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The combined use of paleolimnological and long-term limnological information to identify natural and anthropogenic environmental changes

O uso combinado de informação paleolimnológica e limnológica histórica para identificar mudanças ambientais naturais e antropogênicas

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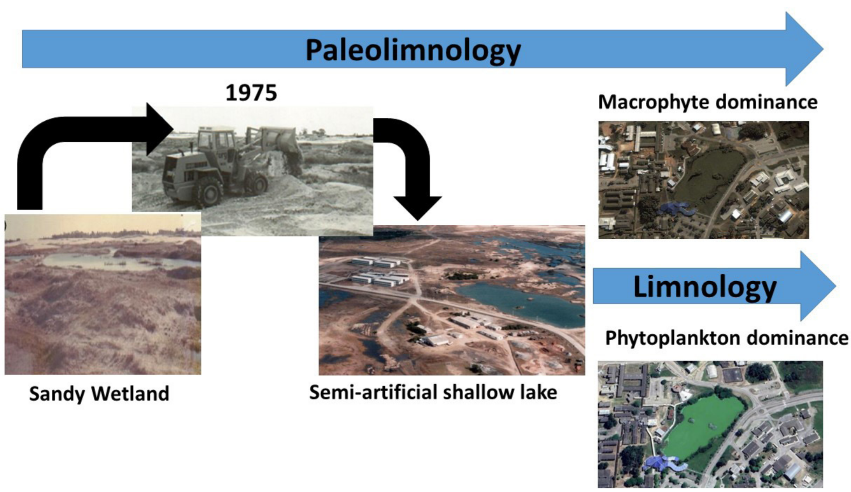
Abstract: Aim: Urbanization leads to rapid changes in ecosystem structure and function. Wetlands on university campuses under urbanization pressure could be used as case studies of multidisciplinary aquatic research and good environmental practices promoting sustainability. **Methods:** A paleolimnological study was undertaken in a semi-artificial lake on a university campus in southern Brazil to trace historical impacts and ecological changes back to the mid-1970s through complementary approaches: historical data, nutrients, $\delta^{13}C$ and $\delta^{15}N$ stable isotopes, diatoms, microplastics and associated microbial community analysis. **Results:** The eutrophication process started to intensify after the lake was used for nocturnal roosting by waterbirds, and especially after the establishment of constructions along the margins with septic tank sanitary sewage, which eventually spilled and leached into the lake. Over decades, we identified a limnological hypertrophication process leading to recurrent cyanobacterial blooms and massive macrophyte proliferation coupled with changes in isotopic ratios and algal occupation with several transitions between shallow lake alternative states. Such a limnological process has resembled the paleolimnological eutrophication trends and isotopic changes in sedimentary organic matter. The microplastic deposition was detected as a proxy for the intensification of urbanization, especially during the construction of the University facilities. **Conclusions:** The combined use of paleolimnological and historical limnological data represents a powerful approach for inferring both natural and cultural impacts on the lake, and identifying management strategies based on such scientific information.

Keywords: diatoms; eutrophication; microplastics; pollution; trophic state.

Resumo: Objetivo: A urbanização leva a mudanças rápidas na estrutura e função do ecossistema. Terras úmidas em câmpus universitários sujeitos ao desenvolvimento da urbanização podem ser usados como estudos de caso de pesquisa aquática multidisciplinar e boas práticas ambientais que promovam a sustentabilidade. **Métodos:** Foi realizado um estudo paleolimnológico em um lago semiartificial em um campus universitário no sul do Brasil para inferir impactos históricos e mudanças ecológicas desde a década de 1970 por meio de abordagens complementares: dados históricos, nutrientes, isótopos estáveis $\delta^{13}C$ e $\delta^{15}N$, diatomáceas, microplásticos e análise da comunidade microbiana associada. **Resultados:** O processo de eutrofização começou a se intensificar após o uso noturno de árvores como poleiros por aves aquáticas, e especialmente após o estabelecimento de construções ao longo das margens com fossa séptica de esgoto sanitário, que ocasionalmente transbordava e carreava material para o lago. Ao longo de décadas, identificamos um processo limnológico de hipertrofização que induziu a florações recorrentes de cianobactérias e proliferação maciça de macrófitas, juntamente com mudanças isotópicas e ocupação de algas com várias transições entre estados alternativos de lagos rasos. Tal processo limnológico foi claramente espelhado nas tendências de eutrofização paleolimnológica e mudanças isotópicas da matéria orgânica sedimentar. A deposição de microplásticos foi detectada como *proxy* para a intensificação da urbanização especialmente durante a construção das instalações da Universidade. **Conclusões:** O uso combinado de dados paleolimnológicos e limnológicos históricos representa uma abordagem poderosa para identificar os impactos naturais e culturais no lago e identificar estratégias de manejo baseadas em tais informações científicas.

Palavras-chave: diatomáceas; eutrofização; microplásticos; poluição; estado trófico.

Graphical Abstract



1. Introduction

Studies with detailed historical records of successive impacts on wetlands and how they change the characteristics of such habitats are scant. We undertook a paleolimnological investigation in a shallow lake under a hypertrophication process of the campus of the Federal University of Rio Grande (FURG), as an input to identify management options. Knowledge of long-term changes in natural systems is fundamental for reducing the vulnerability of their associated socio-ecological systems in terms of livelihood security, resilience, risk and sustainability (Behling, 1995; Ekblom, 2012; Hennemann et al., 2015; McGlue et al., 2015; Velez et al., 2018; Wang et al., 2012). In the case of shallow lakes, paleolimnological reconstructions can support both ecological research and management by providing long-term decadal, centennial and millennial proxy information that would not be obtainable otherwise by direct observations. In this sense, Velez et al. (2018) emphasized that there is an important lack of information on the use of paleoenvironmental data in such assessments, with only a few papers published in journals oriented toward an interdisciplinary readership of both natural and social scientists. Therefore, a study dealing with paleolimnology in socioecological systems remains a timely issue, especially under the availability of direct limnological observations. Learning from past decisions on land use and occupation, as well as the associated environmental consequences, could redirect daily practices, management actions, and eventually mitigate impacts still underway.

