominant species was *Cylindrospermopsis raciborskii* (Woloszynska) Seenayya & Subba Raju, has high-phenotypic plasticity associated with pigments, size and growth rates, being more tolerant within a broad gradient of temperature and light, compared to other ecologically similar species, such as *Planktothrix agardhii* (Gomont) Anagnostidis & Komárek (BONILLA et al., 2012), the latter being a potentially toxic species of occurrence in Santa Lúcia, classified in group S\(_1\).

The functional group H\(_1\) (*Dolichospermum* sp., *Cuspidothrix* sp., and *Anabaenopsis* sp.), represented by heterocystous cyanobacteria, is associated with eutrophic environments, mixed. However, the species *Dolichospermum* sp. was found in Santa Lúcia in all months, being associated
with water column mixing, a characteristic also shared by group $S_1$ (REYNOLDS et al., 2002; PADISÁK et al., 2009).

Total phosphorus, orthophosphate, light attenuation coefficient and conductivity are indicators of anthropogenic pollution, which, in this study, were related to Lagoa do Paó, influencing the occurrence of the groups $X$, $D$ and $L_o$, common in waters with high-nutrient concentrations, represented by pioneer species that invest in rapid cellular replication and whose propagules are easily dispersed by wind or by the body of other animals such as birds and insects (REYNOLDS, 1999).

Urban lakes, without riparian forest, have high potential for phosphorus and nitrogen accumulation, exhibiting high growth of toxic algae, which form a layer on the surface of the water, allowing the shading and increase of turbidity, and consequent limitation by light, also found in the present study (FÉLIX et al., 2015). Moreover, the emission of toxins by cyanobacteria, in addition to compromising water quality, can lead to biological contamination, for example, of fish, generating, this way, bioaccumulation (FÉLIX et al., 2015).

In this work, phytoplankton structure and composition directly responded to changes and alternations in the equilibrium state of the ecosystems, recording high specificity in relation to environmental variables (IZAGUIRRE et al., 2012; CROSSETTI et al., 2013; SANCHÉZ et al., 2015). Thus, this study confirms that phytoplankton composition and structure respond to physical and chemical characteristics, and the classification into functional groups is an effective way to predict possible changes in their equilibrium states.

ACKNOWLEDGMENTS

To the Coordination for the Improvement of Higher Education Personnel (CAPES), for the financial support, and to the Universidade Federal da Paraíba and the Laboratory of Limnology (UFPB/CCA), for the technical and logistic support.

REFERENCES


