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Zooplankton Assemblages of an Argentinean Saline Lake during Three Contrasting Hydroperiods and a Comparison with Hatching Experiments

Santiago Andrés Echaniz *, Alicia María Vignatti and Gabriela Cecilia Cabrera

Faculty of Exact and Natural Sciences, National University of La Pampa, Santa Rosa 6300, Argentina; aliciavignatti@exactas.unlpam.edu.ar (A.M.V.); gabrielacabrera@exactas.unlpam.edu.ar (G.C.C.)

* Correspondence: santiagoechaniz@exactas.unlpam.edu.ar

Abstract: Many saline lakes are temporary, with large variations in salinity, and their biota is adapted to withstand unfavorable periods. Utracan Lake, in a protected area in central Argentina, was studied on three occasions under different environmental conditions. In 2007, depth and salinity were 2 m and 33 g/L, and six species were recorded in the zooplankton. In 2009–2010, its maximum depth was 0.3 m, its salinity exceeded 230 g/L, and only *Artemia persimilis* was recorded. Field studies to compare the active zooplankton of a third period were combined with laboratory tests to ascertain the composition of the egg bank (flotation with sucrose) and zooplankton succession (hatching from sediments). In 2017–2018 (third period), the depth and salinity were 1.75 ± 0.17 m and 47.19 ± 11.40 g/L, respectively. Five species were recorded, and *A. persimilis* was found coexisting with cladocerans, copepods, and rotifers. *Brachionus plicatilis*, *Hexarthra fennica*, *Boeckella poopoenis*, *A. persimilis*, and a single specimen of *Moina eugeniae* were recorded in hatching experiments; however, the latter species was not recorded again. No cladoceran ephippia were recorded in the flotation tests. Salt accumulation on the sediments during the Utracan drought (2010–2016) would have deteriorated the ephippia. The register of *M. eugeniae* in 2017–2018 could be largely because of recolonization by waterfowl. The conservation of Utracan Lake is therefore advisable, and the same goes for other nearby saline lakes, which can act as sources of propagules that cross terrestrial areas through transport by wind or zoochory.



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Keywords: egg bank; hatching experiments; physicochemical parameters; *Artemia persimilis*; *Daphnia menucoensis*; *Boeckella poopoenis*

1. Introduction

Aquatic saline ecosystems, widespread in arid zones throughout the world [1], have salinities >3 g/L [2]. Their ecology has been less studied than freshwater lakes [3], despite representing approximately 44% of the total volume of inland waters and providing many ecosystem services such as the provision of inputs for industries, biotechnology, and aquaculture. They also represent important cultural, spiritual, recreation, tourism, and nature conservation sites [4]. They are currently receiving greater attention because they are sources of elements of high economic and strategic value, such as lithium [5].

Systems have been proposed to classify these lakes based on their salinity [2,4], but, in many cases, assigning a lake to any category is difficult due to large inter- and intra-annual variations in salinity [6]. These changes in water salinity are a major factor influencing biological diversity in saline lake ecosystems [4,7]. Therefore, food web shortening affects ecosystem functioning [1]. In the case of zooplankton, both richness and density have been shown to decrease as the concentration of dissolved solids increases [8–11].

Depending on their location, the topography, and the climate of their basins, many saline lakes are temporary [12] because they can remain dry for some time and fill and contain water (hydrophases) for variable periods [13,14]. The organisms that inhabit these

lakes have adaptations to withstand periods of environmental stress (due to a decrease in temperature, an increase in salinity, or desiccation) [14–18]. These adaptations include the production of dormant or diapausing states, as is the case with the resistance eggs of anostraca, notostraca, cladocerans, and rotifers [15,18–21], or the embryonic stage of the copepods [22–24]. The accumulation of these resistance structures in the lake sediments generates an egg bank, where they can remain viable for long periods and repopulate the lake when conditions become favorable again [17,25]. It has been previously reported that egg banks can contain a greater number of species than those that can be found at any time in active zooplanktonic communities [18] because they integrate structures produced at different times, both in an annual cycle and interannual periods [18,25–27].

The Dry Pampa region is in the center of the Arid Diagonal, which spans Argentina from north to south [28,29]. It contains numerous lakes of a wide range of salinities [11]. Most are in arheic or endorheic basins, without connection to water courses and, as they are fed by precipitation and groundwater discharges, are temporary, with hydrophases that last a few weeks or a few years and dry phases that can last several years [11]. The filling and drying of these lakes are strongly related to the short-term precipitation cycles determined by El Niño and La Niña events, and sometimes, the filling can be rapid, due to torrential precipitation, especially in summer [30]. As water losses typically occur through evaporation, drying can take several months during which pronounced changes in salinity may occur [11].