To address this issue practically, it would be necessary to identify a case for study in which long-term information on the transformation of a natural system by socioeconomic development is available. This would enable the identification of the salient socioeconomic milestones and assessment of their associated effects on the natural environment through a comparison of historical information to sedimentary proxies. Fortunately, lake ecosystems hold high-resolution sedimentary records that can be used to reconstruct paleoenvironmental changes and relate them to well-documented historical anthropogenic information (Smol, 2008). To achieve this, a set of useful paleolimnological proxies could include (i) documented history of local human interference to interpret the historical changes inferred from sedimentary profiles (Burdge, 1991; Smol, 2008); (ii) changes in the composition of diatom communities as biological proxies for environmental change (Smol & Stoermer, 2010); (iii) lake trophic state variability inferred from nutrient concentration and isotopic composition of bulk organic matter as nutrients modulate limnological function and structure (Lamb et al., 2006; Savage, 2005); and (iv) contamination by microplastics (hereafter, MPs) and associated microbial organisms, i.e., the plastisphere (Browne et al., 2011; van Cauwenberghe et al., 2015; Turner et al., 2019; Woodall et al., 2014; Zettler et al., 2013). Plastics are only of use for recent strata in paleolimnology and are made of synthetic thermoplastic polymers that can be classified as macro- (> 5 mm) or micro (< 5 mm) (Castañeda et al., 2014; Galgani et al., 2015; Zalasiewicz et al., 2016). MPs can be transported over long distances, and sedimentation and burial can be accelerated through colonization by organisms (i.e., the plastisphere, Browne et al., 2011; Woodall et al., 2014; Zettler et al., 2013).

We investigated a semiartificial shallow lake, i.e., an excavated wetland transformed into a shallow lake, on the campus of the Federal University of Rio Grande (FURG), whose original formation was precisely dated to 1975 (FURG Master Plan, FURG, 2022), so it is possible to reconstruct its entire history and modifications. There is precise long-term chronological information on anthropogenic actions and limnological research to help explain decadal and interannual lake changes. It is therefore possible to undertake an integrated long-term campus development analysis and infer the interrelation between natural changes and associated university development and to understand the process of environmental transformation. Therefore, the ultimate aim of this paper is to identify milestones in the history of long-term lake changes and propose a set of environmental and social management measures to introduce the concept of environmental management performance (EMP, *sensu* Trumpp et al., 2015), as already suggested by Roos et al. (2020) in higher education institutions, towards achieving best possible campus sustainability (Brinkhurst et al., 2011). Briefly, this concept considers five dimensions: environmental policy, environmental objectives, environmental processes, organizational structure, and environmental monitoring.

2. Material and Methods

2.1. Description of the study area

The coastal plain of southern Brazil was formed during the Quaternary by the juxtaposition of deposits of barrier-lagoon systems designated I to IV (Villwock et al., 1986). Each barrier-lagoon system corresponds to a high-frequency depositional sequence (Rosa et al., 2017), with Lake Biguás (32°04’43”S; 52°10’03”W) sitting on Holocene barrier-lagoon IV (see 1920 aerial photograph, Figure 1) dominated by sandy segments (Dillenburg et al., 2017). The mean annual temperature varies between 13 °C (winter) and 24 °C (summer), and the total annual rainfall ranges between 1200 and 1500 mm yr^{-1}. The climate is characterized by *Cfa* (humid subtropical) *sensu* Köppen classification. Lake Biguás is located at the Federal University of Rio Grande, Brazil (Figure 1). The lake is 1.5 ha in area, and the maximum depth is 2 m but modulated by rainfall. The lake holds aquatic macrophytes with a predominance of emergent and floating forms, with episodes occurring of excessive growth and transitions between alternative states, from clear to turbid waters, especially during the last two decades (Albertoni et al., 2014; Silva et al., 2015). The lake is eutrophic or hypertrophic and subject to bird guanotrophication (Furlanetto et al., 2012; Silva et al., 2015) from the feces of Neotropic cormorants (*Nannopterum brasilianum*, Biguás in Portuguese), as well as a low number of ibises and herons (Barquete et al., 2008a). Waterbirds use trees on two islands of the lake for nocturnal roosting (Barquete et al., 2008b) and can contribute 40% and 75% of the total nitrogen and phosphorous, respectively, in wetland ponds (Kitchell et al.,

