

Limnological characteristics and trophic state of Paso de las Piedras Reservoir: An inland reservoir in Argentina

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Abstract

The current trophic status of Paso de las Piedras Reservoir was evaluated by analysing different physicochemical, biological and environmental variables, in relation to its water quality, and application of two different classification criteria. Water quality sampling was conducted at four sampling sites from June 2004 to June 2005. During this study, 183 phytoplankton taxa were identified. The phytoplankton abundance exhibited a maximum of 368.037×10^3 cells.mL⁻¹ in February 2005, and at least 1.133×10^3 cells.mL⁻¹ in October 2004. An almost exclusive dominance of Cyanobacteria was observed between December 2004 and May 2005, the product of a large relative abundance of *Anabaena circinalis* and *Microcystis natans* which, together with other companions, were the reason for an algal bloom characterized by an average density of 133.05×10^3 cells.mL⁻¹, and an average chlorophyll concentration of 28.7 mg.m⁻³. These study results indicate that the seasonal variations of physical, chemical and biological parameters in the waters of this reservoir were essentially a consequence of environmental and hydrological conditions in the dam area. In contrast, the spatial variations inside the lake were the result of the characteristics of the water inflow provided by its two main tributaries. The N:P ratio suggests neither nutrient is a limiting factor for maximum algal biomass in the lake, indicating that variations in the phytoplankton community structure, and development of phytoplankton blooms, would be more constrained by environmental and hydrological conditions than nutrient competition. The high concentration of measured nutrients could be attributed to the concurrence of various non-point sources. The phytoplankton species richness was high, exhibiting values even higher than those mentioned in previous studies. Considering the two trophic classification systems, and based on total phosphorus data, the reservoir is classified within the hypertrophic category. In contrast, considering only the chlorophyll and turbidity data, the lake would be classified within the eutrophic category.

Key words

Paso de las Piedras Reservoir, phytoplankton community, trophic state, water quality.

INTRODUCTION

Eutrophication is one of the most ubiquitous environmental problems in inland waters, primarily as a result of anthropogenically driven enrichment with two nutrients, phosphorus and nitrogen (UNEP-IETC/ILEC 2001). In the middle of the twentieth century, eutrophication was recognized as a serious pollution problem for many western European and North American lakes and reservoirs, becoming widespread since that time. About 54% of the lakes in Asia, for example, are eutrophic, while

the same is true for 53% of the lakes in Europe, 48% of the lakes in North America, 41% of the lakes in South America and 28% of the lakes in Africa, respectively (ILEC/Lake Biwa Research Institute 1988–1993). Eutrophication can cause changes in the taxonomical structure of aquatic ecosystems, significantly reducing biodiversity by increasing the abundance of some species, particularly cyanobacteria, which have the capacity to form surficial blooms. Increases in total algal biomass can have strong impacts on their periodicities (Smith 1990), with this pattern having been documented for boreal, temperate, subtropical and tropical ecosystems (Havens 2008).

Several indicators, indexes and models have been developed to assess eutrophication and water quality,

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most being based on chemical, physical and biological parameters (e.g. dissolved oxygen, nutrient and/or chlorophyll-*a* concentrations, turbidity, specific composition of phytoplankton) (Carlson 1977; Lampert & Sommer 1997). According to Margalef (1983), because organisms can be precise sensors of the properties of their surrounding environment, the structure of phytoplanktonic communities can be used as good indicator of water quality changes. As urban and agricultural developments continuously increase, concern about the sustainability of the quality of water supplies also increases. Thus, monitoring changes in the hydrology and water quality of reservoirs and basins is critical for properly managing these resources for the long term.

In Argentina, the number of lakes and reservoirs experiencing eutrophication appears to be increasing. Paso de las Piedras Reservoir is the unique water supply source for around 400 000 inhabitants of the cities of Bahía Blanca and Punta Alta, in the south of Buenos Aires Province, Argentina. It also provides raw water for industrial purposes, as well as being used for recreation and fishing. The reservoir has experienced recurrent cyanobacterial blooms during the summer months since 1982 (Gayoso 1993; Pizzolon *et al.* 1999; Echenique *et al.* 2001; Parodi *et al.* 2005).

There is little knowledge about the ecological changes in the reservoir since its creation in 1978, or about the anthropogenic pressures being exerted on it. Intartaglia and Sala characterized the reservoir in 1989 as a non-stratifying, eutrophic lake. Other studies were limited to taxonomical descriptions of phytoplankton, especially diatoms (Sala & Intartaglia 1985; Sala 1990a, 1990b, 1996a, 1996b, 1997; Fernández & Parodi 2005), or to brief reports on massive algal bloom episodes (Gayoso 1993; Guerrero & Echenique 1997; Echenique *et al.* 2001).

The main objective of this study was to assess the current trophic status of the lake by describing different physicochemical, biological and environmental variables, in relation to the reservoir water quality. The goal is to determine which of these variables is most significant in regard to the trophic status of this waterbody. Accordingly, this study incorporates of the weather conditions prevailing in the study area, the physical and chemical properties of the water and their seasonal variations, with special reference to the reservoir's different tributaries, and the attributes of the phytoplankton community and its seasonal variations and spatial relationships with physicochemical and environmental parameters. Using this information, the trophic status of the reservoir was assessed by applying different classification criteria and indexes and, in turn, analysing the differences between

them, and their particular incidence as factors influencing the reservoir's water quality.

METHODS

Study area

Paso de las Piedras Reservoir (38–39°S, 61–62°W) is located in the southern part of Buenos Aires Province (Fig. 1). It was created in 1978 by damming the Sauce Grande River at its confluence with El Divisorio Stream. The reservoir covers an area of 36 km², and has a maximum depth of 28 m (Table 1). It is located in a semiarid temperate region, with an average temperature in the coldest month (July) of 7°C, and 23.5°C in the hottest month (January). The average rainfall is between 650–950 mm. The winds blow primarily from the north/northwest, with greater intensity in the spring and summer (Vouilloud *et al.* 2005).

Land use in the catchment area includes cattle grazing on native and improved pastures, and cultivation of wheat, oats, sunflower, sorghum, crop and soya bean (Aduriz *et al.* 2003). Monoamonic and diamonic phosphates and urea are the most frequently used fertilizers in the catchment area (UNS-ABSA-ORAB-ADA 2004).

Sampling and sample processing

The lake was sampled from June 2004 to June 2005. The samples were collected weekly or biweekly, depending on constituent, between 0800–1100 hours. Four sampling sites were established, including S1 (in water intake tower of purifying plant); S2 (near mouth of Sauce Grande River); S3 (near mouth of El Divisorio Creek); and S4 (in transition area between tail of the reservoir and dam) (Fig. 1).

Samples for qualitative analyses of phytoplankton were taken subsuperficially with a 30- μ m mesh plankton net. Some samples were maintained alive, while others were fixed with 4% formaldehyde. Samples for quantitative analyses were collected with a Van Dorn bottled, and fixed with Lugol's solution. The Utermöhl (1958) method was used to quantify the microalgae.

In addition to water sample collection, *in situ* measurements also were conducted on selected chemical and physical characteristics, including electrical conductivity, temperature, pH and water transparency, utilizing a Horiba U10 multisensor (HORIBA Ltd. Miyano Higashi, Kisshoin, Minami-Ku, Kyoto, Japan) and 40-cm diameter Secchi disk, respectively.

Water samples for nutrient and chlorophyll analysis were collected with a Van Dorn bottle, stored in darkness at 4°C, and processed within 24–48 h after being collected.

