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Limnological Characteristics and Rotifer Community Composition of Northern Mexico Chihuahuan Desert Springs

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Abstract

Desert springs are critical for wildlife and human subsistence, but poorly studied. The springs in the northern Chihuahuan Desert region of Mexico are no exception. While groundwater overexploitation and other anthropogenic activities have endangered these waters, little is known about their water quality or biodiversity. We monitored five springs on a monthly basis for one year, and two others sporadically, to determine their physicochemical characteristics and to study rotifer community composition as an assessment of biodiversity. Seasonal variation in temperature, dissolved oxygen (DO), and conductivity was observed in the five springs. DO was occasionally below the criterion for aquatic life (<4mg/L) in some springs. Ca²⁺ (468-775.2 mg/L) was the dominant ion in all springs except Ojo de Santa María where Na⁺ (180-290 mg/L) dominated. Of 57 rotifer species identified, *Lepadella patella* (Müller, 1786), *Lepadella triptera* (Ehrenberg, 1830), and *Philodina megalotrocha*, Ehrenberg, 1832 were present in most springs. The rotifer community exhibited nestedness among the sites (supported by four null modes, $p < 0.05$) and was correlated with fall season, DO, and nitrate concentration. This work provides information that will aid in management practices and provide important baseline data on water quality and regional diversity of a major component of the zooplankton.

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Keywords

Arid land springs; Limnology; Nestedness analysis; Water quality

1. Introduction

Springs are important water sources for many human cultures and wildlife (Kreamer et al., 2015; Blinn, 2008; Stevens & Meretsky, 2008). Yet they are among the least studied and poorly protected ecosystems on Earth (Kreamer et al., 2015; Stevens & Meretsky, 2008). The springs of the Chihuahuan Desert (CD) are no exception (Alcocer & Bernal-Brooks, 2010). The CD is an arid/semiarid region of 629,000 km² with low annual precipitation (150–400 mm) that extends from northern Mexico into southwestern USA. This large region comprises a rich array of geographical features including flat lands, sand dunes, canyons, and arroyos, as well as a variety of permanent and temporary aquatic habitats (SEMARNAT, 2008; Goettsch & Hernández, 2006). The most well characterized aquatic systems of the CD are those located in Cuatro Ciénegas, a natural protected area of 1500 km² in México. Here groundwater-fed springs have been disturbed by anthropogenic activities such as groundwater mining and conversion of land from desert to agriculture. This situation is likely representative of other springs in the Coahuila region (Hendrickson et al., 2008; Badino et al., 2004). Consequently water quantity and quality have decreased, impacting their natural communities (Goudie & Seely, 2011; Alcocer & Bernal-Brooks, 2010).

The few studies on ground and surface water quality in the CD focused primarily on the Río Bravo and Conchos (e.g., Gutiérrez et al., 2008; Ríos et al., 2004); indeed detailed surveys of arid land waters in Mexico are rare, particularly for CD aquatic systems (Carson & Masa-Benignos, 2014; CNA, 2011; Alcocer & Bernal-Brooks, 2010; Hendrickson et al., 2008; Walsh et al., 2008; INE, 2007). Water scarcity and multiple geographical features often result in high diversity and endemism of biota in arid land springs (Unmack & Minckley, 2008). This is evident in the CD, where endemism in vertebrates is particularly high: 47% of the amphibians and 50% of fishes are categorized as endemic (Carson & Mesa-Benignos, 2014; SEMARNAT, 2008) and several endemic springsnails and amphipods have been described (Hershler et al., 2011; Gervasio et al., 2004). However, few studies have characterized biotic diversity of zooplankton in CD springs (Walsh et al., 2014a, b; Suárez-Morales et al., 2010; Walsh et al., 2008; Stevens & Meretsky, 2008; Wallace et al., 2005). These studies indicate high levels of diversity tied to local water chemistries. For instance, Wallace et al. (2008) reported that about 12% of all described rotifer species have been observed in this ecoregion. Walsh et al. (2008) found that species assemblages were strongly correlated with conductivity. Suárez-Morales et al. (2010) determined the biogeographic origin of 32 copepods species found in CD aquatic systems, but they did not evaluate how this diversity was related to chemical characteristics of the habitats.

To improve management and conservation practices of desert springs, additional information is needed, especially for those in protected areas. Such data should include both basic limnological parameters, as well as data on the biodiversity of key taxa (Carson & Masa-Benignos, 2014; Alcocer & Bernal-Brooks, 2010; de la Federación, 2009; Springer et al.,

2008). Here we report the physicochemical characteristics and rotifer community composition of seven springs located in the Samalayuca region in northern Chihuahua, Mexico. This information is required to (1) evaluate anthropogenic impacts on springs, (2) fill knowledge gaps of desert spring limnology and biodiversity at regional scales, (3) provide more fine scale data on spring classification, and (4) establish baseline biotic inventories for future monitoring, thus establishing assessment of overall ecosystem health (Stevens & Meretsky, 2008).