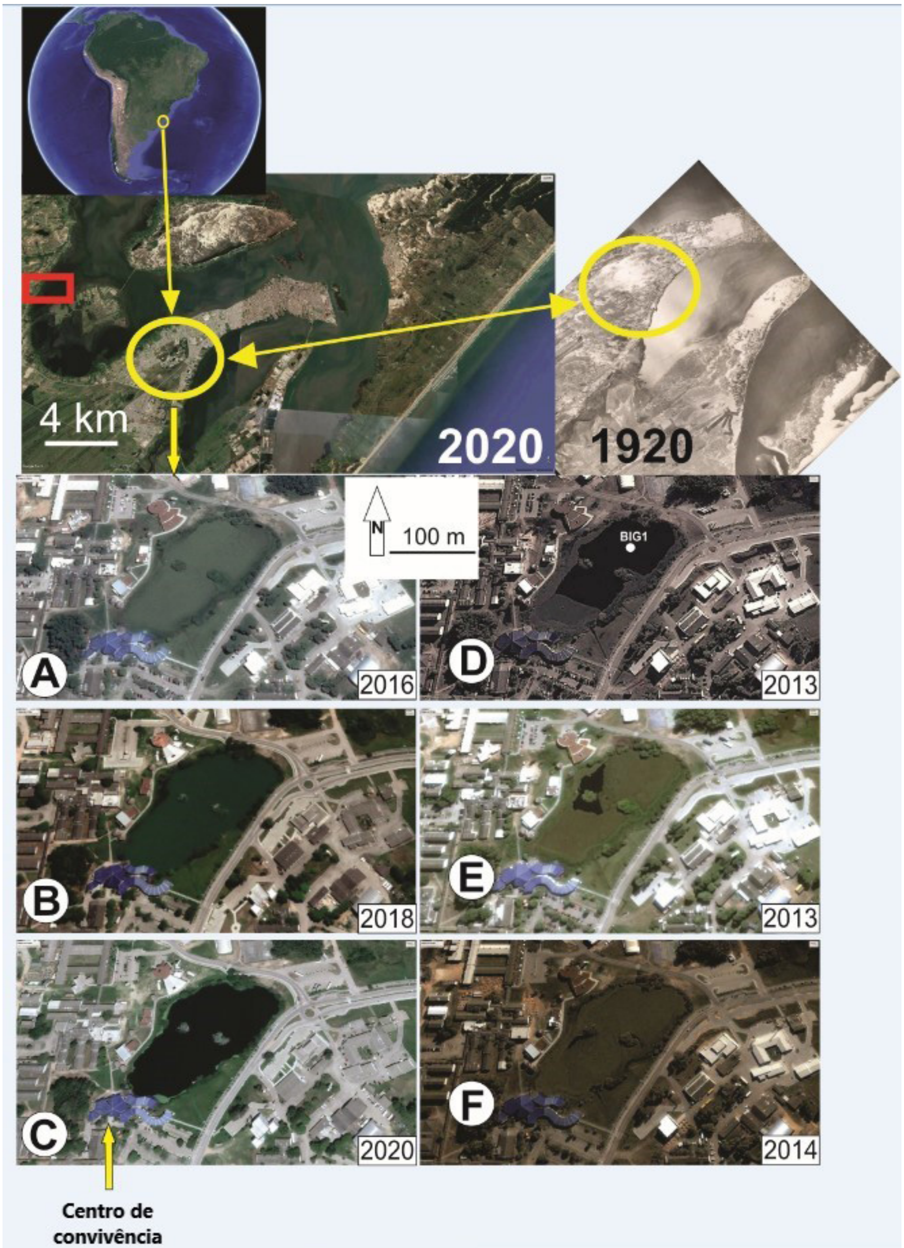


Figure 1. Upper panels show an overview of the city of Rio Grande, indicating the position of the university campus with a yellow circle for both 2020 and 1920. The red box shows the location where the organic matter isotopic was survey carried out by Patterson (2016). (A), (B) and (C) show examples from Lake Biguás under a limnological phase dominated by phytoplankton, while (D), (E) and (F) show examples of a limnological phase dominated by macrophytes under three conditions of macrophyte proliferation. Aerial photographs from Google Earth.

1999). Guanotrophication into Lake Biguás leads to cyanobacterial blooms, anoxic events and occasional fish kills during summer (Trindade et al., 2009). Hence, methanogenic processes play an important role in lake organic matter remineralization (Furlanetto et al., 2012; Palma-Silva et al., 2013). Further information on the lake's limnology has been published elsewhere (Albertoni et al., 2014; Duarte et al., 2020; Furlanetto et al., 2012; Marinho et al., 2009; Palma-Silva et al., 2008, 2013; Pereira et al., 2012a, b; Silva et al., 2015; Trindade et al., 2009).

2.2. The lake's history

The historical information reported herein was obtained from several university offices in the form of documents, architect blueprints, historical photographs and interviews. The first documented human activities of significant social importance occurred between 1923 and 1970 and corresponded to drinking water withdrawal from the wetlands by the Municipality of Rio Grande for the local population. In 1971, a 227-ha lot was awarded to the university to start building the Campus Carreiros facilities, where the lake is located, but the construction of the campus itself did not start until 1975.

A remarkable aspect of the strategic planning was the modification of the vast areas of marshes and other wetlands by filling flooded land using nearby dunes. Lake Biguás was deepened substantially in 1975 to create a semiartificial system to embellish the campus. Aerial photographs before and after 1975 depict the process of landscape accommodation and modification (Figure 2). The oceanographic station became the first facility to be inaugurated in 1978, ~400 m away from Lake Biguás, and the library and lecturing facilities were inaugurated immediately afterward in 1980. After 1981, the campus was expanded, and additional programs other than marine sciences were developed.

In 1990, the construction of the *Centro de Convivência* began. Aerial photographs before and after the construction of the facility, which featured lunch services, coffee shops, banking, information and other leisure activities, are shown in Figure 2. The construction process lasted approximately a decade, and the facility was inaugurated in 2000. Since then, the facility has been visited daily by thousands of people. At this point, the lake originally exhibited clear waters with restricted littoral occurrence of submerged macrophytes (Palma-Silva et al., 2008). This facility does not include an efficient sanitation system because the region does not have sanitation services. Instead, there is only a cesspit for bathroom and kitchen effluents that eventually leach into the lake, and there are two pipes that intentionally channel stormwater runoff into the lake.

Silva et al. (2015) provided a decadal limnological study documenting interannual shifts in shallow lake alternative states *sensu* Scheffer (1998). The lake exhibited variation in trophic conditions, including changes in the species composition and abundance of macrophytes, nutrient content and phytoplankton biomass, which were reflected in a series of interannual turbidity changes and increases in macrophyte volume (Table 1). Between 2000 and 2001, the lake exhibited high transparency and the development of submerged and floating-leafed macrophytes. At the beginning of 2002, the lake experienced severe eutrophication, with massive phytoplankton growth leading to high turbidity; in 2003, the floating macrophyte *Pistia stratiotes* proliferated to cover most of the lake surface. Harvesting activities were necessary to remove