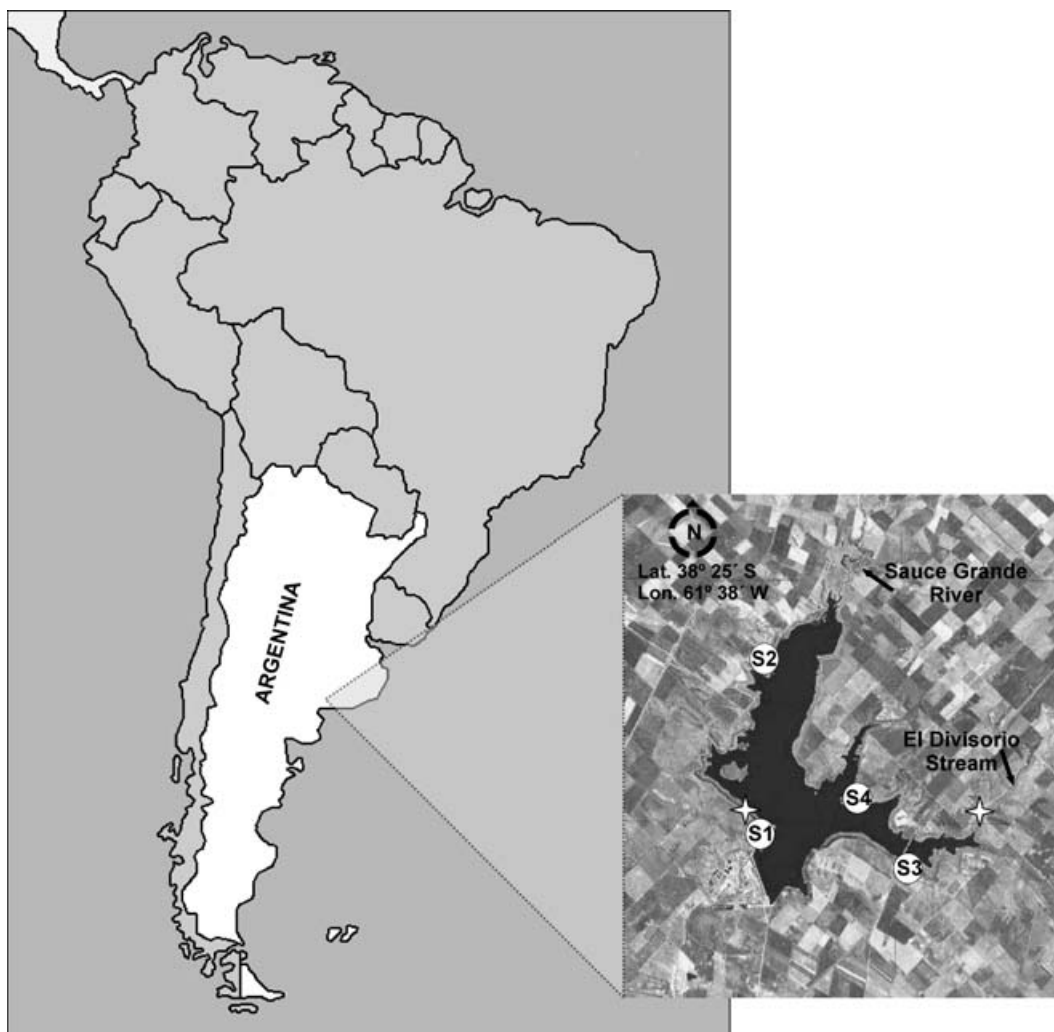


Fig. 1. Location of Paso de las Piedras Reservoir and sampling sites (S1, S2, S3, S4; ✦ indicates location of meteorological stations).

Table 1. Principal morphometric and hydrological characteristics of Paso de las Piedras Reservoir (from Schefer 2004)

Watershed area (km ²)	1620
Shoreline length (km)	60
Surface area (km ²)	36
Maximum depth (m)	28
Mean depth (m)	8.2
Volume (hm ³)	328
Hydraulic retention time (years)	4
Maximum level of dam (m)	165

Nitrate (NO₃⁻), nitrite (NO₂⁻), ammonium, total phosphorus (TP), reactive soluble phosphorus (RSP) and silica were analysed in the Autoridad del Agua (ADA) Laboratory of the Buenos Aires Province, following the methods described in APHA (1992). Total suspended solids (TSS) concentrations also were made in accordance

with the method outlined in APHA (1992) by PROFERTIL. The chlorophyll concentration was measured in accordance with the spectrophotometric technique described by Lorenzen (1967), after being extracted in a 90% acetone solution.

Two meteorological stations located in the vicinity of the mouth of El Divisorio Creek and in the lake area were used to obtain weather data (air temperature, wind speed and direction, precipitation; Fig. 1). The Laboratory of Hydraulic at the Universidad Nacional del Sur provided these data, along with data on lake volume and level, and tributary flows.

Data processing and statistical analysis

The phytoplankton community in each sampling site was analysed in terms of its taxonomic composition (i.e. species richness, abundance and specific diversity (H') using the Shannon index).

The trophic status of the reservoir was ascertained with application of the 'open limit' classification scheme

developed by the OECD (1982), and the trophic status index (TSI) of Carlson (1977). The latter index is based on (i) the Secchi disk transparency (TSI (SD)); (ii) chlorophyll-*a* concentration (TSI (Chl *a*)); and (iii) total phosphorus concentration (TSI (TP)), calculated according to the following formulae:

$$TSI (SD): 60 - 14.41 [\ln (SD)];$$

$$TSI (Chl a): 9.81 [\ln (Chla)] + 30.6;$$

$$TSI (TP): 14.42 [\ln (TP)] + 4.15$$

The Carlson TSI is based on a gradual scale from 0 to 100. A TSI of <30 is commonly considered indicative of oligotrophic conditions; an index ranging between 50 and 70 is considered indicative of eutrophic conditions; and values >70 are commonly considered representative of hypereutrophic conditions (Wetzel 2001).

Box plots were used to compare the values of the TSIs. The non-parametric Kruskal–Wallis test was used to assess differences between the TSI values. If any statistical differences among values were detected, pairwise multiple comparison Dunn's test was applied to distinguish differences between pairs of values.

A principal component analysis (PCA) was performed on the environmental data to determine which variables were correlated, and to summarize environmental characteristics of sampling sites in an ordination diagram. Data were first centred and standardized, once the variables had different units. To determine how phytoplankton was associated with the studied environmental variables, canonical correspondence analysis (CCA) was applied to the data. Phytoplankton data were presented as the density of the main taxonomic classes, after logarithmic transformations. The software used for this analysis was XLSTAT2008 (trial version; Addinsoft, New York, USA).

RESULTS

Environmental conditions in dam area

Total precipitation of 598 mm was recorded in Paso de las Piedras Reservoir during the study period. The greatest rainfall was recorded during December 2004, totalling 168.8 mm for that month. The lowest average precipitation was recorded for the months of May 2005 and June 2004 and 2005. The months exhibiting the greatest number of raining days (13, 12 and 11, respectively) were October, July and December 2004 (Fig. 2). The precipitation was > 30 mm per day only in December. The directions of prevailing winds were north and northwest, with daily average wind velocities ranging between 0.2 and 30 km.h⁻¹, with gusts of up to 77 km.h⁻¹.

The lake water level was mostly influenced by the water input (rainfall run-off). At the beginning of the study, the

reservoir water level was 162.5 m, equivalent to a water volume of 251 hm³. The water level then decreased to the minimum value recorded during this study, being 162.2 m during mid-July 2004, probably due to low precipitation during this period. As a result of an increasing flow of El Divisorio Creek, and other tributaries of the River Sauce Grande, caused by several days of intermediate intensity rainfall, however, the lake level of the lake started to rise in late July, 2004, and remained >163.4 m until mid-March 2005. The peak level of 164.45 m was reached during the last days of December 2004, resulting from heavy rains during the month that caused both the Sauce Grande River and the El Divisorio Creek to flood (Fig. 2).

The air temperature presented a typical seasonal cycle, with a maximum of 27.8°C in December, and a minimum of 1.4°C in June (Fig. 2).

Physical and chemical water quality parameters

The reservoir water temperature oscillated between 23.55°C (January 2005) and 6.38°C (July 2004), being similar at all four sampling sites (Fig. 2).

The pH exhibited a similar behaviour for the four sampling sites, with values ranging between 7 and 9.75 (Fig. 2). The recorded pH values were lower at the beginning of the study, increasing by the end of August. Since that period, the pH values fluctuated around the higher values, except for two decreases on 25 October and 20 December, which correlated with decreases in the Secchi depth. The first decrease in the pH coincided with a small increase in the number of diatoms, while the second decrease corresponded to the beginning of the period of higher lake productivity.

The TSS concentrations ranged between 1.2–18.4 mg.L⁻¹. The highest concentrations were recorded between August and November 2004, with a maximum value in mid-September 2004 (Fig. 2). Overall increases in the TSS concentration coincided with periods of high wind intensity and changes in wind direction.