Utracan Lake is temporary and saline and is located in a protected area in the province of La Pampa (central Argentina) and is therefore currently subject to relatively low anthropogenic influence. It has previously been studied on different occasions and has shown a marked variation in its physicochemical parameters over time [31,32]. During 2007, after a humid period, it had a depth greater than 2 m, its salinity was around 33 g/L, part of its surface was covered with macrophytes, and cladocerans, copepods, and rotifers were recorded in its zooplankton [31]. A second study was carried out after a dry period, between May 2009 and August 2010, during which time it had a maximum depth of 0.3 m, which decreased until drying completely in September 2010. During this period, it had an average salinity greater than 230 g/L and only the anostracan *Artemia persimilis* (Piccinelli & Prosdocimi, 1968) was recorded [32].

Knowledge about the biological and geochemical dynamics of saline and hypersaline lakes remains incomplete [4], and the changes that Utracan Lake underwent caused it to behave like two different lakes. Therefore, the first objective of this study was to determine the lake's physical–chemical parameters and the composition of the active zooplanktonic community during a third period and compare the information with the two previous studies, in relation to the precipitation regime of the region.

Though it is useful to know the salinity thresholds at which species replacement occurs during the hydrophase of a lake, doing so through field studies is relatively difficult because high sampling frequencies are required. Since the determination of biodiversity in the egg bank allows for an exhaustive inventory of the species present and their abundance from a single set of sediment samples [33], the second objective was to ascertain the succession from the egg bank and the changes that occur in the zooplanktonic community with the increase in salinity through laboratory hatching assays.

2. Materials and Methods

2.1. Study Area

Utracan Lake (64°36' W, 37°17' S) (Figure 1) is in a protected area under municipal jurisdiction. It is fed especially by phreatic contributions from the surrounding dunes and to a lesser extent by direct precipitation and surface runoff. It is in an arheic basin, so water losses occur mainly through evaporation. It is in a region where the average annual rainfall measured between 1921 and 2023 in General Acha City (~10 km from Utracán Lake) is 563.6 ± 197.7 mm (www.policia.lapampa.gov.ar/contenidos/ver/lluvias, accessed on 8 May 2024) (Figure 2), with a wet season between September and April when more than 85%

of the precipitation falls [34]. The region is characterized by alternating periods of drought and rainfall, well below or above the historical average rainfall, especially associated with the ENSO phenomenon [30,35,36].

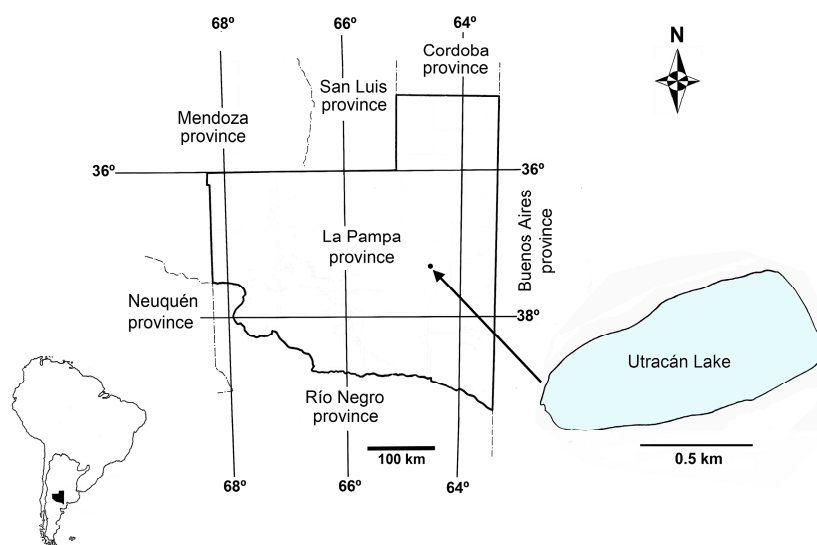


Figure 1. Geographic location of Utracán Lake.