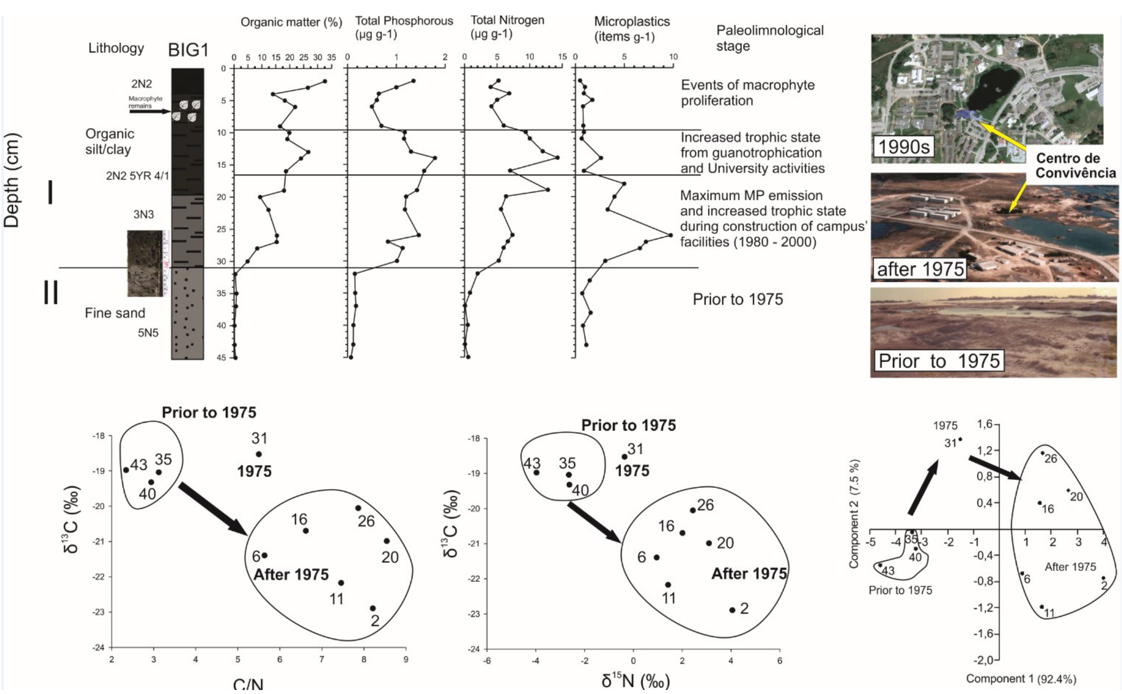


Figure 2. Lithology and vertical distribution of organic matter, total phosphorous, total nitrogen and MPs (upper four panels). Age is shown to the right of the plots, and photographs of the study area showing the environmental settings prior to 1975 AD and after 1975 are shown to the right. The lower panels show information on stable isotope data and C/N ratios and the PCA plot. The numbers next to each point indicate the core depth intervals in cm.

Table 1. Inter-annual changes in limnological conditions of Lake Biguás. Modified from Silva et al. (2015). *Nymphoides indica* is currently renamed *Nymphoides humboldtiana*, and *Potamogeton pectinatus* is currently renamed *Stuckenia pectinata*.

Time (yr AD)	Dominant macrophyte	Lake cover (%)	Chlorophyll-a (µg l^{-1})	Total Phosphorous (mg l^{-1})	Total Nitrogen (mg l^{-1})	Water phase
2000	*Nymphoides indica*	20	6.38 (±0.93)	0.32 (±0.04)	3.33 (±0.02)	clear
2001	*Nymphoides indica*	20	13.98 (±16)	0.15 (±0.07	0.50 (±0.36	clear
2002	*Nymphoides indica*	10	99.47 (±6.97)	0.14 (±0.06)	2.41 (±0.16)	clear/turbid
2003	*Pistia stratiotes*	10-95	52.23 (±45.11)	0.35 (±0.26)	1.40 (±1.01)	turbid/clear/ turbid
2004	*Nymphoides indica*	20	112.9 (±22.23)	0.24 (±0.06)	4.25 (±1.71)	turbid
2005	*Nymphoides indica Pistia stratiotes*	20	137.5 (±44.47)	0.4 (±0.04)	3.6 (±0.74)	turbid/clear/ turbid
2007	*Salvinia herzogii*	10	146.04 (±18.52)	0.64 (±0.40)	3.77 (±0.87)	turbid
2008	*Nymphoides indica Salvinia herzogii*	20	94.25 (±60.06)	0.27 (±0.09)	3.42 (±1.50)	turbid/clear
2009	*Potamogeton pectinatus Chara zeylanica*	95	8.95 (±2.75)	0.05 (±0.02)	1.42 (±0.09)	clear
2010	*Potamogeton pectinatus Chara zeylanica*	95	10.38 (±5.42)	0.04 (±0.02)	0.70 (±0.29)	clear

macrophytes, with a brief return to clear water conditions after harvesting. However, due to the increased nutrient supply, the lake returned to a turbid water phase dominated by phytoplankton. This situation lasted until mid-2005, with a brief period of clear waters in the winter, but soon afterward the lake returned to a turbid state until December 2008. After December 2008, the number of birds on the central islands of the lake decreased markedly due to the death of trees used as roosts, and the water conditions shifted from turbid to clear (2009 and 2010) with the massive proliferation of the submerged macrophytes *Stuckenia pectinata* (previously named *Potamogeton pectinatus*) and *Chara zeylanica*. After this period, the lake maintained the clear water phase until the end of 2021, after the construction of buildings near the margins, removal of littoral vegetation and perturbation of sediments. Albertoni et al. (2014) observed that this may have favored the release of sediment nutrients and enhanced the growth of the macrophyte *Salvinia herzogii*, covering the entire surface during 2013 and the following two years, which suppressed the submerged macrophytes. Duarte et al. (2020) mentioned the removal of floating vegetation in July 2015 and the return to the clear water state with submerged macrophytes growing.

2.3. Sampling and laboratory analysis

A 45-cm-long sediment core (BIG 1, Figure 2) was taken from Lake Biguás in April 2019 with a 7-cm diameter corer. The core was immediately opened, lithologically described according to sediment texture and color using a Munsell chart, and samples were selected for geochemistry, MPs and diatom analyses. The Munsell color system, designates colors based on an arrangement scheme first developed in 1913. The colors are defined by measured scales of hue, value, and chrome, which correspond respectively to dominant wavelength, brightness, and strength or purity. The system is internationally used in geosciences for reporting sediment color codes.