The water transparency, as expressed by Secchi depth values, ranged between a maximum of 2.2 m in November 2004 at site S1, and a minimum of 1.08 m in September 2004 at site S3 (Fig. 2). There were two periods of low visibility at site S1, the first between late December 2004 and February 2005, a period that coincided with major phytoplankton growth. The second period occurred during August and September 2004. This was a period during which, even though the total number of phytoplankton cells was low, there was a predominance of diatom species that, with a larger biovolume, more strongly affected the water transparency. Variations in

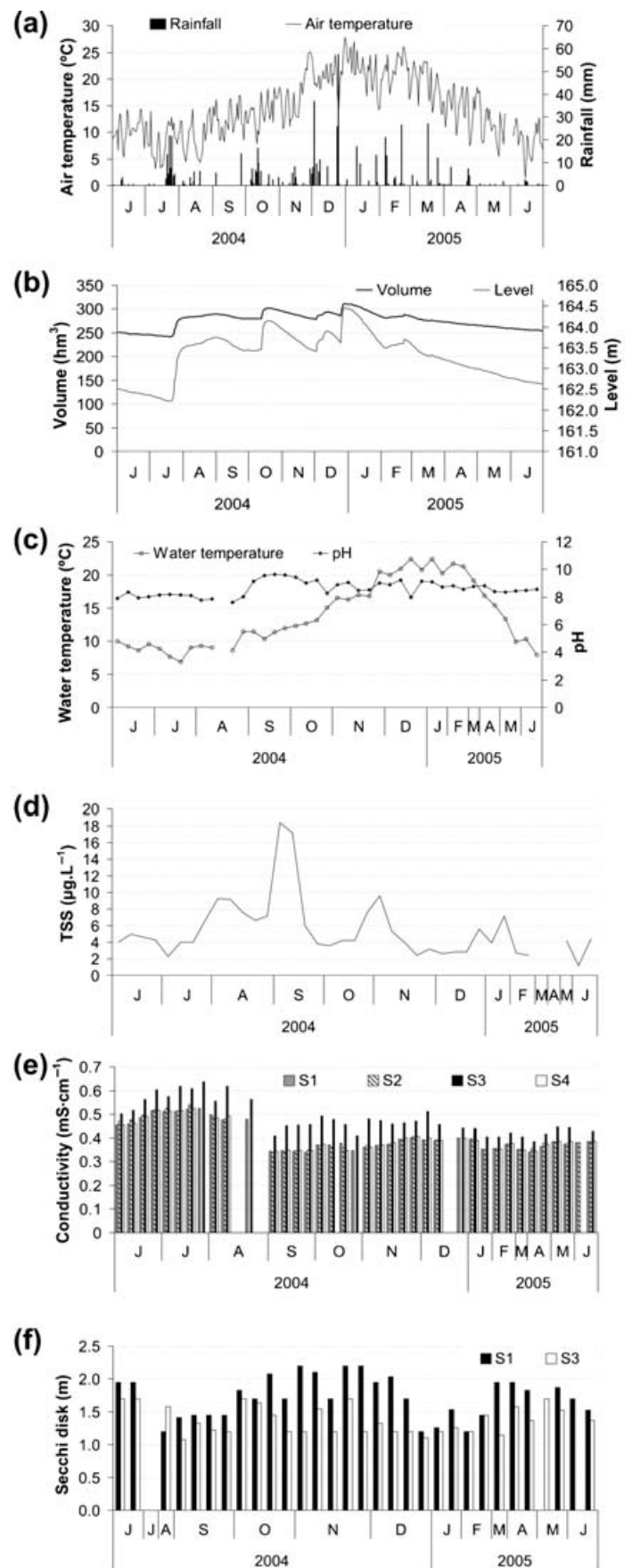


Fig. 2. Daily variation of weather and hydraulic conditions: (a) rainfall and air temperature; (b) lake volume and level of Paso de las Piedras Reservoir, and variation of physical and chemical water parameters: (c) temperature and pH; (d) total suspended solids (TSS); (e) electrical conductivity; (f) Secchi depth (in regard to water temperature, pH and TSS, the figures show the average of the four sampling sites, while the sites are shown separately for conductivity and Secchi depth).

water transparency were less marked at site S3, which exhibited lower, more stable values throughout the year. The low water transparency measured at both sampling sites in early September 2004 also coincided with a sharp increase in the TSS concentration values.

The electrical conductivity ranged from $0.54 \mu\text{S}\cdot\text{cm}^{-1}$ in July 2004 at site S4, to $0.34 \mu\text{S}\cdot\text{cm}^{-1}$ in September 2004 at site S1. Site S3 exhibited higher electrical conductivity values, compared to the other monitoring sites (Fig. 2).

Nitrates were the most important nitrogen compounds, with concentrations ranging from $2.10 \text{ mg}\cdot\text{L}^{-1}$ in late August 2004 at site S1, to $0.8 \text{ mg}\cdot\text{L}^{-1}$ in May 2004 at site S4 (Fig. 3). Nitrites always exhibited concentrations $<0.04 \text{ mg}\cdot\text{L}^{-1}$, with the exception being June 2004, when they reached a maximal concentration of $0.078 \text{ mg}\cdot\text{L}^{-1}$ (Fig. 3). Ammonium concentrations always exhibited values under the $0.05 \text{ mg}\cdot\text{L}^{-1}$ detection limit of the employed method.

The TP concentrations ranged from $1.82 \text{ mg}\cdot\text{L}^{-1}$ in February 2005 at site S1 and $0.23 \text{ mg}\cdot\text{L}^{-1}$ in November 2004 at site S1. The RSP concentrations ranged from $0.83 \text{ mg}\cdot\text{L}^{-1}$ in February 2005 at site S1, to $0.08 \text{ mg}\cdot\text{L}^{-1}$ in November 2004 at site S4. Figure 3 illustrates a marked peak in the RSP concentration during February. Silica exhibited low concentrations, ranging from moderate to a maximum of $6 \text{ mg}\cdot\text{L}^{-1}$ in February 2005 at site S3, and a minimum of $0.02 \text{ mg}\cdot\text{L}^{-1}$ in June 2005 at site S2 (Fig. 3).

The nutrients exhibited a similar pattern at the four sampling sites. The exception was silica, which exhibited marked peaks at site S3, compared to the remaining sites.

The N:P ratio generally remained between 4 and 12, reaching extreme values of 1.8 and 13.8 (Fig. 3).

The chlorophyll-a concentration exhibited a maximum value of $70.03 \text{ mg}\cdot\text{m}^{-3}$ in October 2004 at site S3, and a minimum of $2.17 \text{ mg}\cdot\text{m}^{-3}$ in October 2004 at site S2 (Fig. 3).

Phytoplankton community

In the present study, 183 phytoplankton taxa were identified in the reservoir (Table 2), most (80) belonging to Chlorophyceae. The remaining taxa belonged to cyanobacteria (42), Bacillariophyceae (35), Zygnemaphyceae (10), Cryptophyceae (6), Chrysophyceae (3), Euglenophyceae (3), Xanthophyceae (2) and Dinophyceae (2). The genera *Scenedesmus*, *Navicula* and *Oocystis* exhibited the highest number of species, 11, 10 and 10 species, respectively. The specific richness was lower during the spring, with a minimum of 18 in September 2004 at site S1, and a maximum of 106 in March 2005 at site S3 (Fig. 4).

The phytoplankton abundance exhibited a maximum $368.037 \times 10^3 \text{ cells}\cdot\text{mL}^{-1}$ in February 2005 at site S3, and at

least $1.133 \times 10^3 \text{ cells}\cdot\text{mL}^{-1}$ in October 2004 at site S2. The largest number of cells was recorded between December 2004 and April 2005 (Fig. 4).

Chlorophyceae and cyanobacteria dominated during June and July 2004, the latter to a lesser extent, with *Dictyosphaerium ehrenbergianum*, *Dictyosphaerium pulchellum*, *Microcystis natans* and *Synechocystis* sp.1 being the most abundant species. There was a progressive increase of the relative abundance of diatoms by the end of July, and during August and September 2004, accounted for an increase in the density of *Cyclotella meneghiniana* and *Stephanodiscus* sp. A peak in cryptophyceae was observed between September and October 2004, attributable to an increase in the concentrations of *Rhodomonas lacustris* and *Cryptomonas ovata*. A period of dominance of Chlorophyceae and diatoms then followed. Finally, from December 2004 until May 2005, an almost exclusive dominance of cyanobacteria was observed, the product of a large relative abundance mainly of *A. circinalis* and *M. natans*, together with other companion species. These high concentrations of cyanobacteria characterized a bloom with an average density of $133.05 \times 10^3 \text{ cells}\cdot\text{mL}^{-1}$, and an average chlorophyll concentration of $28.7 \text{ mg}\cdot\text{m}^{-3}$ (Fig. 5).

The Shannon diversity index was lowest (1.4) in January 2005 at site S3. This period coincided with the highest phytoplankton density, caused by an increase of *A. circinalis* that represented $>80\%$ of the total phytoplankton cells. There were two other significant declines in the index of diversity at site S3 beyond the period of major phytoplankton growth, the first being in 20 September 2004 (1.72), the product of an increase in the relative abundance of *Microcystis aeruginosa* ($>70\%$), combined with a very low density of the remaining phytoplankton species. The second (1.95) occurred in 18 October 2004, emanating from an abundance of *Stephanodiscus* sp. in relation to the total number of phytoplankton cells (relative abundance $>70\%$). Declines in the index of diversity from mid-November always coincided with moments of dominance of different cyanobacteria species (Fig. 4).