2. Methods

Five springs (Ojos Altos A, Ojos Altos B, and Ojos Altos C [GPS coordinates: 31.40685 N; -107.6181833 W], Ojo de En Medio [31.37885 N; -106.5877833 W]; and Ojo de La Punta [31.3859166 N; -106.6022666W]) were monitored monthly for one year during 2007 to 2009. Two other springs, Ojo de Santa María (31.1552777 N; -107.3172222 W) and Ojo de La Casa (31.3656166 N; -106.5322333W) were sampled intermittently from 2009 to 2011. Springs and sampling regimes were selected based on geographical location, accessibility, and security concerns. The topography of the Samalayuca region consists mostly of alluvial and lacustrine plains with small sparse mountains “like islands on the sea” (CONANP, 2013), such as La Sierra de Samalayuca. These plains are characterized by sand dunes in the natural protected area of Médanos de Samalayuca (declared Área de Protección de Flora y Fauna on 2009). This region is part of the Northern Closed Basins, which includes parts of three river basins (Casas Grandes, Santa María, and del Carmen) and the Samalayuca Desert. This region is endorheic: localized surface water flows with in arroyos and flood plains only during heavy storms (CONANP, 2013; Alcocer & Bernal-Brooks, 2010). Ojos Altos and Ojo de Santa María are located at the north and southeastern portions of Samalayuca region, respectively (Figure 1 and 2). Ojo de La Casa, Ojo de En Medio, and Ojo de La Punta are located about 7 km northeast of the town of Samalayuca. Annual precipitation is about 1600 mm, most of which occurs from July to September. Minimum and maximum temperatures recorded are -16° and 41.2°C . Due to high temperatures, there is a water deficit of about 10 times the average annual precipitation: e.g., the mean evaporation is $\sim 2,420$ mm (CONANP, 2013).

2.1. Water Quality Analyses

Our monitoring protocol included morphological assessment, water quality analyses, and determination of rotifer community composition. Maximum length, width and depth of springs were measured monthly to identify seasonal changes in volume (triangulation method; Wetzel & Likens, 1991).

Water quality analyses included determination of physicochemical parameters, nutrients, and ions related to salt content (Table 1). Water temperature (T), conductivity (EC), and dissolved oxygen (DO) were measured at each site using a pre-calibrated YSI 556 multi-parameter system. Grab samples for water chemistry analyses were collected according to Mexican regulatory procedures (PROY-NMX-AA-121-SCFI-2005; Secretaría de Economía, 2005). Three grab samples were collected at each site (~ 30 cm below the surface) using a cup sampler. Samples were poured into clean plastic containers and preserved with

concentrated H₂SO₄ for total phosphate and nitrogen (inorganic and organic) or HNO₃ (pH 2) for metal (Ca, K, Na, Mn, Fe, Zn) determination.

Samples were placed on ice for transport and stored at 4°C until laboratory analysis (APHA, 1992).

Chemical parameters were determined by titration (APHA 2330B/total hardness, Ca; APHA 4500/ total nitrogen-Kjeldhal) and UV/Visible spectrometry (Perkin Elmer Lambda 2; APHA 4500C/NO₃⁻ and PO₄⁻²; APHA 4500E/SO₄⁻²). Other metals (K⁺ (766.5 nm), Na⁺ (330.2 nm), Cu²⁺ (324.2 nm), Mn (279.5 nm), and Fe (248.3 nm)) were determined using flame atomic absorption spectroscopy (FAAS: Perkin Elmer spectrophotometer model 3110; APHA, 1992). Appropriate standards were prepared to set metal calibration curves at the 2-80 mg/L range for Na⁺ and at 0.05-1.0 mg/L for Cu²⁺, Mn, and Fe.

Quality control procedures were followed for sampling, field and laboratory equipment calibration, and experimental procedures (Mexican regulation: NMX-EC-17025-IMNC-2006; de la Federación, 2006). Detection limits were determined for nutrients and ions related to salinity: SO₄⁻² (0.048 mg/L), NO₃⁻ (0.016 mg/L), P (0.006 mg/L), Fe (0.02 mg/L), Mn (0.01 mg/L), Cu²⁺ (0.01 mg/L), K⁺ (0.99 mg/l), Na⁺ (0.86 mg/L). Calibration curves for nutrients and ions all had correlation coefficients of (r^2) 0.997.

Because weather patterns in the northern region of Chihuahua, Mexico are characterized by extreme fluctuations in temperature (0 to 40°C; INE, 2007), data collected for each spring were analyzed using Analysis of Variance (one or two factor ANOVA) to determine the significance of seasonal changes in water quality in Ojos Altos A-C, Ojo de En Medio, and Ojo de La Punta. Means were compared using a Tukey *post-hoc* test with a 95% confidence level. Statistical analyses were not conducted for the other two springs since monitoring was irregular.

2.2 Rotifer Community Composition

Two types of samples were collected for rotifer species identification: water and aquatic vegetation. Two water samples were collected by towing plankton net (64 µm) 10 times horizontally through the water column (EPA, 1982). Duplicate samples were divided: one was preserved (70% EtOH or 4% buffered formalin); the other was kept at 4°C. Rotifers were identified from live and preserved material under a Zeiss Axioscope using appropriate taxonomic keys (e.g., Wallace et al., 2006; de Paggi, 2002; De Smet & Pourriot 1997; Segers, 1995).

A Redundancy Analysis (RDA) was implemented to investigate the relationship between physical and chemical parameters and rotifer species richness (*S*) in the habitats following Walsh et al. (2008) in CANOCO5 (ter Braak and Šmilauer, 2012).