The sediment core was dated by relative techniques. We compiled very detailed historical information on the history of the study area, and identified the lithological transition from sandy to silty sediment, corresponding to lake origin in 1975 after excavation operations, and used the top/bottom approach by identifying ulterior changes in MPs distribution and geochemistry to infer paleolimnologcial stages.

Organic matter was determined by weight loss on ignition (LOI) at 550 °C for 2 h (Heiri et al., 2001). For total nitrogen and phosphorus, approximately 2-g sediment samples were dried in an oven at 50 °C for 48 h. Total nitrogen (TN) was determined by the Kjeldahl method according to Allen et al. (1974), and total phosphorus (TP) was measured using the method of Fassbender (1973).

Carbon ($^{13}C/^{12}C$, referred as $\delta^{13}C$) and nitrogen ($^{15}N/^{14}N$, or $\delta^{15}N$) isotope ratios were analyzed simultaneously at the Integrated Analysis Center (CIA-FURG), following Faria et al. (2018). Samples were freeze-dried, ground and homogenized, and approximately 1 mg of sediment was placed into tin capsules. An isotope-ratio mass spectrometer coupled to an elemental analyzer was used for the stable isotope analysis of carbon and nitrogen. Values are provided in delta notation (δ), expressed in ‰ following Bond & Hobson (2012).

For evaluation of MPs, a volume of 5 cm^3 of sediment with variable mass was transferred into 15-ml tubes and dried at 40 °C, weighed with an analytical balance and transferred into beakers containing saline solution (1.2 g cm^{-3} NaCl-Coralife), and MPs were isolated by flotation and then filtered (Hidalgo-Ruz et al., 2012; Pinheiro et al., 2019). Each filter was transferred to a Petri dish, which was closed and stored at 40 °C before analysis under a stereomicroscope (Olympus SZX9) with an attached camera protected by a chapel. Evaluation of filters and blanks consisted of a total scan of each entire filter and identification (fiber or fragments), measurement (length and area) and color of each MP (Masura et al., 2015). Plastisphere microbial biofilm samples were fixed with 1% sterile glutaraldehyde and heated at 40 °C. The MPs were transferred into plastic vials and then covered with gold film for SEM using a JEOL JSM - 6610LV (Agostini et al., 2020). The community of plastisphere was classified into bacteria, fungi and microalgae. Microplastic characteristics (amount, fiber, fragments, length, area, and color) were ordinated using canonical correspondence analysis (CCA) to identify differences in composition among the sediment layers and surface characteristic preferences for plastisphere community occurrence.

Diatom samples were treated with 35% HCl and rinsed with distilled water. Next, 30% H_2O_2 was added to eliminate organic matter, and samples were rinsed with distilled water. Permanent slides were mounted in Entellan®. A minimum of 500 valves were counted at 1000× magnification and identified according to Metzeltin & García-Rodríguez (2003) and Metzeltin et al. (2005). Diatom association zones were identified by cluster analysis using the Morisita index as advised by Hammer et al. (2001) to constrain similarity to contiguous intervals, which was performed for the whole 45-cm-long sediment core for 23 selected sediment intervals. The changes in isotopic composition in $\delta^{13}C$ and $\delta^{15}N$ were evaluated by Principal Component Analysis (PCA). All statistical tests were performed with PAST version 2.17c (Hammer et al., 2001).

3. Results

The lithological analysis identified two stratigraphic units: Unit II (45–31 cm, Figure 2) consisted of light gray (Munsell code 5N5) fine sand sediments; Unit I encompassed the uppermost 31 cm and consisted of organic clay, which was subdivided into three sections related to a darkening pattern in sediment color from bottom to top (Figure 2). Accordingly, the 31–20 cm layer corresponded to the Munsell code 3N3, the 20–4 cm layer to the code 2N2 5YR 4/1, and the top 4 cm to 2N2. The distinct shift in sediment type observed at 31 cm depth was attributed to the lake excavation in 1975. The rationale for this is that the system was originally a natural wetland, which was artificially transformed into a shallow lake by excavating it to make it deeper. Therefore, there was a transition from a wetland to a lake sedimentation system, and sedimentological expression is evident in the photograph presented to the left of the sediment core plot in Figure 2. Photographs confirm excavation in 1975. Because no substantial change in lake morphometry or watershed geomorphology was documented after 1975, a mean historical sedimentation rate of 7 mm yr^{-1} was calculated. Although sedimentation is a variable process depending on environmental factors such as rainfall and wind, regional shallow lakes exhibit historical sedimentation values ranging between 4 and 10 mm yr^{-1}, and the most frequent contemporary historical value is 5 mm yr^{-1} (Azcune et al., 2020; Bueno et al., 2019; 2021; García-Rodríguez et al., 2002).

The geochemical proxies for trophic changes showed similar trends (Figure 2), with the lowest values found in lithological Unit II prior to 1975. In this sense, organic matter increased from 1 to 10%, TP increased from 0.2 to 1 μg g^{-1} and TN increased from 1 to 5 μg g^{-1}. Such an increasing trend in geochemical variables was also supported by changes in isotopic composition of the sedimentary organic matter. The isotopic shift from bottom to top demonstrated a marked decrease in $\delta^{13}C$ values and increases in C/N ratios and $\delta^{15}N$ values from before to after the urbanization around the lake (Figure 2) as indicated by the PCA analysis where the axis 1 gradient accounted for 92.4% of the variance, thus suggesting a high input of organic matter of anthropogenic origin. The continuing increase in organic matter, nitrogen and phosphorous reached maximum values between 16 and 20 cm (i.e., by 1990 and 1996) and then decreased to the 5-cm depth layer, after which there was a second increase in values of geochemical variables near the surface (Figure 2).

