

First characterization of a shallow tropical reservoir at risk in northern Argentina

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ABSTRACT. The study of shallow tropical environments in northwestern Argentina is poorly developed. The Itiyuro reservoir is of major importance for supplying drinking water to more than 100000 people in the San Martín department. The development of cyanobacterial blooms with cyanotoxin-producing species in the reservoir constitutes a health problem. The present study explores the relationship between phytoplankton communities and limnological variables in the Caraparí river and the Itiyuro reservoir, between June 2018 and January 2020, using standardized techniques. Bacillariophyta were dominant in the river, and cyanobacteria and chlorophytes, in the reservoir. A multispecific bloom of cyanobacteria was observed in the summer of the last year sampled. Only 22% of the species in the reservoir were dominant, including *Aphanizomenon gracile*, *Raphidiopsis* spp., *Pseudanabaena limnetica* and *Monoraphidium contortum*; this implies a low average dominance (0.29) in 65% of the samples. In both systems, 50% of the species were rare, and the abundance of the constant species was low. Phytoplankton abundance —and, in particular, of cyanobacteria— was higher in the high-water phase, characterized by a high average temperature (22.7 °C) and higher nutrient inputs from runoff. To date, there is no continuous monitoring of the reservoir by the drinking water service providers. Performing periodic samplings would allow the detection of algal and cyanobacterial blooms and the release of toxins in time, as well as developing management protocols.

[Keywords: Itiyuro, Caraparí, phytoplankton, cyanobacteria, Argentina]

RESUMEN. Primera caracterización de un reservorio tropical somero en riesgo en el norte de la Argentina. El estudio de ambientes tropicales someros del noroeste argentino está poco desarrollado. El embalse Itiyuro es muy importante por abastecer de agua potable a más de 100000 personas en el departamento San Martín. En este embalse, las floraciones de cianobacterias con especies productoras de cianotoxinas constituye un problema de índole sanitario. Este trabajo explora la relación entre las comunidades del fitoplancton y las variables limnológicas en el río Caraparí y en el embalse Itiyuro, entre junio de 2018 y enero de 2020. Mientras que las diatomeas fueron dominantes en el río, las cianobacterias y clorofitas lo fueron en el embalse. Se observó una floración multiespecífica de cianobacterias en el verano del último año muestreado. Sólo el 22% de las especies del embalse fueron dominantes, entre ellas *Aphanizomenon gracile*, *Raphidiopsis* spp., *Pseudanabaena limnetica* y *Monoraphidium contortum*, lo que implica una baja dominancia promedio (0.29) en el 65% de las muestras. En ambos sistemas, el 50% de las especies fueron raras, y la abundancia de las especies constantes fue escasa. La abundancia del fitoplancton —y, en particular, de las cianobacterias— fue más alta en la fase de aguas altas, caracterizada por una temperatura media elevada (22.7 °C) y un ingreso mayor de nutrientes por escorrentía. A la fecha, las prestadoras del servicio de agua potable no realizaron monitoreos continuos en el embalse. La realización de muestreos periódicos permitiría detectar a tiempo las floraciones de algas y cianobacterias y la liberación de toxinas, como así también desarrollar protocolos de gestión.

[Palabras clave: Itiyuro, Caraparí, fitoplancton, cianobacterias, Argentina]

INTRODUCTION

Climate change, anthropogenic pollution and accelerated eutrophication are the main threats to continental freshwater sources. Those regions with the highest population density also correspond to the areas with the highest contamination of surface watercourses and aquifers due to urban or industrial discharges (Pochat 2005). In a context of constant world population growth, the demand for environmental resources and services for economic and subsistence purposes requires sustainable and rational management.

Reservoirs are one of the key tools to satisfy the needs of large populations. Among the services they provide, there are water supply for human and animal consumption, irrigation, hydroelectric power generation, flood regulation, and an innumerable amount of ecosystem services such as habitats for animal and plant species, among others (Bonilla 2009).

Eutrophication is a process of loss and deterioration of resource quality in aquatic ecosystems, although its effects seem to be more evident in shallow lakes and reservoirs.

One of the key causes is the enrichment of nutrients, mainly nitrogen and phosphorus (Reynolds 1984), which leads to an increase in phytoplankton biomass and, therefore, in the productivity of the system, in what is known as algal blooms (Bartram and Chorus 1999). All these conditions the use of the resource and has great ecological, health and economic impacts on a regional scale (Stefouli et al. 2005; Girão et al. 2007), and can interfere in the processes of water purification and treatment of the resource.

Eutrophication problems in Argentina are not new (Aguilera et al. 2017). The massive development of algae and cyanobacteria is quite common in reservoirs intended for drinking water supply (O' Farrell et al. 2019). Out of a total of 31 reservoirs analyzed until 2004, in 29% of them, cyanobacteria exceeded 30% relative frequency (Quirós 2004). Furthermore, based on several studies, it has been concluded that the general trophic state of reservoirs in Argentina varies between mesotrophic and eutrophic, depending on the season of the year, so that the development of blooms would be favored (Bazán et al. 2005; Amé et al. 2010).

The Itiyuro reservoir, located in the north of the province of Salta, supplies more than 100000 people, however it has not been deeply studied previously, and the monitoring carried out is scarce and isolated. Despite not being part of Argentine legislation, limnological and ecotoxicological studies of reservoirs that are

used to supply drinking water should be mandatory. In the province of Salta, most of the studies have been carried out in the larger reservoirs such as Cabra Corral and El Tunal (Salusso and Moraña 2005).

The objective of this study was to analyze the temporal variation of the composition and dynamics of the phytoplankton community in the Itiyuro reservoir and its main tributary (Caraparí river), as well as their relation with abiotic parameters. When comparing lotic and lentic systems, we expect greater proliferation of algae and cyanobacteria blooms in the reservoir than in the river, due to its characteristics (shallow environment, stability of the water column, etc.).