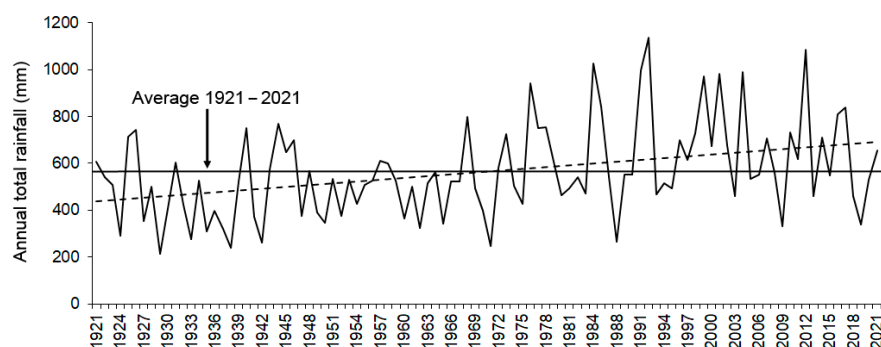


Figure 2. Average annual rainfall between 1921 and 2023 in the region where Utracán Lake is located, determined in General Acha City. Solid line: annual average. Dashed line: time trend.

Utracán is located in the *Espinal* (Thorny Forest) ecoregion [37]. It is surrounded by natural vegetation, with grasslands of *Hyalis argentea* D. Don. ex Hook. & Arn. and *Stipa brachychaeta* Godron and a native forest of *Neltuma caldenia* (Burkart) C.E. Hughes & G.P. Lewis and *Geoffroea decorticans* (Gillies ex Hook. & Arn.) Burkart. It has a rich variety of birdlife, including the Chilean flamingo (*Phoenicopterus chilensis* Molina, 1782) [31,32]. The Utracán watershed has semi-extensive livestock farming, and there is no direct influence of urban settlements [31,32].

2.2. Field and Laboratory Work

There were nine surveys between October 2017 and March 2019. The water temperature, the concentration of dissolved oxygen (Lutron[®] OD5510 oximeter, Lutron Electronic Enterprise, Co., Ltd., Taipei, Taiwan), the water transparency (Secchi disk, made by the researchers who wrote the article), and the pH (Corning[®] PS15 pH meter, Corning Inc., Port Louis, Mauritius) were recorded in situ, and water samples, which were kept refrigerated until analysis, were collected for physicochemical determinations. Zooplankton samples were taken with a 22 cm diameter, 0.04 mm mesh net.

Salinity (dissolved solid concentration) was determined gravimetrically by drying 50 mL of previously filtered water at 104 °C. Chlorophyll-a concentrations were measured

with a spectrophotometer after extractions with aqueous acetone to 90%. Suspended solids were determined with Microclar FFG047WPH fiberglass filters, dried at 103–105 °C until constant weight, and muffled at 550 °C [31,32]. Zooplankton counting was carried out under stereoscopic and conventional optical microscopes using Bogorov and Sedgwick-Rafter chambers, respectively [31,32].

In January 2017, after the lake went through a hypersaline period (salinity > 50 g/L), sediments were collected using cores at six sites in the lake. The sediment was homogenized, air dried in plastic recipients, and stored in a dark cold room at 4–5 °C for about 3 months until the hatching test was performed [27,38].

For the hatching experiments, a ~1 cm thick layer of sediment was placed in four 10 L glass aquaria. To simulate the increasing salt concentration by evaporation, we added from 12 g to 48 g of salts per aquarium from day 59. Salts were obtained from the same lake and were previously sterilized by drying them for 24 h at 160 °C. The aquaria were incubated for 204 days at 22 ± 1 °C with a photoperiod of 16/8 (light/dark). Zooplankton samples were taken from the aquaria by filtering 500 mL of water through a 40 µm mesh. Sampling frequency varied, from every two days at the beginning to biweekly at the end of the assay. Filtered water was placed back into the corresponding aquaria and their level was kept constant by adding demineralized water. When zooplankton samples were taken, we also measured water electrical conductivity (Hanna® HI 8733 conductivity meter, Hanna Instruments Inc., Woonsocket, RI, USA), dissolved oxygen concentration (Lutron® OD 5510 oximeter, Lutron Electronic Enterprise, Co., Ltd., Taipei, Taiwan), and pH (Corning® PS 15 pH meter, Corning Inc., Port Louis, Mauritius). The dissolved oxygen concentration values were corrected to account for the influence of salinity [39]. In order to compare electrical conductivity in the aquaria with salinity determined in field studies, salinity was calculated using the equation $S = C.F$ where S is salinity, C is electrical conductivity, and F is the conversion factor [40]. To determine the “ F ” factor (0.75), solutions of increasing concentration were prepared with the same salts used in the tests. Conductivity was then measured, and total dissolved solids were determined by drying at 104 °C.