The vertical distribution of MPs indicates that the lake appears to be historically subject to MPs contamination, with items identified throughout the sediment core. Minimum levels of MPs were observed within the basal section of the core and peaked (10 items g^{-1}) within the middle section of the core, mainly consisting of fibers. Most MPs found in the sediment layers were fibers as opposed to fragments (Figure 2 and 3A). Fibers were recorded in all sediment layers, while fragments were more abundant in sediments dating from 1975 to 2000 (Figure 1). The most common colors for fibers were black, transparent and orange, which were associated with smaller MPs sizes (<0.0001 mm). On the other hand, fragments recorded were more commonly associated with red and white colors and larger sizes (>0.0002–0.001 mm) (Figure 3A). Fragments were more suitable than fibers for microbial biofilm association, including colonization by bacteria, fungi and diatoms. Both MPs area and color influenced plastisphere occurrence. Bacteria and fungi (stages 1 and 2) were more associated with black and white colors, while diatoms (stage 3) were more associated with transparent and green surfaces. In addition, microbial biofilms on MPs were also recorded in all sediment layers; however, the upper layers (after 1996) showed more intense colonization (Figure 3B).

A total of 14 dominant diatom species were identified throughout the sediment core, with the dominance of benthic pennate over planktonic centric forms commonly observed for shallow lakes (Metzeltin & García-Rodríguez, 2003). The five species of the genus *Gomphonema* (i.e., *G. acuminatum*, *G. laticolum*, *G. gracile*, *G. parvulum* and *G. affinopsis*) were pooled in Figure 3, as these five species coexist under similar environmental conditions (Metzeltin et al., 2005). Similarly, *Pinnularia latevittata* and *P. gibba* and *Epithemia adnata* and *E. sorex* were pooled into their respective genera (Figure 4). Cluster analysis allowed the identification of four diatom association zones (DAZs). DAZ1 was dominated by benthic forms of *Navicula* spp., *Nitzschia* spp. and *Encyomena* spp., together with *Discostella stelligera*. DAZ2 exhibited codominance of *Staurosira longirostris*, *Pseudostaurosira neoelliptica* and *P. trainorii* (Figure 4), which together accounted for approximately 90% of diatoms throughout this DAZ. The dominant diatom species reported

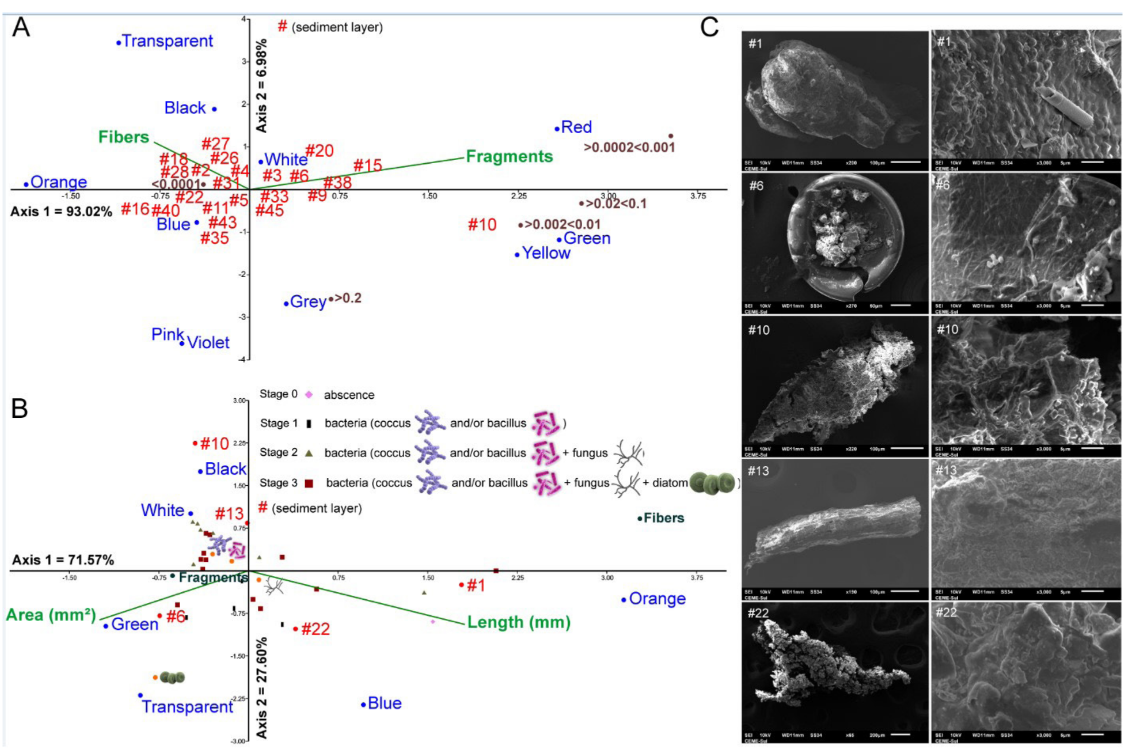


Figure 3. CCA ordination diagram showing MP composition. (A) Microbial biofilm association (plastisphere) regarding MP surface characteristics. (B) MEB plastisphere images for different layers (# cm) (65x and 270x magnification) and the biofilm (3000x magnification) (C) #1 left: coccus bacteria, #1 right: *Aulacoseira granulata* diatom, #6: coccus and bacillus bacteria, fungi and unidentified biological structure, #13: coccus bacteria, #22: coccus bacteria.