Environmental parameters and phytoplankton community

Principal component analysis (PCA) ordination of environmental variables and sampling sites is presented in Fig. 6. The first three factors accounted for 70% of the total variability of the data, thereby being used to interpret the analysis. The samples from the months of December 2004, and January, February and March 2005 were on the right side of axis I, while the samples for June 2004 and 2005 and July 2005 were on the far left. Axis I (36.7% of the total

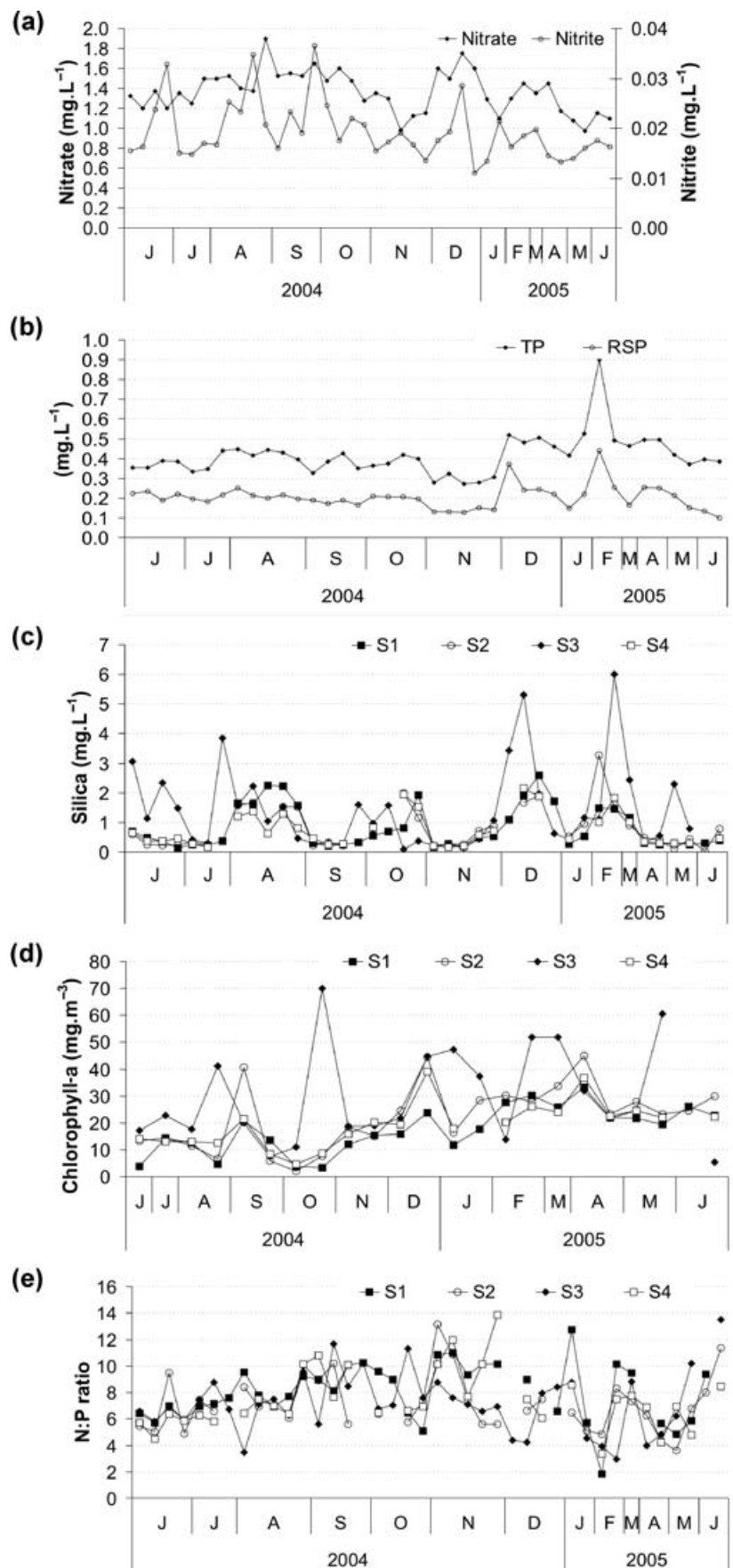


Fig. 3. Variation of (a) nitrates and nitrites; (b) phosphorus compounds (total phosphorus (TP), reactive soluble phosphorus (RSP)); (c) silica; (d) chlorophyll-a; (e) N:P ratio (nitrite + nitrate + ammonium:RSP) (the figures illustrate the average of the four sampling sites for the nitrogen and phosphorus compounds, while the sites are shown separately for silica, chlorophyll-a and N:P ratio as they exhibited differing behaviours).

Table 2. Phytoplankton species composition of Paso de las Piedras Reservoir. The pie chart shows the relative contribution of each taxonomical class to the phytoplankton composition.

CYANOBACTERIA

Chroococcales

Aphanocapsa delicatissima W. et G.S. West
Aphanothece smithii Kom.-Legn. & Cronb.
Chroococcus limneticus Lemm.
Chroococcus minutus (Kütz.) Näg.
Coelosphaerium aeruginum Lemm.
Coelosphaerium dubium Grunow
Coelosphaerium punctiferum Kom. & Kom.-Legn.
Coelosphaerium sp.
Cyanosarcina sp.
Merismopedia sp.
Microcrocis sp.
Microcystis aeruginosa (Kütz.) Kütz.
Microcystis flos-aquae (Wittr.) Kirchn.
Microcystis natans Lemm. ex Skuja
Microcystis protocystis Crow
Microcystis smithii Kom. & Anag.
Rhabdogloea scenedesmoides (Nyg.) Kom. & Anag.
Snowella lacustris (Chod.) Kom. et Hind.
Synechococcus nidulans (Pringsh.) Kom.
Synechocystis aquatilis Sauvageau
Synechocystis fuscopigmentosa Kovác.
Synechocystis sp.1
Synechocystis sp.2
Woronichinia sp.

Oscillatoriales

Borzia sp.
Jaaginema minimum (Gicklhorn) Anag. & Kom.
Lyngbya sp.
Oscillatoria foreau Frémy
Oscillatoria sp.1
Oscillatoria sp.2
Oscillatoria sp.3
Oscillatoria sp.4
Oscillatoria sp.5
Oscillatoria sp.6
Oscillatoria sp.7
Oscillatoria sp.8
Phormidium hamelii (Frémy) Anag. & Kom.
Planktothrix agardhii (Gomont) Kom. & Anag.
Pseudoanabaena mucicola (Hub.-Pest. & Naumann) Schwabe
Spirulina major Kütz. ex Gom.

Nostocales

Anabaena circinalis Rab.
Anabaena sphaerica Born. et Flah.

CHLOROPHYCEAE

Chlorococcales

Ankyra judayi (G.M. Smith) Fott
Botryococcus braunii Kütz.

Chlorella minutissima Fott et. Novák.
Chlorella oocystoides Hind.
Coelastrum astroideum De Not.
Coelastrum indicum Tur.
Coelastrum microporum Näg.
Coenochloris planconvexa Hind.
Coenocystis planctonica Kors.
Crucigenia quadrata Morr.
Dictyosphaerium ehrenbergianum Näg.
Dictyosphaerium pulchellum Wood
Golenkinia radiata Chod.
Kirchneriella contorta var. *elegans* (Playf.) Kom.
Kirchneriella cornuta Kors.
Kirchneriella irregularis (G.M. Smith) Kors.
Kirchneriella irregularis var. *spiralis* Kors.
Kirchneriella obesa (W. West) Schmidle
Kirchneriella sp.
Lagerheimia balatonica (Scherff. in Koll) Hind.
Lagerheimia citrififormis (Snow) Coll.
Lagerheimia genevensis (Chod.) Chod.
Lagerheimia longiseta (Lemm.) Wille
Lagerheimia subsalsa Lemm.
Monoraphidium circinale (Nyg.) Nyg.
Monoraphidium contortum (Thur.) Kom.-Legn.
Monoraphidium dybowskii (Wolosz.) Hind. & Kom.-Legn.
Monoraphidium griffithii (Berk.) Kom.-Legn.
Monoraphidium mirabile (W. & G.S. West) Pankow
Monoraphidium tortile (W. & G.S. West) Kom.-Legn.
Oocystella rhomboidea (Fott) Hind.
Oocystis borgei J. Snow
Oocystis ecbalocystiformis Iyeng.
Oocystis eremosphaeria G. M. Smith
Oocystis lacustris Chod.
Oocystis marssonii Lemm.
Oocystis naegelii A. Br.
Oocystis parva W. & G.S. West
Oocystis solitaria Wittr. in Wittr. & Nordst.
Oocystis submarina Lagerh.
Pediastrum boryanum (Turp.) Menegh.
Pediastrum duplex Meyen
Pediastrum tetras (Ehrenb.) Ralfs
Planktosphaeria gelatinosa G.M. Smith
Pseudokirchneriella major (Bernard) Hind.
Pseudokirchneriella subcapitata (Kors.) Hind.
Pseudoquadrigula sp.
Quadricoccus ellipticus Hortob.
Quadrigula quaternata (W. & G.S. West) Printz
Raphidocelis rotunda (Kors.) Hind.
Rayssiella hemisphaerica Edelst. et Presc.
Scenedesmus acuminatus var. *elongatus* G.M. Smith