To determine the presence of resistance structures in the sediments, especially cladoceran ephippia, the Onbe sugar flotation method was used. This method consists of mixing sediment with a solution of 1000 g of sucrose in 1 L of distiller water, centrifuging at 3000 rpm for 5 min, and observing the supernatant under a stereoscopic microscope [41–43].

We used the classification of continental waters based on salinity proposed by Hammer [2].

A Shapiro–Wilkins test was used to determine data normality. Due to the lack of normality, to examine relationships among environmental variables, and between these variables and zooplankton species, nonparametric correlation coefficients of Spearman (r_s) were used. A Kruskal–Wallis (H) test was performed to look for differences between the medians [44,45]. To determine any tendency to change in the annual precipitation of the region over time, the nonparametric Mann–Kendall test was used [46], using data obtained between 1921 and 2023 by the police of the province of La Pampa in the city of General Acha (www.policia.lapampa.gov.ar/contenidos/ver/lluvias, accessed on 8 May 2024). We used the Past 3 [46] and Infostat 2015p [47] software.

3. Results

3.1. Environmental Parameters

All parameters measured in the lake differed over the three sampling periods (Table 1), except for water temperature. During 2007, the lake depth ranged from 1.8 m to 2.2 m and remained relatively stable. Consequently, salinity showed little variation (29–37 g/L) [31]. During 2009–2010, the maximum depth (0.3 m) was recorded at the beginning of the study, when salinity was close to 155 g/L. Salinity reached a maximum greater than 420 g/L in February 2010, after which the lake dried up [32]. In 2017–2019, the maximum depth (2.02 m) was recorded in October, when salinity was 32.34 g/L. The depth decreased to 1.65 m in August 2018, when salinity was 48.81 g/L. Using all data, a correlation was found between both parameters ($r_s = -0.98$; $p < 0.05$).

Table 1. Physicochemical parameters determined in Utracan Lake during the three periods studied.

	2007 *	2009–2010 **	2017–2018	(H)
Depth (m)	2.00 ± 0.15	0.16 ± 0.09	1.75 ± 0.17	H = 22.43; $p < 0.05$
Salinity (g/L)	32.90 ± 2.70	238.05 ± 92.34	45.89 ± 10.74	H = 24.88; $p < 0.05$
Water temperature (°C)	16.08 ± 7.18	16.53 ± 7.61	18.14 ± 5.09	H = -0.12; $p > 0.05$
Transparency (m)	1.15 ± 0.29	0.11 ± 0.06	0.32 ± 0.11	H = 24.54; $p < 0.05$
pH	9.58 ± 0.13	9.21 ± 0.09	9.46 ± 0.16	H = 22.18; $p < 0.05$
Dissolved oxygen (mg/L)	10.03 ± 2.05	2.61 ± 1.64	8.22 ± 0.74	H = 22.34; $p < 0.05$
Chlorophyll- <i>a</i> (mg/m ⁻³)	1.22 ± 0.92	39.16 ± 22.69	4.23 ± 4.18	H = 23.42; $p < 0.05$
Inorg. susp. sol. (mg/L)	4.30 ± 3.77	1545.58 ± 1108.09	-	H = 16.65; $p < 0.05$
Org. susp. sol. (mg/L)	5.00 ± 1.89	387.65 ± 249.82	-	H = 15.62; $p < 0.05$

* [31]; ** [32].

The ionic composition of the water showed the predominance of Na⁺, which represented more than 90% of the cations across the three study periods, and Cl⁻, which represented between 42.3 and 51.4% of the total anions.

Water temperature followed a well-defined seasonal pattern. In the winter of 2007, it dropped to around 6 °C, and in the summer, it reached almost 27 °C. The range of variation was wider during 2009–2010, when the minimum was 3.4 °C, and the maximum exceeded 31 °C [31,32]. In the 2017–2019 study, sampling was carried out during months with higher temperatures, and the values were similar to those of the corresponding months of 2007.