MATERIALS AND METHODS

Study area and methodology

The Itiyuro reservoir ($22^{\circ}6'12.29''$ S - $63^{\circ}44'21.34''$ W) located in northwestern Argentina (Salta province) is part of an arheic basin originating in southern Bolivia, fed by the pluvial Itiyuro river and formed by the confluence of the Caraparí and Itangüe rivers (Figure 1). The basin covers an area of 1153 km² (670 km² in Bolivia and 483 km² in Argentina) (Baumann et al. 2005). Most of the basin area is located in the semi-arid Chaco Forest and shrubland region, while a small part of the upper basin is characterized as Yungas forests (Cabrera 1994). During flood periods, the Itiyuro River is subjected to frequent

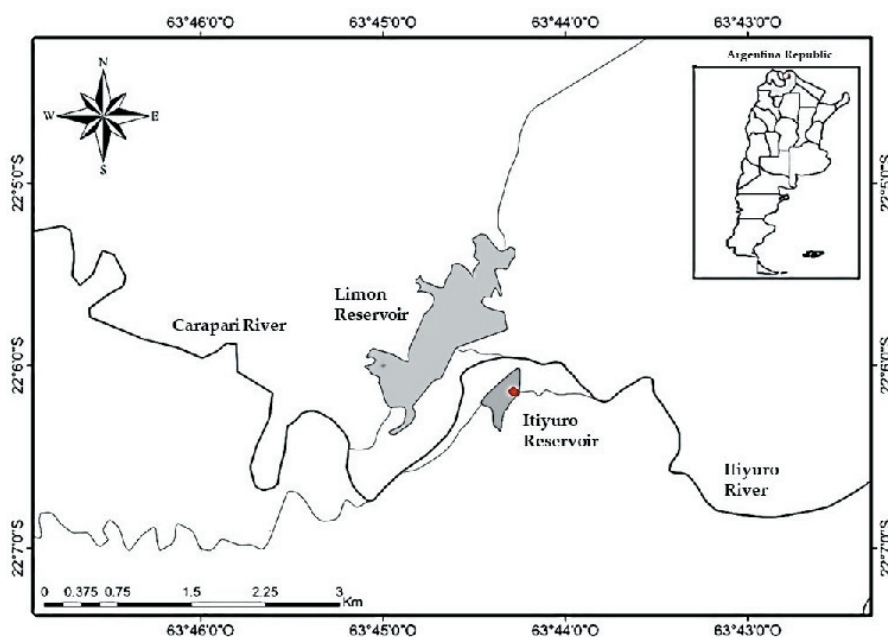


Figure 1. Itiyuro reservoir study area and the main tributary rivers, In red, the sampling area in the reservoir and at the entrance of the river.

Figura 1. Ubicación del embalse Itiyuro y sus principales ríos tributarios, en rojo se señala el área de muestro y el ingreso del río al embalse.

landslides that have reduced the reservoir's initial capacity of 80 hm³ to only 2 hm³, with a useful depth of 2 m and a surface area of 11 ha. Currently, it is used to supply water for a population of more than 100000 inhabitants (INDEC 2010).

The markedly tropical climate exceeds 970 mm of water per year with an average temperature of 21 °C and a dry period extending from April to September (Arias and Bianchi 1996). The average rainfall during the study period was 186.8 mm during the flood season and 23.41 mm during the dry season.

In the period from June 2018 to January 2020, a total of 22 samples were taken in the Itiyuro reservoir (22°06'08.2" S - 63°44'15.2" W) at a depth of a Secchi disk, with a monthly frequency in the period of low water (April to September), and bimonthly in the period of high water (October to March). Bimonthly samplings were carried out in the months with the highest temperature as they were the propitious months for the development of blooms. During the same period, 16 samples were collected in the Caraparí River prior to its entry into the reservoir (22°06'02.8" S - 63°44'17.8" W), 6 samples during low water and the remaining ones during the high-water period. Mainly in the rainy months, the Caraparí river presents abrupt floods that make access to take samples impossible; the months of July, September to December 2018 were sampled, and then from June 2019 to January 2020.

Phytoplankton samples collected by 30 µm mesh net, fixed in 4% formaldehyde, were used for qualitative analysis. Quantitative analysis was performed from samples taken at the depth of a Secchi disk, fixed in acidified lugol and stored at 4 °C until analysis. Counts were performed, after sedimentation for 24 h, in combined chambers using a Zeiss L inverted microscope following Utermöhl (1958). Each sample was counted to obtain at least 100 individuals of the most frequent species. The results were expressed in cells/mL. The cell number per filament was determined by dividing the total filament length by the average cell length (n=20). Organisms without cell content were not considered in the count. Species were identified by image capture using Axio Cam1Cc3 digital camera, and by using specialized bibliography: Komárek and Anagnostidis (1999, 2005), Komárek and Komárková-Legnerová (1969), Komárek et al. (2014) and Krammer and Lange-Bertalot (1986, 1991), among others.

Physical and chemical variables

In each sampling, temperature (°C), electrical conductivity (µs/cm), pH and dissolved oxygen (mg/L) were measured in situ (with an Orion multiparameter sensor), turbidity (with HACH brand turbidimeter [NTU]) and transparency (using a Secchi disk). Samples for physical-chemical analyzes were obtained with a Van Dorn sampler at the depth of a Secchi disk and were refrigerated until their analysis. Total and suspended solids (mg/L), true color, nitrates, nitrites and ammonium (mg N/L), soluble reactive phosphorus (mg SRP/L), chemical oxygen demand (COD) (mg O₂/L) alkalinity (mg CaCO₃/L) and hardness (mg CaCO₃/L) were determined in the laboratory, according to standardized techniques of APHA (2005), and chlorophyll *a* (Chl *a*) with the modified Scor-Unesco technique (Cabrera Silva 1984).