Table 2. Continued

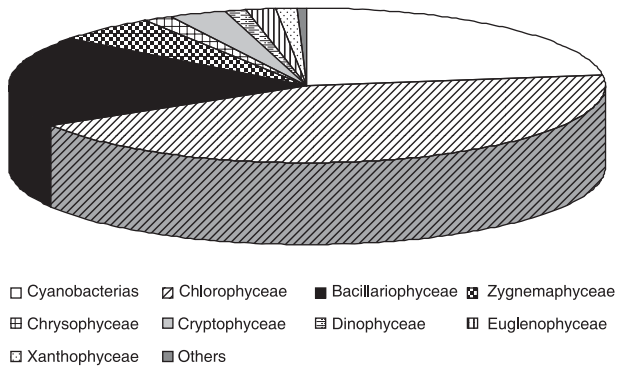
<i>Scenedesmus acutus</i> Meyen	Euglenales
<i>Scenedesmus bicaudatus</i> Dedus.	<i>Euglena</i> sp.
<i>Scenedesmus brasiliensis</i> Boh.	<i>Trachelomonas rotunda</i> Swirenko
<i>Scenedesmus disciformis</i> (Chad.) Fott et Kom.	<i>Trachelomonas</i> sp.
<i>Scenedesmus intermedius</i> Chad.	XANTHOPHYCEAE
<i>Scenedesmus linearis</i> Kom.	Mischococcales
<i>Scenedesmus nanus</i> Chad.	<i>Isthmochloron</i> sp.
<i>Scenedesmus protuberans</i> Fritsch	<i>Tetraplektron torsum</i> (Skuja) Dedus. Scæg.
<i>Scenedesmus quadricauda</i> (Turp.) Bréb.	ZYGNEMAPHYCEAE
<i>Scenedesmus semipulcher</i> Hortob.	Desmidiiales
<i>Schroederia setigera</i> (Schröd.) Lemm.	<i>Closterium aciculare</i> T. West
<i>Schroederia</i> sp.	<i>Closterium acutum</i> Bréb ex Ralfs
<i>Selenastrum capricornutum</i> Printz	<i>Closterium moniliferum</i> (Bory) Ehrenb. ex Ralfs
<i>Selenodictyon</i> sp.	<i>Cosmarium botrytis</i> (Menegh.) Ralfs
<i>Sphaerocystis schroeteri</i> Chod.	<i>Staurastrum chaetoceros</i> (W. & G.S.West) G.M.Sm.
<i>Tetraedron hemisphaericum</i> Skuja	<i>Staurastrum gracile</i> Ralfs ex Ralfs
<i>Tetraedron minimum</i> (A. Br.) Hansg.	<i>Staurastrum hexacerum</i> (Ehrenb.) Wittr.
<i>Tetrastrum komarekii</i> Hind.	<i>Staurastrum</i> sp.
<i>Tetrastrum staurogeniaeforme</i> (Schröd.) Lemm.	<i>Staurodesmus cuspidatus</i> Bréb ex Ralfs
<i>Thorakochloris nygaardii</i> Komárek	Zygnematales
Tetrasporales	<i>Spirogyra</i> sp.
<i>Tetraspora</i> sp.	BACILLARIOPHYCEAE
Ulotrichales	Centrales
<i>Binuclearia eriensis</i> Tiff.	<i>Aulacoseira granulata</i> (Ehrenb.) Simonsen
Volvocales	<i>Aulacoseira granulata</i> var. <i>angustissima</i> (Müller) Simonsen
<i>Chlamydomonas</i> sp1.	<i>Coscinodiscus</i> sp.
<i>Chlamydomonas</i> sp 2.	<i>Cyclotella meneghiniana</i> Kütz.
<i>Chlamydomonas</i> sp 3.	<i>Melosira varians</i> Ag.
<i>Chlamydomonas peterfii</i> Gerloff	<i>Stephanodiscus</i> sp.
<i>Phacotus lenticularis</i> (Ehrenb.) Stein	Pennales
<i>Pteromonas</i> sp.	<i>Amphora ovalis</i> (Kütz.) Kütz.
CHRYSOPHYCEAE	<i>Cocconeis placentula</i> Ehrenb.
Chromulinales	<i>Cymatopleura solea</i> (Breb.) W. Smith
<i>Chromulina</i> sp.	<i>Cymbella</i> sp.
Ochromonadales	<i>Diatoma vulgare</i> Bory de St. Vincent
<i>Ochromonas</i> sp.	<i>Diploneis</i> sp.
Rhizochrysidales	<i>Epithemia sorex</i> Kütz.
<i>Lagynion</i> sp.	<i>Fragilaria vaucheriae</i> (Kütz.) Baye Peter
CRYPTOPHYCEAE	<i>Gomphonema olivaceum</i> (Lyngb.) Kütz.
Cryptomonadales	<i>Gyrosigma spencerii</i> (Quek.) Griff. & Henfr.
<i>Chilomonas</i> sp.	<i>Hantzchia amphyxis</i> (Ehrenb.) Grunow
<i>Cryptomonas ovata</i> Her.	<i>Navicula cryptocephala</i> Kütz.
<i>Cryptomonas marssonii</i> Skuja	<i>Navicula exigua</i> Greg. ex Grunow
<i>Rhodomonas lacustris</i> Pasch. et Ruttn.	<i>Navicula goeppertiana</i> (Bleisch) H.L. Smith
<i>Chroomonas</i> sp.	<i>Navicula peregrina</i> (Her.) Kütz.
<i>Cyanomonas</i> sp.	<i>Navicula pygmaea</i> Kütz.
DINOPHYCEAE	<i>Navicula radiosa</i> Kütz.
Peridinales	<i>Navicula recens</i> (Lange-Bert.) Lange-Bert.
<i>Ceratium hirundinella</i> (O.F.Muell.) Duj.	<i>Navicula subcapitata</i> (Greg.) Ralfs
<i>Peridinium</i> sp.	<i>Navicula tripunctata</i> (O. Müll.) Bory
EUGLENOPHYCEAE	<i>Navicula veneta</i> Kütz.

Table 2. *Continued*

Nitzschia acicularis (Kütz.) W. Smith
Nitzschia dissipata (Kütz.) Grun.
Nitzschia hungarica Grun.
Nitzschia palea (Kütz.) W. Smith
Nitzschia sigmaidea (Ehrenb.) W. Smith
Rhoicosphenia curvata (Kütz.) Grun.
Rhopalodia operculata (Agardh) Håkansson
Synedra acus Kütz.

OTHERS

Not identified flagellated



variance) was mainly related to water and air temperatures. Axis II (17.6% of the total variance) was related to the N:P ratio, being those on the upper end samples from the months of November and September 2004, and January 2005, mainly those pertaining to sites S1 and S4, as well as those at the bottom end from the months of June, July and August 2004, and February, April and May 2005. Axis III (16% of the total variance) was characterized by the concentration of NO_3^- and NO_2^- , remaining at the top of the chart samples from the months of June 2004 at sites S1 and S2, and August 2004 at the four sampling sites, all being characterized by higher concentrations of these two nutrients.

The CCA was performed with just four environmental variables (N:P ratio; air temperature; RSP; NO_3^-). These variables presented the longest arrows in the PCA and, therefore, were most relevant for differentiating sampling sites. The results of CCA are illustrated in Fig. 7.

Bacillariophyceae, Chlorophyceae, Cryptophyceae and Zygnemaphyceae were placed near the origin of the ordination diagram, meaning these phytoplankton groups were present in all samples, and were not associated with one or more of the measured environmental variables. Xanthophyceae seemed to be related to sites and months with high temperatures. Euglenophyceae were associated with environments containing high nitrate levels and low soluble phosphorus levels. In contrast, cyanobacteria were

associated with low nitrate contents and a low N:P ratio, and high RSP contents and temperatures, although just as Chlorophyceae, diatoms and Cryptophyceae occupied a position quite close to the origin. Dinophyceae were associated with environments similar to those that of cyanobacteria, except that they occupied a position much more extreme in the ordination diagram. Finally, Chrysophyceae were associated with low temperatures, a high N:P ratio and high nitrate concentrations.

Groups occupying the most extreme positions within the diagram were those with fewer species and occasional occurrences, whereas groups located closer to the origin were those exhibiting more diversity and numerical occurrence. This denoted that different species within each taxonomic group exhibited evidence of particular adaptations to different environmental conditions. The percentage of variance obtained from the ratio phytoplankton groups-environmental variables was 70% and 23% of axes I and II, respectively. The correlation between phytoplankton groups and environmental variables was highly significant ($P < 0.001$).

Trophic state

Based on the OECD 'open limit' classification scheme, and considering the TP and Secchi depth values, Paso de las Piedras Reservoir lies on the trophic borderline between eutrophy and hypertrophy. If TP alone were considered, there is a very high probability (>85%) to classify the reservoir as hypertrophic. The average and maximum chlorophyll concentrations suggest the lake exhibits mesotrophic, eutrophic and hypertrophic characteristics, with the highest probability of being within the eutrophic category. Sampling sites S1, S2 and S4, however, all exhibited average and maximum chlorophyll-a concentrations lower than that observed for site S3 (Table 3).