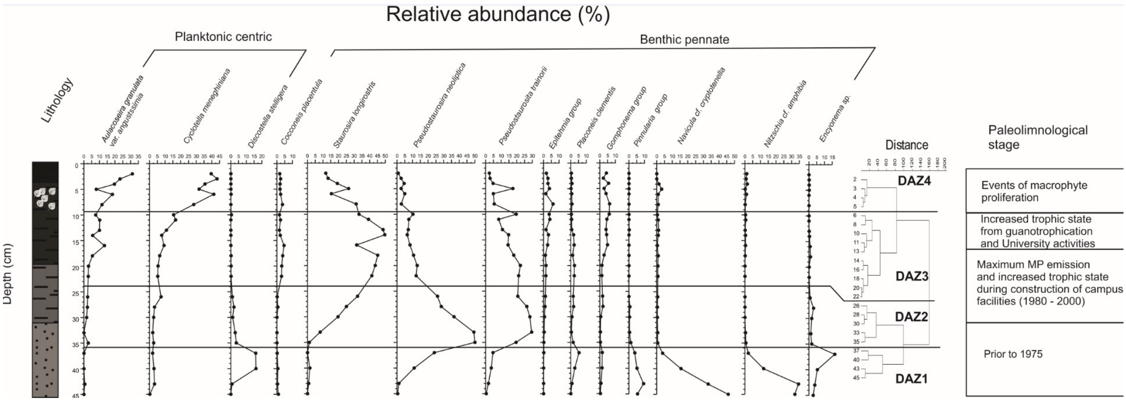


Figure 4. Relative abundances of dominant diatom species. Age and cluster analysis showing the identified diatom association zones (DAZ) are shown to the right of the plot.

for DAZ2 were also observed in DAZ3, but the difference in species composition was associated with an increase in *Gomphonema* and *Epithemia* groups instead of a progressive increase in planktonic forms *Aulacoseira granulata* var. *angustissima* and *Cyclotella meneghiniana*. Finally, DAZ4 was codominated by planktonic forms *A. granulata* var. *angustissima* and *C. meneghiniana*, and there was a progressive decrease in the codominant benthic species reported for DAZ3.

4. Discussion

4.1. Lake paleolimnology and historical limnology

As expected, prior to campus construction, the core lithology shows a Holocene deposit dominated by sandy sediments (see Figure 1, aerial photograph from 1920) of allogenic origin due to successive sea level changes (Dillemburg et al., 2107).The pre-impact landscape was dominated by a series of marshes and other wetlands, in which Lake Biguás represented the largest natural wetland and was excavated and deepened, but the shorelines were fixed only after 1975. Therefore, we have assigned this chronology to the lithological transition observed at 31 cm depth (Figure 2), and this stage was identified as the first paleolimnological stage accordingly. Notwithstanding, the sandy sediments prior to the excavation, hold evidence of contamination, as indicated by the occurrence of MPs within the basal 15 cm of the sequence, so the sediment core does not span preindustrial times. Instead, the system was under the influence of long-distance diffuse MP contamination, probably from industrial and/or urban activities of the city of Rio Grande (Agostini et al., 2018; Quintana & Mirlean, 2019), perhaps with plastics that were transported by wind and then fragmented and deposited. The construction of the university beginning in the early 1970s represented an evident source of MP contamination, as maximum levels of MP items were recorded during this time (Figure 2) and was identified as the second paleolimnological stage. Once the construction of most facilities was completed, MP contamination decreased. Therefore, this is an important record of historical MP contamination, corroborating their use as a sensitive proxy for anthropogenic contamination within the study area. MPs are durable at human time scales (Corcoran et al., 2015), and their longevity in the environment supports their use as an Anthropocene stratigraphic marker (Turner et al., 2019). However, the total number of MPs found could have been even greater if the organic matter digestion method had not been performed (Isobea et al., 2019), so the magnitude of MP contamination was probably underestimated. The adhesion of organisms to MPs increases their density (Lobelle & Cunliffe, 2011; Zettler et al., 2013), although no report is available to our knowledge on the long-term maintenance of biofilms after burial. In the current study, we demonstrate that the plastisphere can be recorded even in deep layers of sediment, representing a decadal scale of variability. In addition, we observed a biofilm preference for MP characteristics such as type (fragment), area (>0.0002 mm^2) and color (black, white, green, transparent), corroborating previous studies in the region on biofilm formation on other kinds of materials such as steel, glass and wood (Agostini et al., 2018).

All geochemical proxies for the initial stages prior to lake formation indicate mesotrophic conditions, with organic composition comparable to that observed by Patterson (2016) in the nearby Patos Lagoon. Patterson (2016) inferred that the organic matter composition exhibited a signal of microalgal origin, which, according to the diatom diagram (Figure 3), corresponded to microbenthic forms. After the formation of the lake, a distinct intensification of eutrophication was initiated, as clearly inferred from the increases in organic matter and nutrients but also from the increases in $\delta^{15}N$ values (Smucker et al., 2018) and C/N ratios (Figure 3), as previously shown, for example, in Lake Geneva, Switzerland, in sediments contaminated by urban effluents (Gascón Díez et al., 2017). In this way, organic matter composition showed a clear difference before and after 1975, with the most plausible explanation being human-induced environmental change coupled with guanotrophication. This is especially noticeable within the top 20 cm (i.e., after 1990) because of the cumulative effect of landscape transformation around the lake and the activities of the fully operational facilities adjacent to the lake (Figure 2). This intensification process of the eutrophication was identified as the third paleolimnological stage. In this regard, the facilities do not have proper sewage systems, and there is underground leaching into the lake. In addition, there are at least two pipes that intentionally channel stormwater runoff into the lake, in addition to plastic waste deposition. Therefore, facilities built next to the lake could be identified as a key historical human impact, which led to a significant and continued deterioration of the environmental quality of the system, especially after the end of the construction process and the beginning of catering service operations.