Statistical analysis

Kruskal-Wallis (H) nonparametric test was used since the analyzed variables did not fulfill the requirements of normality or homogeneity of the variance. The Mann-Whitney test was used to compare both phases of the hydrological cycle. A cluster analysis was carried out according to the method of Bray Curtis, trying to achieve the maximum homogeneity in each group and the greatest difference between the groups, analyzing only the Itiyuro reservoir. The variables considered were suspended solids, soluble inorganic nitrogen (SIN), SRP, turbidity and the nitrogen/phosphorus ratio (N/P). The trophic state of the reservoir was analyzed according to Carlson's Trophic State Index (TSI) based on chlorophyll *a* (Carlson 1977). Spearman correlations were calculated between the physical-chemical and biological parameters to analyze significant differences within each environment and between systems. A canonical correlation analysis was carried out for the Itiyuro reservoir, considering the variables: concentration of nitrites, nitrates, ammonium and orthophosphate; SIN; species richness, diversity according to the Shannon Index and total abundance. Variables considered were not previously transformed. The Itiyuro reservoir was analyzed considering the times of floods and low water flows, but also the transitions between these times. 'Flood' was defined as the period between December and February (coinciding with the rainy season marked in the area), 'low water' as the period between June and September (corresponding

to the dry season), and transitions from low water to high water (TLW-F) considering the months of October and November, and from high water to low water (TF-LW) considering the months April and May.

A principal component analysis (PCA) was performed to see the ordering of the samples and the following variables were used: pH, N/P, SIN, (CLa), COD, SRP and Secchi Disk (SD). The first two components of the analysis did not explain an acceptable percentage of the variation, so it was decided to work with the first 3 components. Variables considered were previously transformed. The Olmstead Tukey test (Sokal and Rohlf 1981) was used for the analysis of species dominance, using the natural logarithm of the abundances and their relative frequencies of appearance. The species were classified as: dominant (high abundance and frequency of appearance), occasional (high abundance and low frequency of appearance), constant (low abundance and high frequency of appearance) and rare (low abundance and frequency of appearance).

The similarity of species between the communities of the reservoir and the river was evaluated using the Jaccard index (J), modified by Ellenberg (1956), where the value closer to 1 indicates greater similarity (Magurran 1988). This index is defined by the following equation:

$$J = C / (S1 + S2 - C)$$

where $S1=N^{\circ}$ of species present in environment 1, $S2=N^{\circ}$ of species present in environment 2 and $C=N^{\circ}$ of species that are present in both samples.

Different statistical programs were used to analyze the results. With InfoStat, correlations

between physical chemical and biological variables were analyzed, multivariate analysis, inference between samples. With PAST v2.14 ecological indices such as Shannon, dominance and similarity were elaborated, and finally with R Studio graphs were created, PCA analysis and Olmstead Tukey test.

RESULTS

Physicochemical variables

Water temperature was 25 °C (± 5.2 °C) on average in both systems. The reservoir showed polymictic behavior throughout the analyzed cycle. During the summer months, the reservoir increased its average water level by one meter. Mean annual SD was 0.72 m (± 0.41 m), with mean values of 1.05 m (± 0.37 m) in winter and 0.46 m (± 0.41 m) in summer. The reservoir showed oxygen supersaturation in 63% of the samples, while the river showed supersaturation in 71% of the samples.

The main results of the physical and chemical variables (Table 1) indicate that the water has a neutral pH to slightly alkaline pH in both systems, while the parameters such as P-SRP, hardness and suspended solids were higher in Caraparí. However, only total solids and electrical conductivity exhibited significant differences between the river and the reservoir ($U=391$, $P'0.05$ and $U=390$, $P'0.05$, respectively). Chlorophyll *a* concentration had an average value of 16.80 $\mu\text{g/L}$. The minimum value in the reservoir (1.12 $\mu\text{g/L}$) was recorded in September 2018, and the maximum value (88.84 $\mu\text{g/L}$) in April 2019.

The grouping analysis according to Bray Curtis, performed on the basis of the physical and chemical variables of the reservoir,

Table 1. Physicochemical variables measured in Itiyuro and Caraparí in 2018-2020 period.

Tabla 1. Variables físicoquímicas medias en Itiyuro y Caraparí en el período 2018-2020

| Variable | Itiyuro | | | Caraparí | | |
|--|---------|------------|------------|----------|------------|------------|
| | Average | Max. value | Min. value | Average | Max. value | Min. value |
| Electrical conductivity ($\mu\text{S/cm}$) | 664.6 | 962 | 399.3 | 844.9 | 1930 | 421.8 |
| pH | 7.5 | 8.49 | 6.69 | 7.43 | 8.26 | 6.82 |
| Total Solids (mg/L) | 635.6 | 1736 | 160 | 742.2 | 2029 | 513 |
| Suspended solids (mg/L) | 55.9 | 557 | 3 | 145.5 | 660 | 2 |
| Nitrites (mg N- NO_2/L) | 0.02 | 0.08 | 0.001 | 0.01 | 0.03 | 0.0003 |
| Nitrates (mg N- NO_3/L) | 0.57 | 2.1 | 0.01 | 0.46 | 3.2 | 0.05 |
| P-SRP (mg P/L) | 0.26 | 1.36 | 0.02 | 0.46 | 3.2 | 0.05 |
| Alkalinity (mg CaCO_3/L) | 125.36 | 236.7 | 30 | 130.58 | 269.6 | 34 |
| Hardness (mg CaCO_3/L) | 322.61 | 1781 | 135 | 360.7 | 2204 | 69 |

showed two distinct seasons in 2019 (Table 2). A first group (floods) comprising sampling from February to May and the second half of December 2019, and a second group (low water), from June to the first half of December 2019. Only differences in turbidity and suspended solids ($U=50$, $P<0.005$) were significant between phases.