Utilizing TP data with the Carlson index, the TSI (TP) values were very high, ranging between 83 and 98, which would place the reservoir in a hypertrophic category. In contrast, intermediate values of Secchi depth and chlorophyll were obtained, with the TSI (SD) values ranging between 49 and 59 and the TSI (Chl a) value ranging between 30 and 72, which would place the lake in a eutrophic category. The TSI (TP) was statistically higher than the TSI (Chl a) which, in turn, exhibited higher values than the TSI (SD) ($P < 0.05$). Significant differences between monitoring sites were found for TSI (TP) and TSI (SD), with statistically higher values of those indexes exhibited at site S3, regarding to site S1 ($P < 0.05$) (Fig. 8).

DISCUSSION

The results of this study indicate the seasonal variations of physical, chemical and biological parameters in the waters

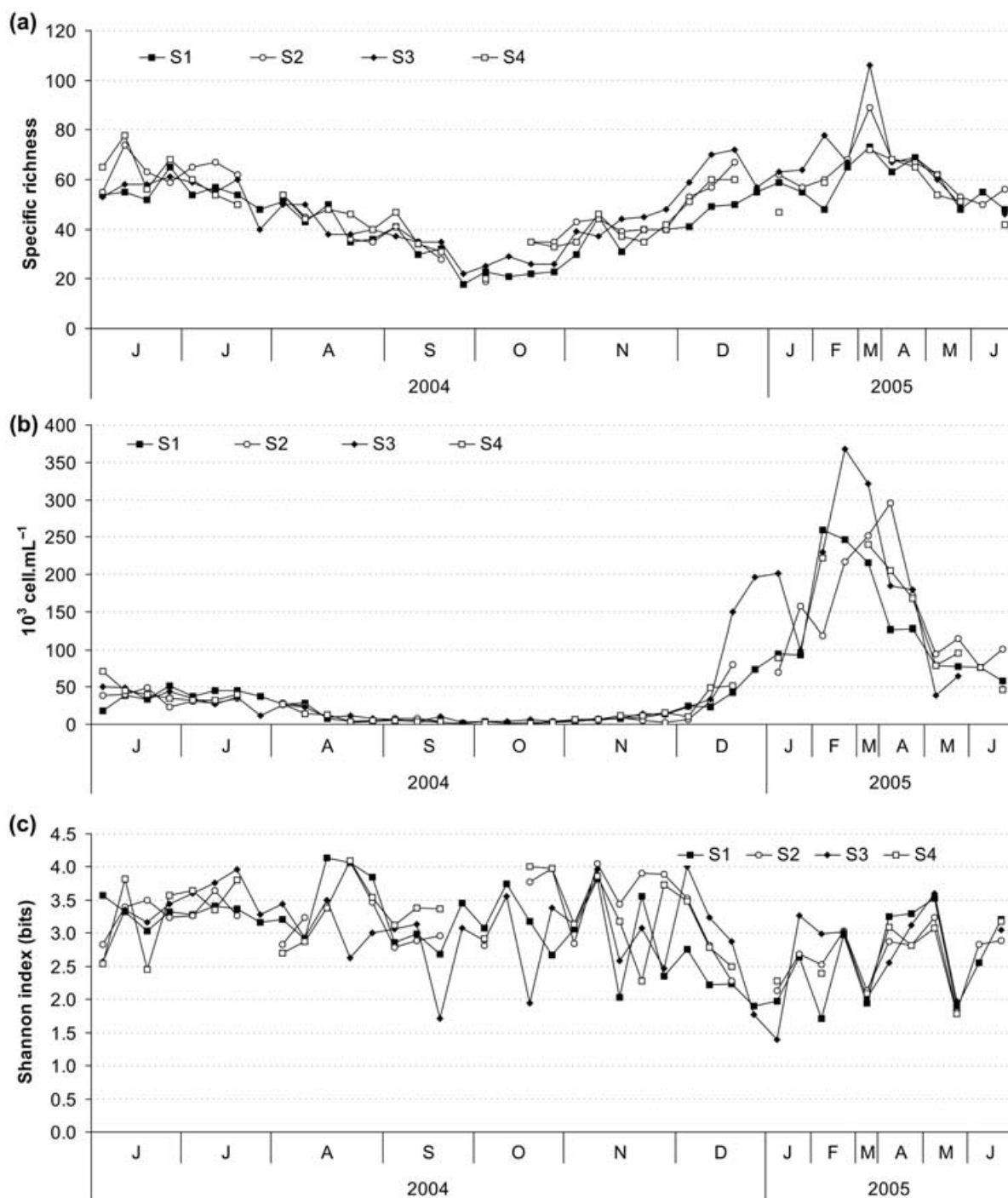


Fig. 4. Variations of (a) specific richness; (b) total phytoplankton abundance; and (c) Shannon diversity index (H') for four sampling sites.

of Paso de las Piedras Reservoir were essentially a consequence of the environmental and hydrological conditions in the dam area, while the spatial variations within the lake resulted from the characteristics of the water inflows from the two main tributaries. This can explain why some parameters (e.g. electrical conductivity; Secchi depth; silica and chlorophyll concentrations) exhibited a different behaviour at site S3 than for sites S1,

S2 and S4. The S3 values generally indicated a lower water quality, considering that site S3 is located near the mouth of El Divisorio Creek, which brings more turbid waters to the lake as it carries a significant quantity of particulate matter and dissolved salts. Nevertheless, as the water intake of El Divisorio Creek is ≈ 10 times smaller than that of the Sauce Grande River, which contains clearer water, the material provided by El Divisorio Creek was quickly

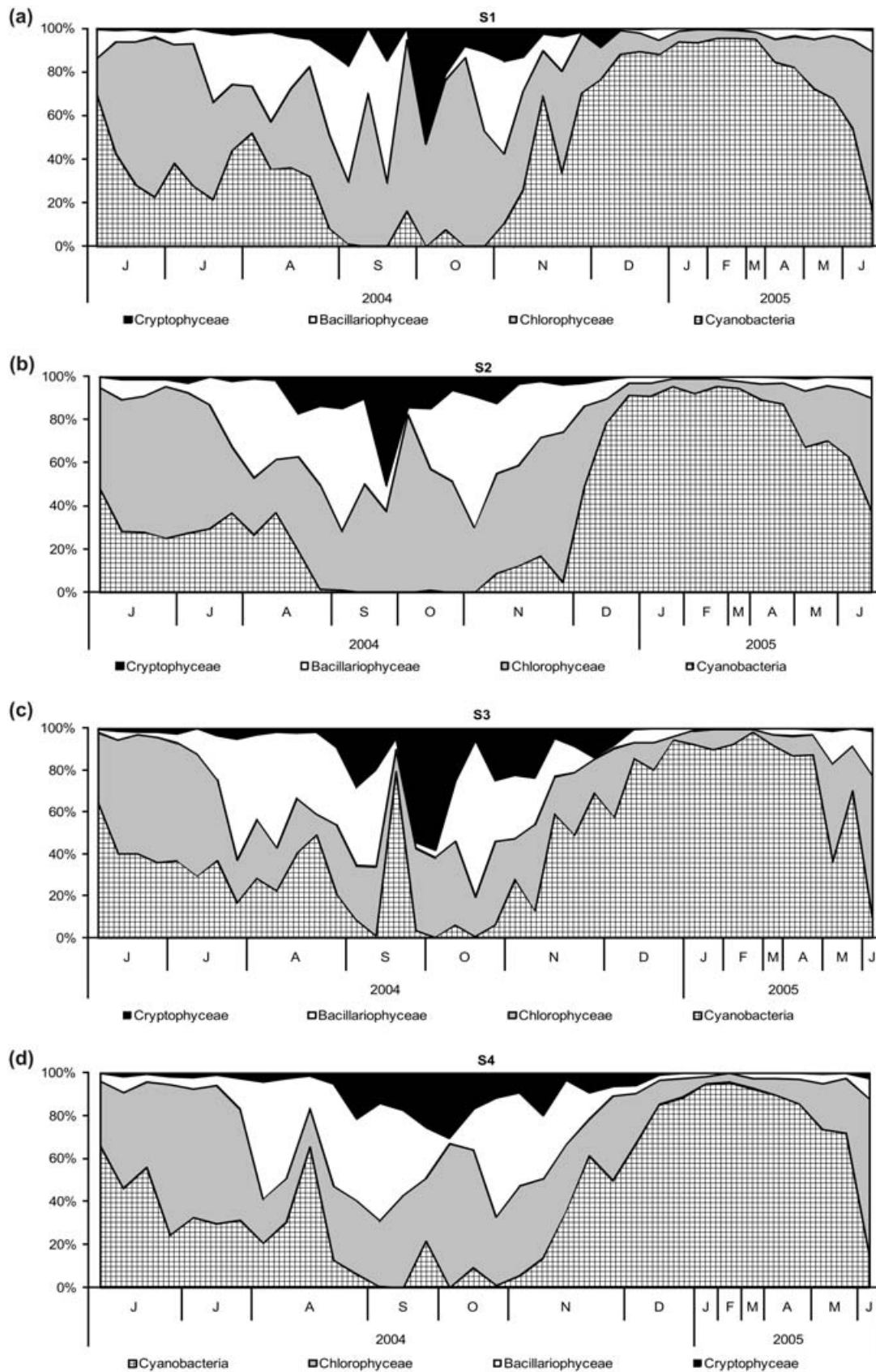


Fig. 5. Variations of relative abundance of each phytoplankton class for the four sampling sites.