Dissolved oxygen concentration in 2007 ranged between almost 7 mg/L and more than 14 mg/L, and in 2009–2010, between 0.11 and 5.97 mg/L [31,32]. In 2017–2019, the concentrations were similar to the corresponding months of 2007 but did not show any seasonal variation since no correlation was found between oxygen concentration and water temperature. Dissolved oxygen concentration, on the other hand, was inversely correlated to salinity ($r_s = -0.92$; $p < 0.05$).

The average water transparency and phytoplankton chlorophyll values were different between the three periods studied. During 2007, the transparency was greater than 1.1 m, with a maximum greater than 1.4 m (December), and a reduced concentration of chlorophyll and suspended solids, both organic and inorganic, was found [31]. During 2009–2010, transparency was around 0.10 m, the chlorophyll concentration was higher, close to 40 mg/m³, and the amounts of suspended solids were high [32] (Table 1). During 2017–2019, an intermediate situation was recorded, with transparency close to 0.3 m and low chlorophyll values (suspended solids were not determined).

3.2. Zooplankton

Seven species were recorded across the three study periods: one anostracan, two cladocerans, two copepods, and two rotifers (Table 2). In 2007, the highest richness (six species) was recorded in mid-autumn (May) and the lowest during the coldest months (June, July, and August), when three crustaceans but no rotifers were recorded. In this period, the calanoid *Boeckella poopoenis* Marsh, 1906 was the most common species (Table 2) [31]. In 2007, total density was highest in summer (February: 1548.7 ind/L) and lowest during autumn, when it dropped to a minimum of 166 ind/L (April). On all occasions, *B. poopoenis* accounted for the highest density. Cladocerans were recorded during the warmer months with *Moina eugeniae* Olivier, 1954 being the most abundant, with a maximum close to 190 ind/L in March. Rotifers were scarce and both species (Table 2) reached maximums close to 100 ind/L in summer. No significant correlation was found between zooplankton density and the environmental parameters analyzed during this period [31].

Table 2. The species recorded in zooplankton, the frequency of appearance in the samples (%), and the mean, minimum, and maximum density (ind/L) during the three periods studied in Utracan Lake.

	2007 *		2009–2010 **		2017–2018	
	Freq. (%)	Mean Density (ind /L)	Freq. (%)	Mean Density (ind /L)	Freq. (%)	Mean Density (ind /L)
<i>Artemia persimilis</i> Piccinelli & Prosdocimi, 1968	-	-	68.8	56.9 (0–399.3)	100	53.84 (2.4–148)
<i>Daphnia menucoensis</i> Paggi, 1996	41.7	0.36 (0–2.3)	-	-	-	-
<i>Moina eugeniae</i> Olivier, 1954	83.3	59.76 (0–187.8)	-	-	57.14	71.97 (0–419)
<i>Boeckella poopoensis</i> Marsh, 1906	100	113.1 (16–251.7)	-	-	85.71	193.76 (0–1224)
<i>Cletocamptus deitersi</i> (Richard, 1897)	83.3	1.94 (0–5.8)	-	-	-	-
<i>Brachionus plicatilis</i> Müller, 1786	41.7	9.96 (0–103.3)	-	-	71.43	8.93 (0–10.6)
<i>Hexarthra fennica</i> (Levander, 1892)	66.7	14.7 (0–101.7)	-	-	71.43	424.80 (0–2905)

* [31]; ** [32].

During 2009–2010, only *Artemia persimilis* was recorded (Table 2) (although absent between November 2009 and February 2010), reaching maximum densities in the autumn of both years, 180.75 ind/L in June 2009 and 399.3 ind/L in April 2010 when the naupliarian stages predominated in the population [32].

Between 2017 and 2018, five species were recorded (Table 2), all previously found in the lake, and maximum richness (all species) was recorded in February 2018. No correlation was found between total richness and the environmental parameters. Although the community maximum density was recorded in January 2018 (2980 ind/L), no correlation was found neither with the water temperature nor with any other variable. In this third period, *Artemia persimilis* and *B. poopoensis* were recorded in all samples, followed by the two rotifers, found in 72% of the samples. The species that reached the highest average density was *Hexarthra fennica* (Levander, 1892) (424.80 ± 1093.73 ind/L), which reached more than 2900 ind/L in January. It was followed by *B. poopoensis* (which, including naupliar stages, reached 353.57 ± 743.86 ind/L) and *M. eugeniae* (71.93 ± 154.47). Both species showed their maximum densities in late spring and early summer. Despite this, the correlation between the abundance of both species and the water temperature was not significant. However, a significant correlation ($r_s = 0.79$; $p < 0.05$) was found between the density of *M. eugeniae* and salinity, since this species stopped being recorded in the lake when the concentration of dissolved solids was close to 50 g/L. During this period, *A. persimilis* showed an average density of 53.84 ± 64.08 ind/L, an amount similar to that of 2009–2010. This species showed a peak of 148 ind/L in October 2017, and no correlations were found between its density and any environmental parameter.