Structure and composition of phytoplankton in the reservoir

A total of 91 genera and 145 species were identified in Itiyuro. The reservoir presented a dominant richness of Chlorophyta (46 species), followed by Bacillariophyta (42 species) and

Cyanobacteria (31 species). All cyanobacterial species recorded in the system corresponded to potential cyanotoxin-producing taxa, with the orders Nostocales (14 species) and Synechococcales (11 species) predominating, followed by Chroococcales (3 species), Oscillatoriales (2 species) and Spirulinales (1 species).

In Itiyuro, cyanobacteria had a 34% average representation of the total phytoplankton (Figure 2), followed by 25.8% chlorophytes and 22.36% Bacillariophyta. In all the samplings, the remaining algal groups (Euglenophytes, Dinophytes, Chrysophytes, Cryptophytes, Ocrophytes, Xanthophytes) were a minority,

Table 2. Physical and chemical variables by groups considered in 2019 in Itiyuro reservoir, according to Bray Curtis.

Tabla 2. Variables físicas y químicas por grupos considerados en 2019 en el embalse Itiyuro, según análisis de Bray Curtis.

| Group | Variable | Units | Average | S.D. | Min. | Max. | Significant differences |
|-------|------------------|-------|---------|-------|-------|-------|-------------------------|
| 1 | Suspended solids | mg/L | 87.6 | 38.8 | 34 | 125 | ($U=50$, $p=0.0025$) |
| | Turbidity | NTU | 61.02 | 19.52 | 31.5 | 86.4 | ($U=50$, $p=0.0025$) |
| | SIN | mg/L | 1.55 | 0.86 | 0.7 | 2.99 | |
| | P-SRP | mg/L | 0.07 | 0.04 | 0.02 | 0.12 | |
| | N/P | | 26.95 | 1.35 | 12.13 | 49.8 | |
| 2 | Suspended solids | mg/L | 10.57 | 5.65 | 3 | 20 | ($U=50$, $p=0.0025$) |
| | Turbidity | NTU | 6.62 | 7.05 | 1.49 | 21.2 | ($U=50$, $p=0.0025$) |
| | SIN | mg/L | 0.7 | 0.09 | 0.56 | 0.81 | |
| | P-SRP | mg/L | 0.24 | 0.43 | 0.04 | 1.2 | |
| | N/P | - | 9.99 | 7.03 | 0.53 | 19.23 | |

*SIN: Soluble inorganic nitrogen; SRP: Soluble reactive phosphorus; N/P: nitrogen phosphorus ratio

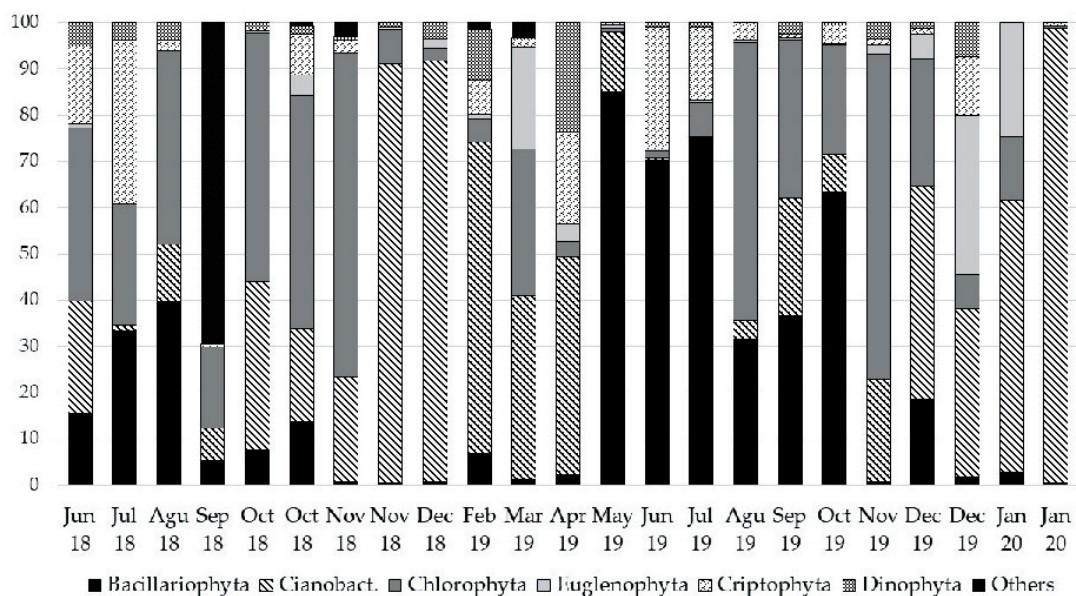


Figure 2. Relative abundances of the main groups of phytoplankton in the Itiyuro reservoir in the sampling period. *Haptophytes, Ocrophytes, Xanthophytes, Chrysophytes are classified as 'Other'.

Figure 2. Abundancias relativas por grupos principales del fitoplancton en el embalse Itiyuro en el período de muestreo. *Haptofitas, Ocrofitas, Xantofitas y Crisófitas son clasificadas como 'Otras'.

except for September 2018, when Haptophytes dominated, with a representation of 69%, explained by the species *Chrysochromulina parva*. In contrast to the rest of the groups, Bacillariophyta decreased its abundances in the reservoir in the warmer months, coinciding with the stability of the water body. In winter, the mean representativeness of Bacillariophyta was 24.9%, the genera *Fragilaria*, *Nitzschia* and *Cyclotella* stood out as the most frequent, they were found in more than 80% of the samplings. Haptophytes, Ocrophytes, Xanthophytes, Chrysophytes are classified as 'Other' in Figure 2.