We followed the methods of Meksuwan et al. (2014) to test the hypothesis that rotifers inhabiting our study sites are a nested subset of those sites with larger species assemblages. Our data matrix was constructed with presence-absence data of species (*S*) and habitat (*H*). The matrix was then rearranged (packed) using the computer program ANINHADO (Guimarães & Guimarães, 2006), with the outcome being the most concentrated grouping of

S and H (Ulrich et al., 2009). Although Ulrich et al. (2009) notes that several metrics have been applied to evaluate nestedness, we abridged our analyses by using the same four nestedness metrics as used by Meksuwan et al. (2014). These include the following: (1) Matrix temperature (T°); (2) Discrepancy shifts (DS); (3) Species segregation (SS); and (4) Spatial turnover (ST) (i.e., percent singletons). Where T is a metric that indicates the level of compactness of the packed matrix; DS counts the number of unexplained presences (= outliers); SS , also known as anti-nestedness, counts the number of reciprocal exclusions in all 2×2 sections of the matrix (Almeida-Neto et al. 2008). Using the Guild Ratio as described by Obersteiner et al. (2011) we assessed the frequency of SS pairings with feeding guilds (i.e., microphagous and raptorial). ST is the number of taxa associated with only one habitat, which may simply mean rarity or it could indicate habitat specific requirements (Baselga, 2010). However, we also recognize that insufficient sampling may be a factor in the incidence of singletons. We tested the significance of the packed matrix against four null models with 1000 simulations for each null model (Meksuwan et al., 2014).

3. Results

3.1. Spring Morphology

All springs sampled are shallow systems (<0.8 m depth). The estimated water volume ranged from 1.8 to 6.6 m³ in Ojos Altos, 0 to 37.3 m³ in Ojo de En Medio and 90.8 to 225.6 m³ in Ojo de La Punta. Volume varied by season among springs (Ojos Altos, ANOVA, $F=4.06$, $p=0.05$; Ojo de En Medio, Ojo de La Punta ($F=3.61$, $p=0.04$). A significant decrease in volume occurred in Ojos Altos B between spring and summer ($HSD=2.72$, $p<0.05$), and from fall to winter in Ojo de En Medio and Ojo de La Punta ($HSD=27.2$, $p<0.05$; Table 2). Although morphology in Ojos Altos A and C varied through time, volume changes did not differ ($p>0.05$).

3.2. Water Quality Monitoring

Physicochemical parameters varied through season and/or among springs. Seasonal fluctuations were detected in temperature in Ojos Altos A-C (ANOVA, $F=19.06$, $p=0.0001$; $HSD=3.12$, $p<0.05$), Ojo de En Medio and Ojo de La Punta (ANOVA, $F=18.04$, $p=0.0001$; $HSD=5.33$, $p<0.05$; Fig. 3A), and DO in Ojo de En Medio and Ojo de La Punta (ANOVA, $F=5.47$, $p=0.003$; $HSD=3.05$, $p<0.05$; Fig. 3B). Hypoxic episodes (<2-3 mg O₂/L) occurred in Ojo de En Medio (2mg/L) and Ojo de La Punta (0.9 mg/L) during the summer (Table 3), and for most of the year in Ojos Altos A and Ojos Altos C (Fig. 3B). Ojos Altos B was oxygen saturated most of the time, while Ojo de Santa María was supersaturated. Due to intermittently sampling, insufficient data from Ojo de Santa María was available to determine patterns in DO concentration (Table 3). Significant variations were also observed on Na⁺ in Ojo de En Medio and Ojo de La Punta (ANOVA, $F=3.31$, $p=0.03$) and sulfate concentrations in Ojos Altos A-C (ANOVA, $F=5.45$, $p=0.003$). Sodium level decreased (18%) in Ojo de La Punta from summer to fall but increased (74%) in Ojo de En Medio ($F=3.31$, $p=0.03$; Fig. 3D), while sulfate concentration reached 2554, 2072 and 2177 mg/L in summer and dropped to 1142, 1313, and 1215 mg/L in winter, respectively in Ojos Altos A-C ($HSD=751.13$, $p<0.05$; Fig 4). Insufficient data from Ojo de En Medio and Ojo de La Punta were collected for statistical analysis.

Other ions and nutrients were found at low levels in all of the springs (<5 mg/L; Table 4):

1mg/L of Mn and Cu were detected; phosphate content (0.09-1.1mg/L), inorganic N (0.3-5.0 mg/L), and organic N (0.07-1.3 mg/L) concentrations were similar among springs. The highest level of Mn was observed in Ojos Altos A ($F=33.34$, $p=0.001$). No significant differences were found in other nutrient concentrations among Ojos Altos springs.

Differences in pH, conductivity, and cation concentrations were detected among sites.

Values of pH varied significantly between Ojos Altos A (6.8) and Ojos Altos B (7.2; ANOVA $F=3.35$; $p=0.047$; HSD=0.44, $p<0.05$), as well as conductivity: 2.9 mS/cm³ and 2.5 mS/cm³, respectively (see Fig. 3E; ANOVA, $F=73.89$, $p<0.0001$; HSD=0.09, $P<0.01$).