3.3. Hatching Experiments

No differences were found between dissolved oxygen concentration, pH, or salinity determined in the treatments, so average values were used. Although dissolved oxygen concentration in the aquaria remained relatively stable at around 8.75 mg/L until day 95, it progressively decreased to 7 mg/L at the end of the assays (Figure 3A). A correlation was found between dissolved oxygen concentration and salinity ($r_s = -0.80$; $p < 0.05$). The pH decreased during the first few days and stabilized at values close to 9 starting on day 11. It decreased after the addition of salts (Figure 3A), and a negative correlation was found between both parameters ($r_s = -0.93$; $p < 0.05$). The mean salinity of the aquariums at the beginning (day 0) was 0.86 ± 0.18 g/L and increased to approximately 5.09 g/L on day 9. It then remained stable until day 59, when the addition of salts began, causing salinity to reach 65.08 ± 1.08 g/L on day 204, at the end of the tests (Figure 3B).

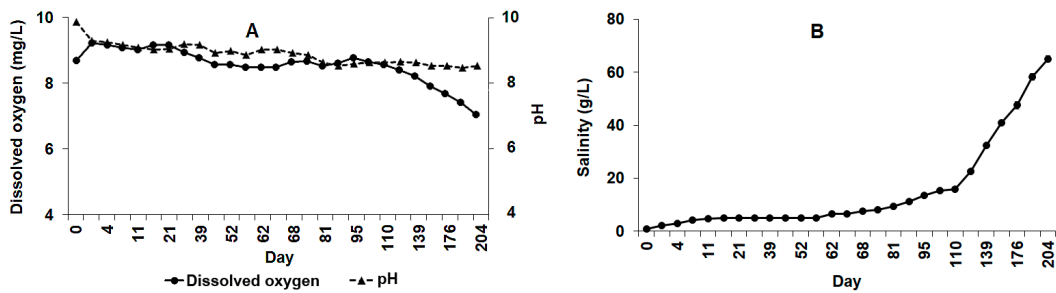


Figure 3. Variation in dissolved oxygen concentration and pH (A) and salinity (B) throughout the hatching bioassays from the sediment of Utracan Lake.

No differences were found between the richness and density of zooplankton in the different treatments. Throughout the experiment, only five taxa were registered: a cladoceran, a copepod, an anostrachus, and two rotifers (Figure 4). During the first month, no hatchings were recorded, but on day 32, when the salinity was 5.03 ± 0.07 g/L, *Brachionus plicatilis* Müller, 1786 and *Hexarthra fennica* were found with densities close to 1.5 ind/L. *Brachionus plicatilis* was recorded in all samples almost until the end of the experiments and reached high densities, with a peak of 922.5 ± 1494.2 ind/L on day 73 (Figure 5). *Hexarthra fennica* was not recorded in some samples, and its density remained low, with a maximum of 70.12 ± 133.37 ind/L on day 81 (Figure 5). On day 39, when salinity was 5.12 ± 0.06 g/L, nauplii of *B. poopoensis* were found with densities around 0.5 ind/L and showed a peak of 566.51 ± 1116.85 ind/L on day 52. After that day, their density decreased markedly until day 183 when they were not recorded again. From day 66, adults and copepodites of this species were found and their density increased towards the end of the tests, and from day 176, their density exceeded 100 ind/L (Figure 5). On day 124 (salinity: 22.59 ± 0.13 g/L), an *M. eugeniae* specimen was found, but the species was not recorded again (Figure 5). On day 73, specimens of *A. persimilis* were found when the salinity was 8.08 ± 0.21 g/L. Although it was registered until the end of the experiments, its density remained low, below 1 ind/L (Figure 5).