The average annual richness of the reservoir was 19 species, with a maximum of 36 species in September 2018. The average annual phytoplankton abundance in the Itiyuro reservoir was 7060 cells/mL, with a maximum of 64560 cells/mL (January 2020), which was 98% explained by the abundance of cyanobacteria; the only cyanobacterial bloom event of the entire period studied was observed in this sampling. During the bloom, the species *Aphanocapsa delicatissima* (32940 cells/mL) and *Pseudanabaena catenata* (21110 cells/mL) stood out. The maximum values of Bacillariophyta (11216 cells/mL) and Chlorophyta (8708 cells/mL) were observed in autumn 2019 and spring 2018, respectively.

Phytoplankton abundances differed between phases of the hydrological cycle in the reservoir, with higher records in floods, especially of Cyanobacteria, coinciding with

higher temperatures, greater availability of nutrients that enter through runoff and lower transparency due to algal proliferation. In Itiyuro reservoir, a 40% increase in phytoplankton was recorded in times of floods characterized by high average temperatures (22.7 °C) with an average of 9984 cells/mL, when the stability of the water column favors the proliferation and accumulation of phytoplankton. In the November and January samplings, of the first and last year, respectively, the abundance of cyanobacteria was above the order of 104, while, in the sampling conducted in May 2019, it was Bacillariophyta that exceeded this order, with a bloom of *Cyclotella* sp. (11170 cells/mL) (Figure 3). Precipitation had a marked influence on the establishment and development of the different phytoplankton groups, positively influencing the abundance of cyanobacteria ($R=0.58$, $P=0.0040$) and Euglenophytes ($R=0.74$, $P=0.0001$).

Among the species recorded in Itiyuro, 50.3% are categorized as rare species, 25.5% as occasional species and only 22% as dominant species (Table 3, Figure 3). On the other hand, 44% of the total number of species corresponded to species that were present in only one sampling, regardless of their abundance. Only 7 of the 31 species of cyanobacteria recorded in the reservoir were classified as dominant, the Raphidiopsis morphotype (*Raphidiopsis mediterranea* + *Raphidiopsis curvata*) stood out for its frequency

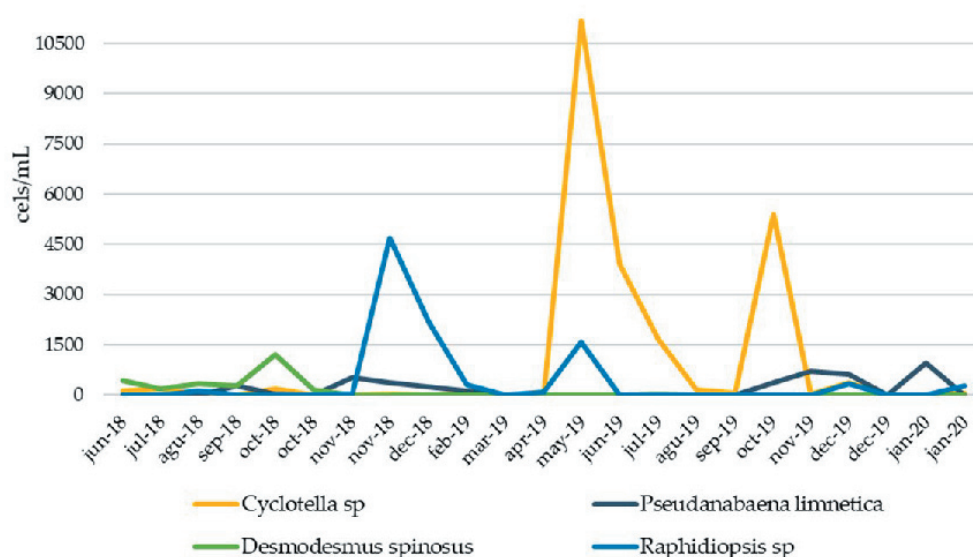


Figure 3. Abundances of some of the species classified as 'dominant' in Itiyuro, according to the Olmstead Tukey test.

Figura 3. Abundancia de algunas de las especies clasificadas como 'dominantes' en Itiyuro, según la prueba de Olmstead Tukey.

Table 3. Examples of dominant, constant, occasional and rare species in Itiyuro reservoir and Caraparí River, according to the Olmstead Tukey test.

Tabla 3. Ejemplos de especies dominantes, constantes, ocasionales y raras en el embalse Itiyuro y en el río Caraparí, según la prueba de Olmstead Tukey.

| Classification | Itiyuro Reservoir | Caraparí River |
|----------------|----------------------------------|--------------------------------|
| Dominant | <i>Aphanizomenon gracile</i> | <i>Nitzschia palea</i> |
| | <i>Pseudanabaena limnetica</i> | <i>Carteria multifilis</i> |
| | <i>Raphidiopsis curvata</i> | <i>Nitzschia reversa</i> |
| | <i>Raphidiopsis mediterranea</i> | <i>Nitzschia acicularis</i> |
| | <i>Monoraphidium contortum</i> | <i>Monoraphidium minutum</i> |
| | <i>Scenedesmus obtusus</i> | <i>Monoraphidium contortum</i> |
| | <i>Achnantidium minutissimum</i> | |
| | <i>Cyclotella meneghiniana</i> | |
| Constant | <i>Snowella litoralis</i> | <i>Anomooneis sphaerophora</i> |
| | <i>Ankistrodesmus falcatus</i> | |
| | <i>Nitzschia reversa</i> | |
| Occasional | <i>Aphanocapsa delicatissima</i> | <i>Desmodesmus spinosus</i> |
| | <i>Merismopedia elegans</i> | <i>Nitzschia acicularis</i> |
| | <i>Planktolyngbya limnetica</i> | <i>Monoraphidium caribeum</i> |
| | <i>Monoraphidium arcuatum</i> | <i>Scenedesmus obtusus</i> |
| | <i>Cyclotella stelligera</i> | |
| Rare | <i>Merismopedia punctata</i> | <i>Chlamydomonas elegans</i> |
| | <i>Microcystis aeruginosa</i> | <i>Pseudanabaena limnetica</i> |
| | <i>Coelastrum indicum</i> | <i>Lepocinclis ovum</i> |
| | <i>Hantzschia amphioxus</i> | <i>Tetraedron regulare</i> |
| | <i>Navicula capitatoradiata</i> | <i>Nitzschia dissipata</i> |
| | <i>Aphanotese zularinare</i> | |

as well as for being potentially toxic species. Bacillariophyta were represented mostly by 'rare' species, except for the genus *Cyclotella* that exhibited high frequency and a maximum peak of 11170 cells/mL (May 2019) (Figure 3).