After 2000 and 2001, the lake exhibited a clear water phase with a relatively low chlorophyll concentration, when macrophyte cover reached 20%, which expression in the sedimentary profile is resembled by the high content of macrophyte remains at 10 cm depth. This explains the decrease in nutrient concentration in this section of the sedimentary profile, as during macrophyte development, phytoplankton biomass is depleted (Meerhoff & Jeppesen, 2009; Scheffer, 1998; Scheffer & van Nes, 2007) and less organic particulate material is deposited in the sedimentary environment accordingly. This was the beginning of the documented process of recurrent events of macrophyte proliferation (Table 1) and was identified as the fourth limnological stage (Figure 2). Thus, in 2002, conditions shifted to a succession of water phases from clear to turbid to clear, with chlorophyll concentrations reaching values significantly higher than those in previous years. This condition worsened within the next few years (until 2008), with chlorophyll values reaching one order of magnitude higher than all historical values, due to cyanobacterial blooms. Additionally, planktonic diatoms reached their maximum historical abundances in the sedimentary record (Figure 3). The reasons for this historical eutrophication process include increases in both nitrogen and phosphorus caused by the natural supply of bird droppings together with the anthropogenic construction and operation of campus facilities around the lake. The transition between alternative states in shallow lakes proposed by Scheffer (1998) occurs differently in tropical and subtropical aquatic systems, where floating plants together with high concentrations of nutrients and generally high water column stability occur (Meerhoff & Jeppesen, 2009; Scheffer & van Nes, 2007). When the water level is low, opportunistic macrophyte proliferation is observed, and in extreme cases, the whole lake can be invaded, as observed for Lake Biguás from 2009–2010 (Table 1) and 2013–2014 (see Figure 1D-F). Similar processes of shallow lake massive macrophyte proliferation have been reported elsewhere (García-Rodríguez et al., 2002; Inda et al., 2016). Eutrophication and its consequences, e.g., massive macrophyte growth or phytoplankton blooms, are synergistic processes that reflect organic matter accumulation in the sediment, as demonstrated in the studied profile. The accumulation of organic matter in the sediment promotes a feedback effect, increasing the concentrations of nutrients in the water column and facilitating the growth of primary producers. This phenomenon is common in eutrophic lakes and known as internal fertilization (Schauser & Chorus, 2007) and promotes resuspension of refractory material (uric acid) contained in bird feces (Adhurya et al., 2020).

The role of cormorants and other waterbirds in eutrophication at Lake Biguás seems limited to certain periods, only when cormorants are roosting on its islands. Because cormorants feed on estuarine fish from Patos Lagoon (Barquete et al., 2008a) and piscivorous herons feeding on the Patos Lagoon estuary have higher $\delta^{13}C$ values than limnetic birds (Britto & Bugoni, 2015; Faria et al., 2016), the decrease in $\delta^{13}C$ values could be more parsimoniously attributed to cultural eutrophication than to guanotrophication. Likewise, the increase in $\delta^{15}N$ is also in agreement with this inference (Britto & Bugoni, 2015; Faria et al., 2016). The limited influence of birds in freshwater environments seems to occur frequently when drainage sources are important (Adhurya et al., 2020). Therefore, the continued cultural eutrophication process to which this lake is subject led to organic contamination resulting in blooms of cyanobacteria, alternating with massive macrophyte proliferation. Thus, an increased interannual limnological instability leading to critical transitions in alternative states is also identified as a symptom of lake environmental quality deterioration. Moreover, after the macrophyte invasion of 2013–2014, the system shifted to a state of greater turbidity. From 2014–2015, another massive floating macrophyte growth occurred, and management for removal was carried out (Duarte et al., 2020). According to the authors, the central islands were permanently occupied by birds, and due to the continuity of nutrient input, a new turbid water state occurred, with extensive growth of phytoplankton. Data from routine monitoring of Lake Biguás showed values of 530 $\mu g\ L^{-1}$ and 184 $\mu g\ L^{-1}$ during 2018 and, in August 2019, 944.9 and 456.8 $\mu g\ L^{-1}$ TP and chlorophyll-*a*, respectively (Duarte et al., 2020). These values indicated ecosystem hypertrophy. In 2020, cyanobacterial blooms were noticeable, even by visual inspection.

4.2. Paleolimnology, limnology and environmental management

We demonstrated the long-term degradation caused by cumulative natural and human impacts. Not surprisingly, none of the published papers on the limnology of Lake Biguás (Furlanetto et al., 2012; Marinho et al., 2009; Palma-Silva et al., 2013; Silva et al., 2015; Trindade et al., 2009) interpreted the hypertrophication process as a consequence of human impacts, which seems clear from the multiproxy paleolimnological data presented here, particularly soon after the 1990s. Instead, they exclusively attributed the intensified eutrophication process and limnological instability to bird guanotrophication (e.g., Silva et al., 2015). Consequently, the negative human impacts were accordingly neglected, given the lack of pre-disturbance limnological data available prior to 2004 (Trindade et al., 2009). It is under conditions of lack of long-term information that paleolimnological approaches provide the long-term historical perspective to improve environmental diagnostics and identify the best possible management measures. From this perspective, and based on the paleolimnological data introduced in this paper, together with previous limnological data, we have identified a number of general lake management measures for attempting to slow the hypertrophication process and minimize lake transitions between alternative states, *sensu* Scheffer (1998). The general measures involve meeting with university authorities to introduce this investigation, show them the potential of the combined use paleolimnological and limnological research. In this regard, we emphasize and agree with the implementation of the ongoing university project for designing and constructing an additional new cesspit or connection with the sewage collection system and treatment station to be built within the university campus. Given that the littoral zone around the lake has abundant litter, several days of cleanup should be necessary, with the involvement of students, employees and owners of the businesses operating at the facilities. We also encourage the continued limnological surveys to monitor lake trophic state and macrophyte cover as input for adaptive management. Since Biguás is a small lake, we recommend deploying permanent freshwater advanced aquatic sensors for nutrients, chlorophyll and turbidity.

5. Conclusion

The long-term science perspective introduces a different and complementary understanding of the eutrophication process relative to that exclusively inferred from direct observational limnological data. Limnology had previously identified guanotrophication via fecal nutrient content from the population of cormorants as the main cause of organic enrichment. However, the paleolimnological perspective indicates that lake eutrophication started to intensify itself after 1990 AD due to human impacts. In addition, pollution by MPs, potentially perceived as recent, was demonstrated to be present long ago in lacustrine sediments. Therefore, we identify the combined effects of natural and cultural impacts as triggers for environmental changes. The lake management measures proposed for rehabilitation must be accompanied by social actions aiming to sensitize stakeholders of many different social and ethnographic origins.

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