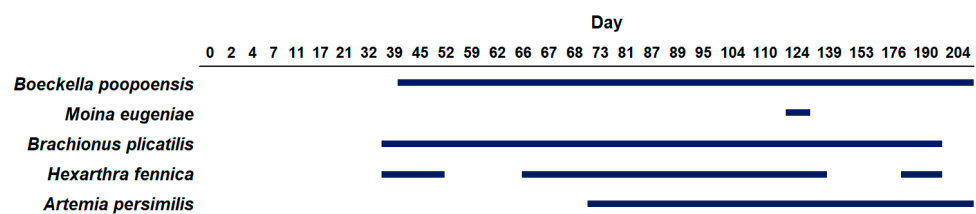


Figure 4. Species that hatched from the egg bank of the sediment of Utracan Lake and the periods during which they were recorded in the hatching bioassays.

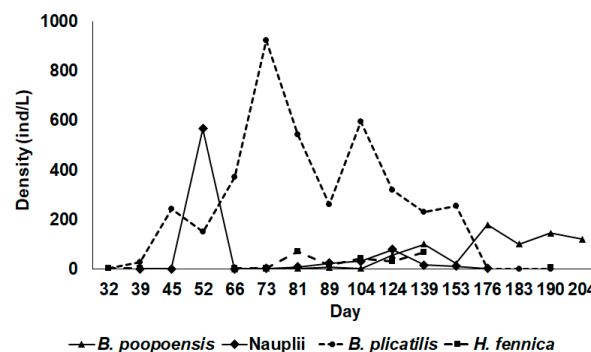


Figure 5. Variation in the average density of the three typical species of the mesosaline periods registered throughout the hatching bioassays from the sediment of Utracan Lake. *Boeckella poopoensis* includes copepodites and adults.

Contrary to expectations, no cladoceran ephippia were recorded in the sucrose flotation tests.

4. Discussion

From 1996 to 2007, a wet period was recorded in central Argentina, so the total annual rainfall in the region where Utracan Lake is located greatly exceeded the historical annual average (except in 2003 and 2005) (Figure 2). Thus, rainfall in 2007, the first period studied, caused the lake to have a depth greater than 2 m and an average salinity barely greater than 30 g/L. These conditions along with the high transparency of the water allowed the development of *Ruppia cirrhosa* (Petagna, Grande), a cosmopolitan rooted macrophyte, capable of tolerating large changes in salinity and water level [48,49], which covered almost 50% of the surface of Utracan [31]. At that time, the characteristic species association of mesosaline lakes in central Argentina was recorded in the zooplankton, which was composed of native halophilic crustaceans such as *D. menucoensis*, *M. eugeniae*, and *B. poopoensis* and euryhaline cosmopolitan rotifers as *Brachionus plicatilis* and *Hexarthra fennica* [11].

Unlike the aforementioned period, 2008 and 2009 were years with little rainfall (Figure 2), which caused the lake to decrease its depth to 0.3 m and the salinity to increase to more than 155 g/L. Although precipitation exceeded the historical average throughout 2010, this was not reflected in the lake's conditions, since its level decreased until it dried out after August [32]. This may be because most of the water entering Utracan is underground, and the water table would have dropped markedly during the previous dry period, losing connection with the basin. In this period, no macrophyte cover was developed and only *A. persimilis*, the only native species of the genus in Argentina (and also found in other hypersaline lakes of La Pampa), was recorded in the zooplankton of Utracan [32].

Starting in 2011, a new wet cycle developed, and annual rainfall once again exceeded the historical average (Figure 2), so when the third study period began, in October 2017, the lake had similar conditions to those of 2007, especially with depth and salinity. During this period, most of the sampling was carried out in spring or summer, so the mean temperature was slightly higher than in previous periods, which included complete annual cycles. In this third period, no macrophytes were recorded and differences were found in zooplankton. Neither *D. menucoensis* nor *Cletocamptus deitersi* (Richard 1897) were recorded; however, *A. persimilis* was found on all occasions. Most of the correlations calculated between the densities of the different species and the environmental variables were non-significant. However, the less tolerant *M. eugeniae* showed negative densities with a salinity increase (it was not recorded when this parameter exceeded 50 g/L), and rotifers showed lower densities when temperatures dropped below 12 °C.