Shannon diversity in Itiyuro ranged from 0.56 to 2.77 bits/ind., with an average of 1.79 bits/ind. The highest values corresponded to the warm season (spring and summer), while the lowest diversity was recorded in the cooler months (June to August). The effective number of species, weighting each one by its relative abundance, was 7 on average. Simpson's dominance had a mean estimate of 0.29, with very variable extremes (0.2 to 92% probability of extracting two individuals of the same species in a sampling), exceeding 70% probability in 65% of the samplings.

When we analyze the similarity of species in the reservoir between climatic seasons, it was observed that the community presented a similarity of 20% between spring 2018 and 2019. In the case of winter, the similarity was quite lower, reaching only 10% (again comparing the periods 18-19). When comparing winter and spring with each other, the similarity of the reservoir between seasons was on average 18%.

Structure and composition of phytoplankton in the Caraparí river

A total of 88 species and 51 genera were identified, 32 taxa were exclusive to the river. Most of taxa corresponded to the Bacillariophyceae class (50), followed by Chlorophytes (24) and Cyanobacteria (7). The average richness was 12 spp., with a peak of 32 spp. in September of the first year sampled. The average abundance for the entire period analyzed was 1246 cells/mL (± 1245 cells/mL), with a maximum of 4920 cells/mL in November 2019. Peak values for Chlorophyta (3400 cells/mL) and Bacillariophyta (1656 cells/mL) occurred in spring 2019. These values were explained in 73%, by the species *Carteria multifilis* - *Monoraphidium minutum* (in the case of Chlorophyta) and in 75% by *Nitzschia palea* (in the case of Bacillariophyta). In the Caraparí river, 51% corresponded to rare species, 25% to occasional species and 22.7% to dominant species. As in the Itiyuro reservoir, the constant species were the least abundant. Chlorophyll *a* concentration in the Caraparí River averaged a slightly higher value than in the reservoir with 29.69 $\mu\text{g/L}$ (± 45.9 $\mu\text{g/L}$) and a minimum of 1.01 $\mu\text{g/L}$, registered in July 2019.

Shannon diversity ranged from 0.74 to 2.77 bits/ind. with an average of 1.83 bits/ind. The

effective averaged number of species was 7, with a variation between 2 and 25. Dominance was slightly higher in Caraparí, with a minimum of 0.41 and a maximum of 0.91.

The highest alpha diversity was obtained in the reservoir (19 species on average) and the lowest in the river (13 species on average). The similarity between environments (river and reservoir) using Jaccard's index (based on the presence or absence of the species), showed a value of 22% on average for the total period considered, with a maximum of 39% in November 2019, being higher between both systems in winter (26%), in relation to spring (20%) and summer (17%).

Trophic state of the reservoir

Carlson's trophic state index (TSI) (Carlson 1977), based on chlorophyll *a* concentration, yielded values between 25 and 73. Stability was observed in the condition of the system, which remained mesotrophic (average chlorophyll *a* concentration 16.8 µg/L and average TSI=50.3) throughout most of the hydrological cycle, reaching the oligotrophic state in a sampling in winter 2018. The eutrophic condition was only observed in four samplings, in which Cyanobacteria dominated in April and December 2019 and Chlorophyta dominated in August and September 2019, according to the values established by OECD (1982). When applying the index based on the Secchi disk depth values, the system was classified as eutrophic, with general averages of TSI=74.8 and Secchi=0.71 m, even reaching the hypertrophic state on three occasions (October 2018, March and April 2019).

Canonical correlation analysis and principal component analysis

The value of the first canonical correlation was 0.92. The significance test indicated that only this canonical correlation was statistically significant ($P=0.0003$). The proportion of the total variance of the data explained by the first pair of canonical variables (first canonical correlation) was 84%. The construction of the first pair of canonical variables (C1-1=dependent; and C2-1=independent) based on the canonical (standardized) coefficients of the linear combinations, resulted in the following formulas.

Dependent canonical variable or criterion or endogenous variable:

$$C1 - 1 = 1.09 * (\text{total abundance}) - 0.3 * (\text{richness}) + 0.57 * (\text{Shannon})$$

Independent canonical variable or predictor or exogenous variable:

$$C2 - 1 = 0.36 * (\text{N-NO}_2) + 1.15 * (\text{N-NO}_3) + 0.57 * (\text{N-NH}_3) - 0.54 * (\text{SIN}) - 0.37 * (\text{P-SRP}) - 0.34 * (\text{N / P})$$

In the plot of the first canonical correlation, we can observe the spatial distribution of the samples obtained in the Itiyuro in the different months studied (Figure 4). There is an 'outlier' towards the positive part of both canonical variables corresponding to the sample collected in the month of January 2020, registering a cyanobacterial bloom with the species *Aphanocapsa delicatissima* and *Pseudanabaena catenata* in that sample.

Towards the positive part of the first canonical axis are the sites with the lowest phytoplankton abundance (with the exception of the site with bloom of cyanobacteria=65460 cells/mL) and the lowest species richness. Towards the negative part, the samples with the greatest richness and abundance are mostly located, with the predominance of low-water and transitions samples.