Sodium (70.25-126.25 mg/L vs 70.25-97.13 mg/L; $F=7.19$, $p=0.002$; HSD=20.58 $P<0.01$) and K concentration (150.62-203 mg/L vs 94.83-138.2 mg/L; ANOVA, $F=15.53$, $p=0.0002$; HSD =24.28, $p<0.01$) were higher in Ojos Altos A than in Ojos Altos B and Ojos Altos C (Fig. 3D-F). In comparison, pH (6.6) and Na⁺ content (59.8 mg/L; Fig. 3D) in Ojo de En Medio were lower than in Ojo de La Punta (7.9 and 107.2 mg/L, respectively) ($F=6.07$, $p=0.005$; HSD=1.15, $p<0.05$ and $F=31.23$, $p=0.0001$; HSD=30.29, $p<0.01$).

Even though data from Ojo de Santa María was not statistically analyzed, it is important to note that its conductivity values were low (0.9-1.0 mS/cm³; Table 3). Moreover, Na⁺, not Ca²⁺, was the dominant cation in this spring (180-290 mg/L).

3.3. Rotifer Species Richness

Of the 57 rotifer taxa (2 genera) identified (Table 5 & 6), ~ 30% have been found in other CD aquatic systems (Walsh et al., 2014a; Walsh et al., 2008; Wallace et al., 2005; Wallace et al., 2008). *Lepadella patella* (Müller, 1773) was found in all springs. Among these springs, unique species were identified in all with the exception of Ojos Altos A and Ojo de La Casa. Ojo de La Punta contained the highest S and most unique species (Table 5). Patterns of S through the monitored period are shown in figure 2G. RDA showed that only 22% of species variation (axis 1: 9%, axis 2: 6% was explained by environmental factors ($F=0.7$, $p=0.056$). Most species are positively associated with increasing DO, nitrate concentration, and the fall season. In addition, several species were associated with increased depth (*Lecane hornemanni* (Ehrenberg, 1834), *Cephalodella sterea* (Gosse, 1887) and unidentified bdelloids; Fig. 5, see Discussion).

The combined rotifer community of these seven springs demonstrated nestedness ($T = 21.9^\circ$; Figure 6) with support from all four null models tested, as defined by ANINHADO (Guimarães & Guimarães, 2006). The S of these habitats varied widely, 2-29, with a mean S per site (mean α diversity =) = 15.3. The β diversity ($\beta_p = \gamma /$) was 3.7. Nestedness of species within these spring systems indicates that smaller species assemblages were composed of non-random nested subsets of communities with greater species richness. If these springs were not nested, species would be assembled randomly among the sites.

Although all four null models supported this dataset, there are a large number of discrepancy shifts (DS) for species ($n = 39$). There also is a large number ($n=23$) of reciprocal exclusions (SS) of species in the packed matrix. Of these exclusions 47.8% were of taxa with the same within a feeding guild; that is, both species were raptorial or both microphagous: e.g.,

Testudinella patina (Hermann, 1783) versus *Lecane undulata* Hauer, 1938. The other pairings crossed guild lines: e.g., raptorial: *Resticula melandoca* (Goose, 1887)) versus microphagous: *Platyias quadricornis* (Ehrenberg, 1832).

As illustrated by the high number of singletons ($n = 32$, i.e., 56% of the species pool of these springs), this assemblage also showed high species turnover (*ST*). This change in community assembly probably reflects the idiosyncratic needs of the species. However, there was no obvious pattern in their feeding type (e.g., 17 microphagous v. 15 raptorial species).

Nine species had unusually high idiosyncratic T° defined as being greater than > 1 SD or $2>$ SD of the mean matrix temperature. Those with > 1 SD were all microphagous: i.e., *Cephalodella cf. physalis* Myers, 1924, *Lecane cornuta* (Müller, 1786), *Lecane inermis* (Bryce, 1892), *Lecane quadridentata* (Ehrenberg, 1830), and *Lecane undulata* Hauer, 1938. Of those with > 2 SD, two were microphagous (*Cephalodella cf. graciosa* (Wulfert, 1956) and *Cephalodella megalcephala* (Glascott, 1893)) and two raptorial (*Pleurotrocha petromyzon* Ehrenberg, 1830, and *Pleurotrocha sigmoidea* Skorikov, 1896).

4. Discussion

Spring water quality is determined in part by the origin of the system, temperature regime, geochemical processes, surrounding vegetation, and human impacts. In terms of origin, Ojos Altos C is likely geothermal (mesothermal; Springer et al., 2008; Alfaro and Wallace, 1994) while the other monitored springs were hypothermal ($< 35^\circ\text{C}$), similar to Poza Azul (30°C ; CC: Cuatro Ciénegas, Coahuila, Mexico), Media Luna (28.3°C ; SLP: San Luís Potosí, Mexico), and San Salomon (25.4°C ; Texas; Hendrickson et al., 2008; Walsh et al., unpublished data). Water properties of the Samalayuca springs, such as pH and conductivity, are likely related to the dissolution of calcareous rocks or other natural soil components: neutral and moderately basic pH (6.5-10) are a consequence of carbonate ions release by water (Springer et al., 2008; Hem, 1989; Horne and Goldman, 1994). Neutral pH values (6.5-7.6) have also recorded in numerous other CD springs in the U.S.A and Mexico (Hendrickson et al., 2008; Blinn, 2008; Walsh et al., unpublished data) and other arid regions of the world (Al-Taani et al., 2012; Kaya et al., 2010). The difference in degree of mineralization (conductivity) between Ojo de En Medio and Ojo de La Punta, and Ojos Altos may be related to their origin (i.e., the Mimbres Bolson which underlies Ojos Altos A and B), evaporative processes, soil leaching, and/or mineral dissolution (gypsum, calcite, and dolomite) from soil (CONANP, 2013; Springer et al., 2008; INE, 2007; APHA, 1992). Based on Hammer's classification, Ojos Altos springs are subsaline systems, as are most other CD springs (Hendrickson et al., 2008; Blinn, 2008; Ortega et al, 1992; Walsh et al., 2008). However, concentration of Ca^{2+} in the monitored springs (90.9-680.4 mg/L; Fig. 3H) was higher than those reported for semiarid aquatic systems at Cuatro Ciénegas (< 357 mg/L; Hendrickson et al., 2008), semiarid coastal permanent (< 189.6 mg/L), and inland temporal Spanish springs (< 92.8 mg/L; Kalff, 2003; Horne and Goldman, 1994; Ortega et al., 1992). In contrast, Na^+ concentration (37-234 mg/L; Fig. 2D) was similar to that found in Cuatro Ciénegas springs: Anteojo, Churince, Escobedo, Juan Santos, Matorral Oeste, Poza Azul, Santa Tecla and Tío Cándido (41-149.5 mg/L; Hendrickson et al., 2008). Presence of potassium and sulfate in Samalayuca aquatic systems may be related to calcite and gypsum