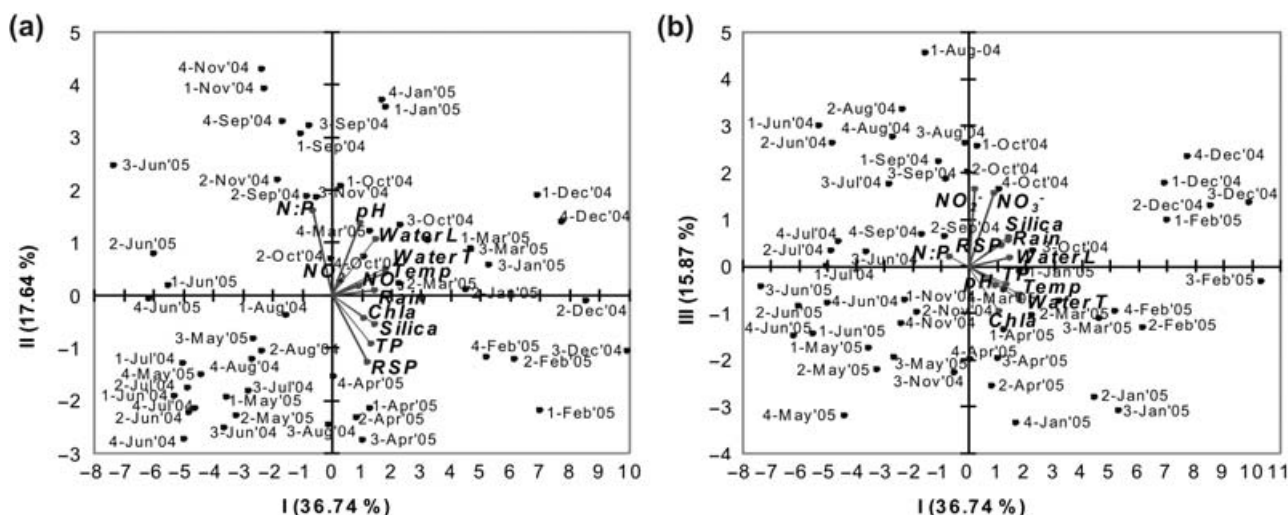


Fig. 6. Principal component analysis performed on environmental parameters (Sampling site codes: First number corresponds to sampling site (1 = S1; 2 = S2; 3 = S3; 4 = S4) and subsequent ones to month and year. Environmental variables: Rain (cumulative rainfall in the days previous to sampling); NO_2^- (nitrite concentration); NO_3^- (nitrate concentration); RSP (soluble reactive phosphorus concentration); TP (total phosphorus concentration); silica (silica concentration); N:P (nitrogen to phosphorus ratio); pH; Water L (water level in lagoon); Water T (water temperature); Temp (air temperature); Chl a (Chlorophyll-a concentration)).

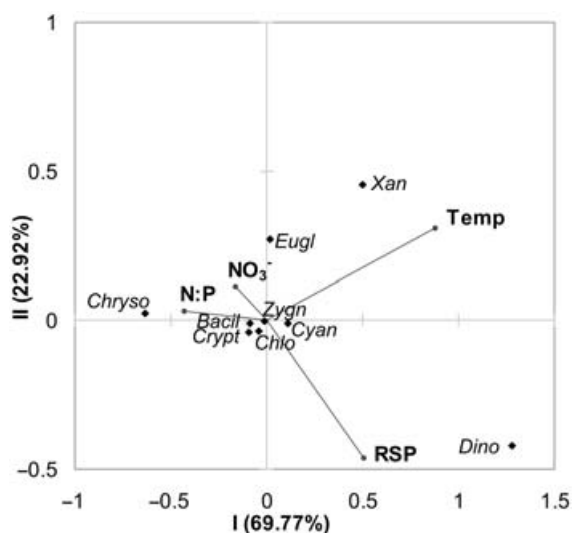


Fig. 7. Canonical correspondence analysis performed with phytoplankton groups (Environmental variables: NO_3^- (nitrate concentration); RSP (soluble reactive phosphorus concentration); N:P (nitrogen to phosphorus ratio); Temp (air temperature). Phytoplankton: Xan (Xanthophyceae); Eugl (Euglenophyceae); Zygn (Zygnemaphyceae); Cyan (Cyanobacteria); Chlo (Chlorophyceae); Dino (Dinophyceae); Crypto (Cryptophyceae); Baci (Bacillariophyceae); Chryso (Chrysophyceae)).

diluted, thereby not affecting the remaining sampling sites. Furthermore, the direction of the prevailing winds (north-northwest) promoted accumulation of particles inside site S3.

Regarding physicochemical variables whose values are regulated by international agencies, namely TSS ($<500 \text{ mg.L}^{-1}$ in drinking water, according to EPA 2003), electric conductivity ($<800 \mu\text{S.cm}^{-1}$ in drinking water, according to the World Health Organization (MDBC 1999)), and pH (6.5–8.5 in drinking water, according to EPA 2003). Only the pH presented values higher than the recommended interval, conditions consistent with the system’s high productivity level.

In regard to nutrients, the recorded nitrate values were always lower than the thresholds for drink water sources with conventional treatment established in Law 24051, Decree 831/93, of 10 mg.L^{-1} . The NO_2^- only exceeded briefly the guideline value of 0.05 mg.L^{-1} in two dates in June 2004, indicating that using the reservoir as a drinking water source would not a health risk to the general population. Nevertheless, both nitrogen and phosphorus exhibited high concentrations, compared to other lakes with similar characteristics, examples being Ben Chifley Reservoir in Australia (TN = $0.5\text{--}2.2 \text{ mg.L}^{-1}$; RSP = $0.003\text{--}0.017 \text{ mg.L}^{-1}$; TP = $0.03\text{--}0.057 \text{ mg.L}^{-1}$) (Rahman *et al.* 2005), and Gargalheiras Reservoir in Brazil (TP = 0.763 mg.L^{-1} ; TN = 1.098 mg.L^{-1}) (Chellappa & Medeiros Costa 2003). They were always well above the limitation values for phytoplankton growth of $\approx 5 \mu\text{g.L}^{-1}$ for RSP, and $20 \mu\text{g.L}^{-1}$ for TN (Ryding & Rast 1992).

Moreover, the N:P ratio ranged around the Redfield ratio (atomic ratio 16 : 1 or mass ratio of 7 : 1), indicating neither nutrient is the main limiting factor for the maximum algal biomass. This observation has important

Table 3. Trophic state of Paso de las Piedras Reservoir according to "open limit" criteria from OECD (1982).

Variable		Paso de las Piedras Reservoir								Clasificación
		Oligotrophic	Mesotrophic	Eutrophic	Hypertrophic	S1	S2	S3	S4	
Total Phosphorus ($\mu\text{g.L}^{-1}$)	X	8.0	26.7	84.4		350	368	401	362	10–12% eutrophic
	X \pm 1 SD	4.85–13.3	14.5–49	48–189						88–92% hypertrophic
	X \pm 2 SD	2.9–22.1	7.9–90.8	16.8–424						
	Range	3.0–17.7	10.9–95.6	16.2–386	750–1200					
	<i>n</i>	21	19 (21)	71 (72)	2					
Total Nitrogen ($\mu\text{g.L}^{-1}$)	X	661	753	1875		1441	1370	1363	1278	eutrophic
	X \pm 1 SD	371–1180	485–1170	861–4081						
	X \pm 2 SD	208–2103	313–1816	395–8913						
	Range	307–1630	361–1387	393–6100						
	<i>n</i>	11	8	37 (38)						
Chlorophyll a ($\mu\text{g.L}^{-1}$)	X	1.7	4.7	14.3		14.67	18.33	24.64	17.43	5–21% mesotrophic
	X \pm 1 SD	0.8–3.4	3.0–7.4	6.7–3.1						48–62% eutrophic
	X \pm 2 SD	0.4–7.1	1.9–11.6	3.1–66						15–48% hypertrophic
	Range	0.3–4.5	3.0–11	2.7–78	100–150					
	<i>n</i>	22	16 (17)	70 (71)	2					
Chlorophyll a Peak Value ($\mu\text{g.L}^{-1}$)	X	4.2	16.1	42.6		33.26	44.97	70.03	39.03	5–27% mesotrophic
	X \pm 1 SD	2.6–7.6	8.9–29	16.9–107						51–55% eutrophic
	X \pm 2 SD	1.5–13	4.9–52.5	6.7–270						20–40% hypertrophic
	Range	1.3–10.6	4.9–49.5	9.5–275						
	<i>n</i>	16	12	46						
Secchi Depth (m)	X	9.9	4.2	2.45		1.73		1.34		2–10% mesotrophic
	X \pm 1 SD	5.9–16.5	2.4–7.4	1.45–4						28–50% eutrophic
	X \pm 2 SD	3.6–27.5	1.4–13	0.9–6.7						38–70% hypertrophic
	Range	5.4–28.3	1.5–8.1	0.8–7.0	0.4–0.5					
	<i>n</i>	13	20	70 (72)						