The coexistence of *A. persimilis* with other species is of interest, since in most previous cases, in high-salinity lakes of La Pampa, this species had been found alone [50] or coexisting only with *B. poopoensis* in lakes of salinities of up to 110 g/L [51], as found in southern Chile [52]. In 2017, it was recorded in Utracan in coexistence with *M. eugeniae*, *H. fennica*, and *B. plicatilis*, a situation similar to that recorded by Pilati et al. [53] during a period of meromixis in another saline lake in La Pampa. These authors found that *A. persimilis* coexisted with *B. poopoensis*, *C. deitersi*, *D. menucoensis*, and *Metacyclops mendocinus* Wierzejski, 1892 in the zooplankton of the low-salinity surface layer (mixolimnion). However, the disappearance of less tolerant taxa, especially cladocerans, when the lake lost stratification and salinity exceeded 45 g/L suggests that this coexistence is modulated and limited by fluctuations in salinity [52].

Regarding the hatching tests, the increasing salinity in the water until approximately day 9 (5.09 g/L) was likely produced by the resolubilization of the salts present in the sediments. That value remained relatively stable until day 59 when salt addition began. This also affected dissolved oxygen concentrations, which decreased as salinity increased. The first hatchings were recorded a month after the tests began and were limited to the appearance of rotifers. This contrasted with the results of hatching assays on sediments

from a subsaline lake in La Pampa, in which cyclopoid copepodites were found at 48 h after initiation (Vignatti et al., unpublished data). The high number of *B. poopoensis* nauplii recorded in the aquariums on day 52 could have been due to a massive emergence of nauplii from the sediments, which would have remained viable due to the high tolerance of this species to salinity. The fact that reproductive adult specimens were not recorded until days 66–73 is probably because this species takes between 13 and 22 days to reach maturity in temperatures similar to those used in these trials [54]. The delay in *A. persimilis* hatching (almost 70 days when the salinity was close to 8 g/L), is an aspect that should be explored in depth, in order to ascertain whether or not this is the minimum value at which this species can hatch, or if it is due to combinations of other environmental factors.

The absence of *C. deitersi* from the hatching tests could be due to the fact that they are typically benthic. Their appearance in the water column in shallow lakes field sampling is usually the result of wind mixing, an effect that was not simulated in the bioassays.

All the species that hatched in the aquaria were halotolerant, typical of the zooplanktonic associations recorded in Utracan in previous hydroperiods. Although the cladocerans *D. menucoensis* and *M. eugeniae* had been recorded in natural conditions in the lake, only a single specimen of the latter species was recorded in the tests. This situation coincides with that recorded by Pilati et al. [55], who, when carrying out hatching tests at low salinity, did not find cladocerans that had been present in the active community of the saline lake from where sediments were collected (Parque Luro East Lake). Taking into account the fact that Utracan dried up between 2007 and 2017, the accumulation of a large amount of salts on the sediments could have damaged the ephippia, inhibiting their hatching, a situation similar to that recorded by Shadrin et al. in hypersaline lakes of the Crimea and whose causes are not well known [56]. Moreover, sugar flotation tests and observations under a stereoscopic microscope showed only *A. persimilis* eggs and, despite what was expected, a total absence of ephippia in the sediments collected in January 2017. However, at the beginning of the third sampling period (October 2017), an established population of *M. eugeniae* was recorded, which remained at high densities until February 2018, when salinity exceeded 50 g/L. The absence of ephippia in the sediments, coupled with the finding of *M. eugeniae* in October 2017, might suggest that zoochory, especially by aquatic birds [15,57], plays an important role in the recolonization of Utracan by this and other species, such as *D. menucoensis*. This slight discrepancy between the records of the zooplankton in the hydrophases and egg bank is an aspect that should be studied further since it relativizes the usefulness of emergency tests compared to field studies to determine the diversity of saline lakes. It has been proven that, in the case of rotifers, only 80% of the species are recorded by both methods. This could be due to the small size of sediment samples, inadequate hatching conditions, or the fact that the obligatory diapause periods for some species are longer than the duration of the bioassays [33].

Although Utracan is in a protected area, there is no management and monitoring plan to ensure its correct management. In the past, salt exploitation was performed, but currently, the main ecosystem services of the lake and the surrounding area are related to recreation and tourism. Given that abundant birdlife is also recorded in Utracan, it is a place where educational activity, bird observation, and nature photography are taking place. Thus, knowing more about its ecology will benefit the formulation of effective protection strategies against growing natural and anthropogenic threats [4]. Furthermore, the adequate conservation of other nearby saline lakes, currently not officially protected, would also be advisable, because they can be the source of propagules of zooplankton species that cross the “dry ocean” through transport by wind or animals [15].

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