dissolution processes (CONANP, 2013; Kolahchi and Jalali, 2006). Potassium levels in all monitored springs (Fig. 3F; 1.34-203.4 mg/L) were above those reported by Ortega et al. (1992) for inland temporary ponds (<29.5 mg/L) but similar to those found in systems connected to groundwater (25.3-256 mg/L). Sulfate values in these springs (1214.78-2553.98 mg/L) were higher than those observed in other springs in the CD (10.8-1328 mg/L; Hendrickson et al., 2008; Blinn, 2008), and inland temporary ponds in Spain (10-88 mg/L; Ortega et al., 1992). Phosphate levels in Samalayuca springs were similar to those determined by Ortega et al. (1992) in small aquatic systems in Spain (0.03-1.3 mg/L).

Other cations, such Mn (0.02-0.9 mg/L), Cu (0.02-0.1 mg/L) and Fe (0.2-3.2 mg/L) were detected at higher levels than those reported by Horne and Goldman (1994) for natural aquatic systems: <35 µg/L (Mn), 0.01 mg/L (Cu), and 0.1 mg/L (Fe). Particularly in Ojos Altos A, soil color indicates presence of manganese oxides, because under the anoxic conditions prevailing in this spring (DO=1.3-4.6 mg/L) and the pH >6, manganese is likely dissolved in water, thus occurring as Mn²⁺ (Hem, 1989).

Through this research we detected significant differences in volume, temperature, dissolved oxygen, conductivity, potassium, sodium, hardness, and sulfate either within the monitored springs and/or among seasons. These changes clearly indicate that water chemical characteristics varied not only between regions as reported by Walsh et al. (2008), but also within a local spring system (Ojos Altos A-C and Ojo de En Medio-Ojo de La Punta). DO, for example, was below the criterion established by EPA for aquatic life protection (4 mg/L; Quality Criteria for Water; EPA, 1982) in Ojo de En Medio, Ojos Altos A, and Ojos Altos C; as are other CD springs in Cuatro Ciénegas (Anteojito and Escobedo; Hendrickson et al., 2008) and San Luis Potosí (Media Luna, Los Peroles & San Sebastián; Walsh et al. unpublished). These low levels of DO may be related to salinity, because an excessive amount of ions related to salinity decreases the amount of dissolved oxygen in water (Horne and Goldman, 1994). In contrast, Ojos Altos B (9.3 mg/L), Ojo de la Punta (10.4 mg/L), and Ojo de Santa María (8-20 mg/L) were saturated or supersaturated with oxygen (Fig. 3B, Table 3). DO supersaturation may be produced by aquatic vegetation photosynthesis during sampling (around midday) in Ojos Altos B (filamentous microalgae), Ojo de La Punta (*Chara*), and Ojo de Santa María (*Chara* and filamentous microalgae; Fig. 2). Recent data (T, pH, DO, and EC) collected during a small survey (July-November, 2017) of Ojo de La Casa and Ojo de La Punta were within the ranges detected during this research. Unfortunately, Ojo de En Medio is no longer flowing: our preserved samples of zooplankton are now of historical relevance.

In our study we found that hypoxic springs (1.32-4.57 mg/L) with elevated hardness levels (1589-2302 mg/L) provided poor habitat for rotifer population development and likely accounted for the lack of species diversity observed in Ojos Altos A (conductivity of 2.46-3.04 mS/cm³, greater than that of mineralized waters (0.5-1.0 mS/cm³).