Towards the positive part of the canonical axis 2 are placed mostly the samples taken in the flood months, and also those that had the highest SIN values. On the contrary, in the negative part of the second component, the samples of low water and transitions are observed, corresponding to lower values of SIN and lower values of nitrite (N-NO₂).

On the other hand, if the behavior of the cyanobacteria between the phases of flooding, low water and transitions is observed, only the low water period was significantly different from the rest of the phases ($H=14.7$, $P=0.0021$). The averages of cyanobacteria in cells/mL were 11281 (± 23552) for floods, 2774 (± 3844) for TF-LW, 1554 (± 231) for TLW-F and 109 (± 114) in the low water. In the case of chlorophytes, these were significantly different only in the transition period from low water to flood ($H=14.09$, $P=0.028$), while Bacillariophyta ($H=4.05$, $P=0.2558$) and total phytoplankton ($H=7.73$, $P=0.0518$) did not show differences between phases.

In the principal components analysis (PCA) carried out with the limnological variables measured in the reservoir, the first three

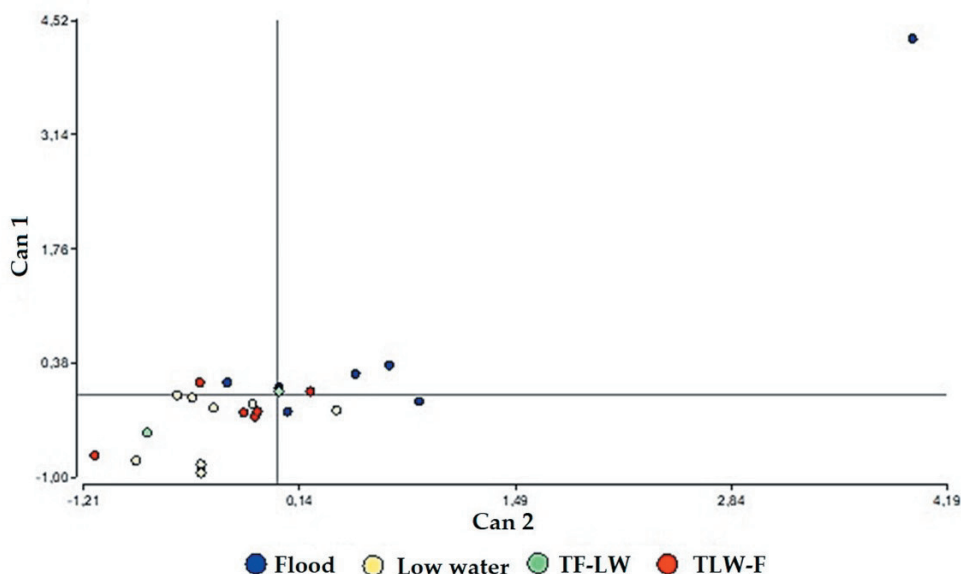


Figure 4. Canonical correlation analysis for Itiyuro. *TF-LW: flood-to-low water transition. TLW-F: transition from low to high water.

Figura 4. Análisis de correspondencia canónica para Itiyuro. TF-LW: transición de crecida a estiaje; TLW-F: transición de estiaje a crecida.

components explain 66% of the variability. The principal components are defined according to the following equations.

$$CP1 = -0.26 \text{ Ph} + 0.43 \text{ SIN} - 0.43 \text{ SRP} + 0.51 \text{ N/P} - 0.51 \text{ COD} + 0.24 \text{ Cla} - 0.47 \text{ SD}$$

$$CP2 = 0.49 \text{ Ph} - 0.15 \text{ SIN} - 0.27 \text{ SRP} - 0.28 \text{ N/P} - 0.65 \text{ COD} + 0.07 \text{ Cla} - 0.36 \text{ SD}$$

$$CP3 = -0.36 \text{ Ph} + 0.10 \text{ SIN} + 0.46 \text{ SRP} - 0.39 \text{ N/P} - 0.14 \text{ COS} + 0.65 \text{ Cla} - 0.18 \text{ SD}$$

Based on this analysis, a three-dimensional space can be defined where the sample units (SU) are distributed according to the values of the original variables, obtaining a scatter plot (Figure 5) that allows relating the values of the limnological variables with the algal and cyanobacteria dominance over the rest of the phytoplankton.

The SU with higher NIS and N/P values and lower transparency of the water column (Secchi disk) tend to be distributed towards the region of space defined by the positive values of PC1, corresponding mostly to those samples dominated by cyanobacteria. Analyzing the PC2 axis, it is observed that the variable with the highest positive weight corresponds to pH, while the one with the highest negative weight is COD; it is observed that the SU dominated by chlorophytes are located towards the positive region of PC2 and those dominated by Bacillariophyta towards the

negative values. Samples with higher biomass (expressed as chlorophyll *a*) and SRP values tended to cluster towards positive PC3 values. The only sampling that resulted dominated by chrysophytes (September 2018), recorded the highest COD value (350 mg O₂/L) of the entire cycle analyzed.

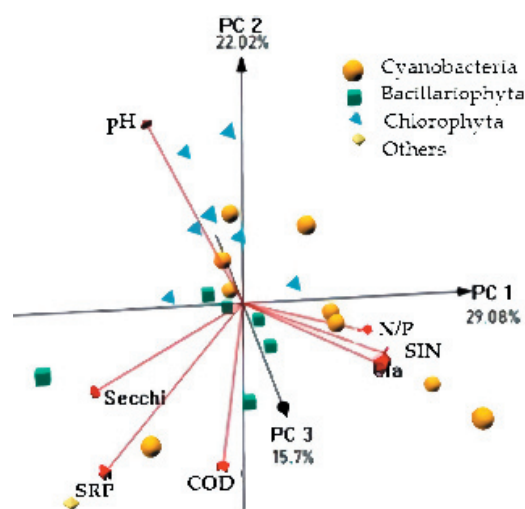


Figure 5. Principal component analysis in the Itiyuro reservoir (pH, N/P, SD, SRP, SIN, COD, Cla). With references, it is shown which group of the phytoplankton dominated in each sample.