X, geometric media

SD, standard deviation

(), the values between parenthesis correspond to the number of variable (*n*) used in the first calculation.

consequences from an ecological perspective, mainly because the high nutrient availability established a scenario in which variations in the community structure, and the development of phytoplankton blooms, would be more constrained by environmental and hydrological conditions than by nutrients.

Variations in the concentrations of different phosphorus compounds observed throughout the sampling period were less marked than changes in the nitrogen concentrations. This could indicate the phosphorus inflow into the water column is more closely related to phosphorus remobilization from the bottom sediments than from an external input, so that the phosphorus dynamics would not be as readily influenced by environmental factors than the nitrogen dynamics. It has

been pointed out by numerous authors that sediments can act both as a source and as a storage of nutrients into lakes and reservoirs, and that recycling the same interface in the sediment–water plays a critical role in the development of eutrophication, and in establishing algae blooms which, in turn would be strongly influenced by physical factors such as water temperature and renewal (Ryding & Rast 1992; Al Bakri & Chowdhury 2006).

The high nutrient concentration recorded in Paso de las Piedras Reservoir could be explained by the concurrence of various diffuse sources, including: (i) intense agricultural and cattle activity throughout the basin; (ii) the characteristics of the land area in which the reservoir is located (i.e. fertile soils with high phosphorus contents); and (iii) increasing urbanization and use of watercourses as drainage basins.

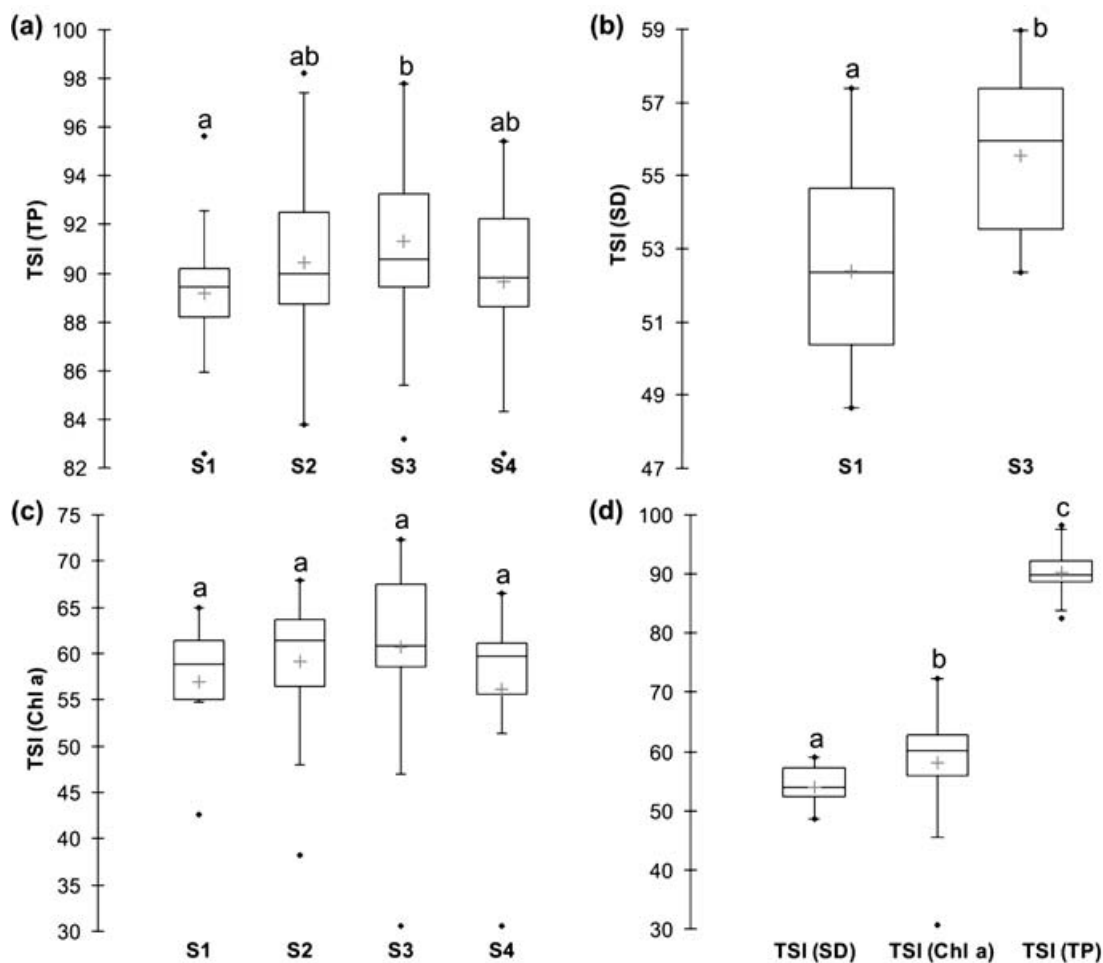


Fig. 8. Box plots comparing the values of trophic status indexes (TSI): (a) box plot compares values of trophic status index (TSI) based on total phosphorus values in the four seasons: TSI (TP); (b) box plot compares values of TSI based on Secchi depth values for sampling site S1 and S3 (the other stations were sampled from shore): TSI (SD); (c) box plot compares values of TSI based on chlorophyll values for the four sampling sites: TSI (Chl a); (d) box plot compares values of the three calculated indexes among themselves. Same letters indicate no difference among means ($P < 0.05$). The ● indicates suspected outlier and + indicates the mean).

The species richness was high, with values even higher than those reported in previous studies (Intartaglia & Sala 1989). This is surprising if it is considered that one eutrophication symptom is a reduction in the number of species (Ryding & Rast 1992), together with a reduction in the diversity index (Margalef 1983). Nevertheless, many eutrophic lakes exhibit a high phytoplankton diversity, which contradicts the competitive exclusion principle (Hardin 1960). This dilemma, also known as Hutchinson's 'paradox of the plankton', has been explained by non-equilibrium environmental fluctuation (Hutchinson 1961).

The phytoplankton taxa structure was highly correlated with environmental variables in Paso de las Piedras Reservoir, with switching between dominant taxa being observed throughout the study period. Four periods can be highlighted, the first characterized by Chlorophyceae

dominance, followed by a shorter period of Bacillariophyceae dominance, and a peak of Cryptophyceae dominance. The more conspicuous period was undoubtedly that characterized by the dominance of cyanobacteria, which lasted all summer and autumn, leading to development of an algal bloom exhibiting a succession of different species, many being potentially toxicogenic.

Considering the two classification systems used in this study to assess trophic status (i.e. OECD 'open limits' scheme; Carlson TSI), and based on TP data, the reservoir fits within the hypertrophic category. Considering the chlorophyll and Secchi depth data, however, the lake would fit within the eutrophic category.

Using two or more variables in the Carlson TSI can produce differences in its values, as observed for Lake Skadar (Rakocevic-Nedovic & Hollert 2005) and Foz de

Almargem coastal lagoon (Coelho *et al.* 2007). According to Carlson (1977), the accuracy of the index values based on TP depends on the assumption that phosphorus is the main algal biomass limiting factor, and that the concentration of all forms of phosphorus present in the waterbody is a function of algal biomass. These assumptions are not easy to demonstrate in many waterbodies and, therefore, Carlson suggested giving priority to biological parameters that represent visible symptoms of eutrophication, such as chlorophyll, when classifying a lake's trophic status, consistent with observations also presented by Ryding and Rast (1992).

While there are differences in the calculated trophic state for Paso de las Piedras Reservoir, based on different data, there is nevertheless a clear trend indicating a high degree of productivity in the system, which coincides with the cyanobacterial bloom phenomena observed during the months of summer and early autumn. The high trophic state values calculated from the TP concentration, related to other variables, also would indicate that the phytoplankton structure in Paso de las Piedras Reservoir is probably limited by factors other than phosphorus.

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