Based on RDA results seven species from the genus *Lecane* were positive correlated with DO concentration (Figure 4). As anticipated, more species were found in waters containing higher levels of dissolved oxygen (this research) and lower salt concentrations (as in Walsh

et al., 2008). Such is the case of Ojo de La Punta, the spring with the highest S ($n=28$). This spring is characterized by a high concentration of DO (4.7–10.4 mg/L), neutral to moderately basic pH (6.4–8.2), large temperature fluctuations (1.3–24.3°C), low levels of hardness (402–794 mg/L) and Ca^{2+} (90.9–267.3 mg/L), intermediate concentrations of K^+ (60.8–103.9 mg/L), and a relatively stable conductivity (1.06–1.3 mS/cm). Seasonal variation in temperature and physicochemical conditions of this spring likely provided suitable habitat for seasonal succession of rotifer species. Similar ranges of DO (8.0–20.2 mg/L), pH (8.02–9.0), and conductivity (0.9–1.0 mS/cm³) to those observed in Ojo de La Punta were present in Ojo de Santa María, the spring with the next highest richness. Although K^+ (3.5–6.2 mg/L), Na^+ (216.8–281.7 mg/L), hardness (22.2–32.9 mg/L), and Ca^{2+} (16.3–19.3 mg/L) levels differed from those detected in Ojo de La Punta, these differences provide a unique system where numerous rotifer species flourished ($S=25$; 7 of which were unique to this spring). Interestingly, 15 rotifer species were identified in Ojo de En Medio, a hypoxic aquatic system (DO=2.5–4.7 mg/L) where the presence of reeds may prevent water aeration (Fig. 2). Probably low levels of dissolved K (15.1–68.1 mg/L), Na (36.8–138.3 mg/L), and Ca salts (158.5–440.4 mg/L; ions related to salinity), as compared to the other monitored, resulted in the successful colonization of six rotifer species found in this springs but not in the others (Table 6), since a decrease in S has been related to salinities > 1.5 mS/cm in some inland brackish water systems (Walsh et al., 2008; Kaya et al., 2010).

We identified rotifer species from 25 genera (in addition to unidentified bdelloids) in springs that range in volumes from 0.3 to 194 L during the year, very small systems compared to the 5.1×10^7 L Montezuma Well, where Blinn (2008) found only two genera of monogonont rotifers (*Lecane* and *Testudinella*) and unidentified bdelloids. Walsh et al. (2008) reported 59 species in 48 saline sites in the CD; of these 33 were found in the seven Samalayuca springs (Table 6). Ten of the identified species have not been recorded previously in these or other Chihuahuan Desert Springs (Walsh et al., 2014a, 2014b & 2008; Wallace et al., 2008; Walsh unpublished data): *Pleurocha petromyzon* Ehrenberg, 1832, *Filinia brachiata* (Rousselet, 1901), *Keratella tropica* (Apstein, 1907), *Synchaeta pectinata* Ehrenberg, 1832, *S. tremula* (Müller, 1786), *Cephalodella globata* (Gosse, 1887), *Notommata glyphura* Wuelfer, 1935, *Testudinella patina* (Hermann, 1783), *Filinia saltator* (Gosse, 1886) and *Proalides tentaculatus* Beauchamp, 1907 (Table 7). More recently, during a short survey of the same springs (July–August 2017), we identified eight species not previously recorded in Ojo de La Casa: *Colurella obtusa* (Gosse, 1886), *Dipleuchlanis elegans* (Wierzejski, 1893), *Filinia pejeri* Hutchinson, 1964, *Hexarthra fennica* (Levander, 1892), *Keratella cochlearis* (Gosse, 1851), *Lecane lateralis* Sharma, 1978, *L. papuana* (Murray, 1913), *Platonuspatulus* (Müller, 1786) and *Ptygura beauchampi* Edmondson, 1940 (not recorded previously in the CD). Thus it is evident that Samalayuca springs have a disproportionally high biodiversity in rotifers given their small size and more sampling will likely reveal additional species.

5. Conclusions

Arid land springs are at risk from anthropogenic activities and climate change. In the northern CD groundwater pumping for agricultural and domestic uses is drawing down aquifers at the same time that drought conditions are limiting natural recharge. We sampled seven at-risk springs stressed by these factors to determine water quantity and quality and

rotifer diversity. Overall water quality varied among springs and seasons. One spring was a mesothermal aquatic system; five springs were oxygen limited at least part of the year, four contained brackish water, and temperature varied significantly between seasons in all springs. *S* was positively correlated to dissolved oxygen concentration, low conductivity, low nitrate, and the fall season, and was quite high considering the small, isolated nature of these springs. It is not surprising that communities found within these regional springs were highly nested, with the largest spring containing the majority of species. Through this research we provided basic information to understand how changes in water physicochemical characteristics may affect species richness in aquatic systems and demonstrate that small but permanent springs exposed to variable temperature regimes and specific chemistries are important in maintaining regional species pools. Thus these results provide much need regional research. By characterizing spring type and providing an update faunal inventory, we provide important information for water management and species conservation.

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Highlights

- Physicochemical features and rotifer diversity were determined for 7 springs.
- Temperature, conductivity and oxygen level varied seasonally among springs.
- Sixty species of rotifers showed nestedness among the sites.
- Species richness was correlated to dissolved oxygen, nitrate, and fall season.
- Ten rotifer species were added to those reported in Chihuahuan Desert springs.

Fig. 1.
Location of springs sampled in the Samalayuca region in northern Chihuahua, Mexico.

Fig 2.
Springs of the Samalayuca region, Chihuahua, Mexico: upper panel images correspond to Ojos Altos A, B and C, respectively; lower panel images are D) Ojo de Santa María, E) Ojo de En Medio, and F) Ojo de La Punta.

Fig. 3.