Figura 5. Análisis de componentes principales para el embalse Itiyuro (pH, N/P, DS, PRS, NIS, DQO, Cla). Con referencias, se muestra que grupo del fitoplancton dominó cada muestreo.

DISCUSSION

The phytoplankton of the Itiyuro reservoir presented a considerable number of species, highlighting the richness of green algae over other phytoplankton groups, while in the Caraparí river, Bacillariophyta were the dominant ones. Previous studies carried out in rivers of northwestern Argentina, report a clear predominance of the Bacillariophyceae class, not only in relation to their abundances but also to richness (Seeligmann et al. 2001; Mirande and Traccana 2005; Martínez Demarco et al. 2011; Galea et al. 2014; Taboada et al. 2018). On the other hand, larger reservoirs in northwestern Argentina (Cabra Corral and El Tunal) that have been studied in the past were characterized by the dominance of Chlorophyta (Salusso and Moraña 2014), in agreement with that observed in Itiyuro. Reservoirs in Argentina generally present a mesotrophic state with a tendency to eutrophic (Bazán et al. 2005; Ame et al. 2010).

The Itiyuro reservoir is located close to the Limón reservoir, in the same basin and subjected to similar climatic conditions. However, the latter presents very frequent cyanobacterial blooms, even in winter (Vidaurre et al. 2018), while in Itiyuro the blooms are isolated events, limited to only one sampling during the period considered by this study.

The high sediment content that the Itiyuro reservoir receives from the Caraparí river could act as a limiting factor in the establishment and development of cyanobacteria, favoring other algal groups. On the other hand, the similarity between the phytoplankton composition of both environments was relatively low (22% average), especially in the case of cyanobacteria. The transfer of water from the Limón reservoir to the Itiyuro reservoir in times of water shortage to reinforce the supply of the population (especially in the months of greatest demand), could be identified as a point of cross-contamination, recognizing that Limón experiences recurrent blooms.

Cyanobacteria were dominant in only 9 of the 23 samples taken in the reservoir, and all the species recorded correspond to taxa with the capacity to produce and release cyanotoxins, a situation that in recent decades has been recognized as a serious problem in Argentine reservoirs (Aguilera et al. 2017; O'Farrell et al. 2019) and deserves special attention. The critical abundance, from which an event can be considered as a cyanobacterial bloom, is

delimited at 5000 cells/mL, and the criterion has been updated and related to the knowledge that we have about toxic cyanobacteria at regional level (Chorus 2001). According to this criterion, the Itiyuro reservoir presented a cyanobacterial bloom event during the period corresponding to sampling of summer of 2020.

Although numerous studies indicate that due to eutrophication and climate changes cyanobacterial blooms have increased in freshwater ecosystems (Paerl and Huisman 2009; Markensten et al. 2010), some authors (Kosten et al. 2012) agree that not all species will respond in the same way to environmental variations triggered by global warming in water bodies.

The increase in temperature would favor the predominance of nitrogen-fixing cyanobacteria (Mehner et al. 2010), which is in line with what was observed in the reservoir, with the species corresponding to the order Nostocales making the greatest contribution to richness. In relation to temperature, shallow reservoirs are generally characterized as polymictic due to the strong wind action, which causes continuous mixing of the water column, preventing a strong and persistent stratification, which favors species with high growth rates such as those of small dimensions (Scheffer 1998). Several phytoplankton genera are indicators of shallow water column depth and high nutrient concentrations. The most important are *Monoraphidium*, *Pandorina*, *Eudorina*, *Coelastrum*, *Golenkinia*, *Pediastrum*, *Scenedesmus* and *Cryptomonas* (Reynolds et al. 2002). With the exception of *Golenkinia* and *Eudorina*, all the others were described with high frequency in the Itiyuro reservoir.

The phytoplankton diversity of Itiyuro, was relatively low (1.79 bits/ind. average, 0.56-2.77 bits/ind.) in relation to the values found by Vidaurre et al. (2018) for the Limón reservoir (1.34-3.57 bits/ind.). On the other hand, the values corresponding to the Caraparí river were even lower at the entrance of the river to the dam. It would be expected that the entry impact of a water body with a lotic regime into another with a lentic regime would generate destabilization and alterations in the assemblages.

In lotic water bodies, variations in the composition and abundance of phytoplankton, in general, would be more representative of changes in their trophic state than a diversity index in itself (Stoermer and Smol 1999).

In Caraparí, the highest diversity values were recorded in winter and spring, when phytoplankton abundance was low (<700 ind./mL) compared to other samples. Several authors affirm that those communities subjected to disturbances of medium intensity and with a considerable frequency, are the ones that present a higher diversity (Holzman 1993; Reynolds 1993; Sommer et al. 1993). The entry of a lotic course with variable flows into the reservoir constitutes an important and intense disturbance on the phytoplankton community, among many things, due to the contribution of sediments and nutrients, which varies over time, in addition to the instability generated by the turbulence of the river itself in the water column (De León and Chalar 2003).

The Itiyuro reservoir constitutes an environment vulnerable to eutrophication and at risk due to the blooms of cyanobacteria that have been reported in this study. The similarity in the phycoflora between the reservoir and the river was low, especially in the contribution of cyanobacteria, which is important to consider in the management of the reservoir. The significant contribution of sediments from the Caraparí River is a morpho hydrological threat that requires stabilization and management at the basin level. The detection of cyanobacterial blooms with species with eco-toxicological potential requires periodic sampling that considers the possible release of toxins, as well as the development of management protocols.

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