Physicochemical characteristics and species patterns in seven springs located in near Samalayuca, México: A) temperature (T), B) dissolved oxygen (DO), C) hardness (as CaCO₃ mg/L, D) sodium concentration (Na), E) electric conductivity (EC), F) potassium concentration (K), G) species richness (S), and H) calcium concentration. Abbreviations: W = winter, Sp = spring, Sum = summer, A = autumn. OEM = Ojo de En Medio; OLP = Ojo de La Punta; OAA = Ojos Altos A; OAB = Ojos Altos B; OAC = Ojos Altos. Bars indicate one standard error of the mean.

Fig. 4. Seasonal sulfate concentration in Ojos Altos springs, Samalayuca, Mexico. Bars indicate one standard error of the mean.

Fig. 5.

Redundancy Analysis (RDA) of seven springs in Samalayuca, Chihuahua, Mexico: Analysis included species and environment factors as variables. Abbreviations are as follows: Bdel = unidentified bdelloid rotifers; BranAngu = *Brachionus angularis*; CephGrcl = *Cephalodella gracilis*; CephSter = *C. sterea*; LecaAega = *Lecane aeganea*; LecaBull = *L. bulla*; LecaClos = *L. closterocerca*; LecaHorn = *L. hornemanni*; LecaIner = *L. inermis*; LecaLunr = *L. lunaris*; LecaPyri = *L. pyriiformis*; LecaThal = *L. thalera*; LecaUndu = *L. undulata*; LepaPate = *Lepadella patella*; and SyncTrem = *Synchaeta tremula*. DO=dissolved oxygen, NO₃= nitrate, CE=electrical conductivity, T=temperature, PTotal=total phosphorus, V=volume.

Fig. 6.

Rotifer species nestedness in seven northern Chihuahuan Desert springs. Supported by 4 null models ($P < 0.001$). Idiosyncratic species are identified with T° 1SD ($n=8$) or 2SD ($n=3$) of the mean matrix $T^\circ = 12.1$.

Table 1.

Water quality parameters monitored in Samalayuca springs, Chihuahua, Mexico. Temp = temperature; EC = electrical conductivity; DO = dissolved oxygen; TDS = total dissolved solids.

Site	Physical and Chemical Properties	Nutrients	Ions that determine salinity
Ojos Altos A-C		N, P, Cu, Mn and Fe	
Ojo de En Medio & de La Punta	Temp, pH, EC, DO, TDS	P, NO ₃ ⁻ and Mn	Ca ⁺² , Mg ⁺² , Na ⁺¹ , K ⁺¹ , CO ₃ ⁻² , HCO ₃ ⁻¹ , SO ₄ ⁻² and Cl ⁻¹
Ojo de Santa María		Cu ²⁺ , Zn ²⁺ , Mn, Fe and NO ₃ ⁻	

Table 2.

Water volume (m³) of surveyed springs averaged by season: Ojos Altos A-C (2007), Ojo de En Medio and Ojo de La Punta (2008). *Indicates significance at $p < 0.05$, ANOVA followed by Tukey test.

Site (Ojos)	Winter	Spring	Summer	Fall
Altos A	1.5	1.7	1.9	3.0
Altos B	7.0	7.8*	5.0*	6.4
Altos C	6.3	7.03	5.05	6.4
De En Medio	27.8*	21.1	17.3	0.3*
De La Punta	194*	165	149	131*

Table 3.

Ranges of physical and chemical parameters measured in surveyed springs in Samalayuca, Chihuahua, Mexico. EC=Electrical conductivity; DO= Dissolved oxygen.

Site (Ojos)	Temp (°C)	pH	EC (mS/cm ³)	DO (mg/L)
Altos A	11-23	6.0-7.8	2.7-3.2	0.9-4.9
Altos B	17-34	6.4-7.6	2.4-2.5	1.4-15.5
Altos C	30-36	6.6-7.7	2.4-2.5	1.3-4.45
De En Medio	7-24	5.6-7.7	0.9-3.3	2.0-7.7
De La Punta	7-24	6.3-9.0	1.0-1.4	0.9-16.3
De La Casa	4-25	5.4-8.3	1.5-2.9	5.9-8.3
Santa María	17-32	7.0-9.0	0.9-1.0	8.0-20.2

Table 4.

Concentration of nutrients (ranges) in monitored springs in Samalayuca, Chihuahua, Mexico. — Not determined; a= Inorganic nitrogen; b= Organic nitrogen; c=Total phosphorus as phosphates.

Sites	NO ₃ -N ^a (mg/L)	N-Org ^b (mg/L)	PO ₄ -P ^c (mg/L)	Mn (mg/L)	Fe (mg/L)	Cu (mg/L)
Altos A	0.3-0.7	0.25-0.5	0.09-0.3	0.05-0.25	0.6-3.2	0.03-0.04
Altos B	0.2-0.8	0.19-1.3	0.3-0.35	0.02-0.06	0.47-2.1	0.03-0.05
Altos C	0.3-0.5	0.2-0.5	0.2-0.3	0.02-0.04	0.5-1.1	0.02-0.04
De En Medio	0.3-1.3	0.2-1.1	0.90-1.6	0.03-0.9	0.02-1.3	0.02-0.04
De La Punta	0.04-0.7	0.07-0.6	0.1-1.1	0.01-0.9	0.3-0.6	0.02-0.03
De Santa María	2.5-5	—	—	0.01-0.06	0.4-2.3	0.002-0.1
De La Casa	0.06-4.6	—	—	—	—	—

Table 5.

Rotifer species richness in Samalayuca springs in 2007-2011; (2017).

Site	Richness (S)	Unique Species
Ojo de Santa María	25	7
Ojo de La Casa	16 (25)	0 (5)
Ojo de En Medio	16	6
Ojo de La Punta	29 (10)	12 (0)
Ojos Altos A	2	0
Ojos Altos B	12	2
Ojos Altos C	9	1
Total	56 (8)	28 (5)

Table 6.

List of rotifer species found in Samalayuca springs, Chihuahua, Mexico. Species in bold have not been previously recorded in these springs. X indicates species presence in springs and F indicates rotiferan family: 1 Brachionidae, 2 Collothecidae, 3 Dicranophoridae, 4 Proalidae, 5 Euchlanidae, 6 Filiniidae, 7 Flosculariidae, 8 Ituridae, 9 Lecanidae, 10 Lepadellidae, 11 Notommatidae, 12 Philodinidae, 13 Proalidae, 14 Synchaetidae, and 15 Testudinellidae.

F	Species	Sites (Ojos)				
		La Casa	En Medio	La Punta	Altos	Santa María
					A B C	
1	Brachionus angularis Gosse, 1851			X		
	<i>Brachionus calyciflorus</i> Pallas, 1766			X		
	<i>Brachionus quadridentatus</i> Hermann, 1783	X		X		X
	<i>Keratella tropica</i> (Apstein, 1907)			X		
	Notholca acuminata (Ehrenberg, 1832)		X			
	Platylabus quadricornis (Ehrenberg, 1832)			X		
2	<i>Collotheca coronetta</i> (Cubitt, 1869)		X			
3	<i>Dicranophorus epicharis</i> Harring & Myers, 1928			X		
4	<i>Proalides tentaculatus</i> Beauchamp, 1907					X
5	Euchlanis dilatata Ehrenberg 1830	X				X
6	<i>Filinia brachiata</i> (Rousselet, 1901)			X		
	<i>Filinia longiseta</i> (Ehrenberg, 1834)			X		
	<i>Filinia saltator</i> (Gosse, 1886)					X
7	<i>Limnias ceratophylli</i> Schrank, 1803					X
8	<i>Itura</i> sp.					X
9	Lecane aeganea Harring, 1914					X
	Lecane arcula Harring, 1914		X	X		
	Lecane bulla (Gosse, 1851)	X	X	X	X X X	
	Lecane closterocerca (Schmarda, 1859)			X	X X	
	Lecane cornuta (Müller, 1876)	X		X		
	<i>Lecane furcata</i> Murray, 1913			X		
	<i>Lecane hamata</i> (Stokes, 1896)	X		X		X
	Lecane hornemanni (Ehrenberg, 1834)				X	
	Lecane inermis (Bryce, 1892)				X X	
	Lecane luna (Müller, 1776)	X		X		X
	Lecane lunaris (Ehrenberg, 1832)				X	
	Lecane nana (Murray, 1913)					X
	Lecane pyriformis (Daday, 1915)				X	X
	Lecane quadridentata (Ehrenberg, 1830)					X
	Lecane thalera (Harring & Mayers, 1926)			X	X	
	Lecane undulata Hauer, 1938				X X	
10	<i>Colurella obtusa</i> (Gosse, 1886)				X	

F	Species	Sites (Ojos)					
		La Casa	En Medio	La Punta	Altos		Santa María
					A	B	C
	<i>Lepadella acuminata</i> (Ehrenberg, 1834)			X			
	<i>Lepadella patella</i> (Müller, 1773)		X	X	X	X	X
	<i>Lepadella triptera</i> (Ehrenberg, 1830)	X	X	X			X
11	<i>Cephalodella catellina</i> (Müller, 1786)	X					X
	<i>Cephalodella forficula</i> (Ehrenberg, 1838)		X				X
	<i>Cephalodella gibba</i> (Ehrenberg, 1830)	X					X
	<i>Cephalodella globata</i> (Gosse, 1887)		X	X			
	<i>Cephalodella cf. graciosa</i>		X				
	<i>Cephalodella gracilis</i> (Ehrenberg, 1830)			X		X	X
	<i>Cephalodella cf. marina</i>			X			
	<i>Cephalodella</i> sp2			X			
	<i>Cephalodella megaloccephala</i> (Glascott, 1893)		X				
	<i>Cephalodella cf. physalis</i>		X				
	<i>Cephalodella sterea</i> (Gosse, 1887)			X		X	X
	<i>Eosphora ehrenbergi</i> Weber, 1918						X
	<i>Notommata glyphura</i> Wulfert, 1935	X					X
	<i>Notommata haueri</i> Wulfert, 1939		X				
	<i>Pleurotrocha petromyzon</i> Ehrenberg, 1830		X				
	<i>Pleurotrocha sigmoidea</i> Skorikov, 1896	X	X				
	<i>Reticula melandoca</i> (Gosse, 1887)		X				X
12	<i>Philodina megalotrocha</i> Ehrenberg, 1832	X	X	X			X
13	<i>Proales similis</i> Beauchamp, 1907		X				X
14	<i>Polyarthra dolichoptera</i> Idelson, 1925	X	X				
	<i>Synchaeta pectinata</i> Ehrenberg, 1832			X			
	<i>Synchaeta tremula</i> (Müller, 1786)			X			
15	<i>Testudinella patina</i> (Hermann, 1783)	X					X
	- Unidentified bdelloid	X	X	X	X	X	X