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Spatial Distribution, Temporal Variation and Specificity of Microhabitat of *Tropisternus* Species (Coleoptera: Hydrophilidae) in Permanent Ponds

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Abstract

The spatial distribution and temporal variation of 11 species of *Tropisternus* were analyzed in two permanent ponds located in the province of Corrientes, Argentina. Samples were collected every 15 days, between October 2010 and March 2011. The species recorded were *Tropisternus collaris* (Fabricius), *Tropisternus ovalis* Castelnau, *Tropisternus laevis* (Sturm), *Tropisternus lateralis limbatus* (Brullé), *Tropisternus longispina* Fernández & Bachmann, *Tropisternus carinispina* Orchymont, *Tropisternus bourmeisteri* Fernández & Bachmann, *Tropisternus apicipalpis* (Chevrolat), *Tropisternus dilatatus* Bruch, *Tropisternus obesus* Bruch, and *Tropisternus ignoratus* Knisch. The first four were present in higher proportions than the remaining during most of the study period. The spatial distribution of individuals was mostly related to the homogeneity or heterogeneity of the ecosystem in relation to microhabitats with aquatic vegetation: In ponds with different microhabitats, individuals were mainly aggregated, whereas in ponds with homogenous features, individuals were randomly distributed. However, when species were analyzed individually, the spatial distribution and the use of microhabitat by each species were different with respect to preference and behavior.

Introduction

The genus *Tropisternus* (Coleoptera: Hydrophilidae) has 29 species in Argentina. They are known as silver water beetles because their sternal pubescence retains a thin layer of air that confers a characteristic ventral silver color when submerged in water (Oliva *et al* 2002). This group is very well represented with respect to the diversity of aquatic coleopterans in the area (Gómez Lutz *et al* 2012). Information on their distribution allows for learning on their life histories, behavior, preferences, and plasticity. It also helps to describe the distribution patterns of animals and the use of space, which is ecologically significant and necessary to design different sampling methods for specific aims (Poole 1974, Southwood 1978). The spatial distribution of individuals is often affected by several factors such as environmental

homogeneity and variation over time (Pielou 1977, Fernández & Kehr 1995).

The main goals of the present study were (1) to calculate the relative abundance and temporal variation of *Tropisternus* species in two ponds of Corrientes Province, Argentina, (2) to determine the spatial distribution and the possible factors affecting it, and (3) to study the microhabitats occupied by the different *Tropisternus* species. The present study is a contribution to the bioecological studies of aquatic beetles in northeastern Argentina, an area with many water bodies, where several studies related to the fauna associated with the aquatic vegetation have been carried out (Poi de Neiff 1977, 1983, 1992, Poi de Neiff & Neiff 1977, 2006, Poi de Neiff & Carignan 1997, Torres *et al* 2012, Libonatti *et al* 2013).

Material and Methods

Ponds studied

The study was conducted in two permanent ponds located 12 km from Corrientes City, in the province of Corrientes, Argentina ("Tendalero" pond and "Don Luis" pond) (Fig 1). The "Tendalero" pond ($27^{\circ}28'S$, $58^{\circ}43'W$) has an elliptical shape, with a length of approximately 150 m, a width of 80–100 m and a depth of 0.8–1.5 m. The pond has three clearly differentiated microhabitats on its surface, two of them formed by aquatic vegetation—*Hydrocleys nymphoides* and *Limnobium laevigatum*—and the other without aquatic vegetation ("open area"). The "Don Luis" pond ($27^{\circ}29'S$, $58^{\circ}43'W$) has a circular shape and a diameter of approximately 80 m; its depth in the central part is about 1.2 m. The aquatic vegetation is dominated

by *Ludwigia* sp. and different grass species. This pond does not present evident microhabitats on its surface. The aquatic vegetation is homogeneously dispersed.

Experimental design and analytical procedure

Samples were collected every 15 days between October 2010 and March 2011, corresponding to spring, summer, and early autumn, a time of maximum insect activity. The large size of the two ponds led us to select the sample area arbitrarily, considering it as a representative portion of the main features of the two ponds. The number of sampling units was proportional to the surface of the two ponds (seven and four sampling units, respectively). For independence reasons between the samples, the distance between the sampling units was of 3 m. Samples were collected usually between 10 a.m.

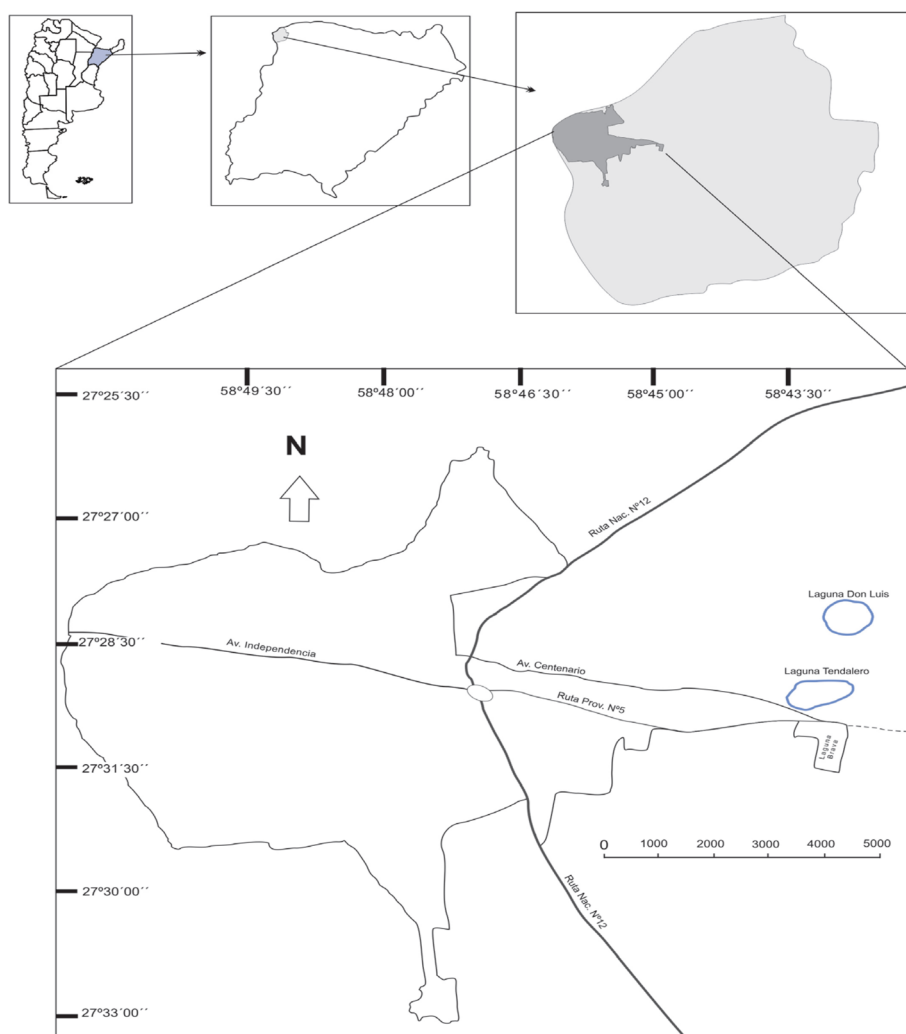


Fig 1 Location of the two sampling sites in Corrientes Province: Tendalero pond and Don Luis pond.

and 1 p.m. and fixed in situ in 5% formaldehyde, transferred to the laboratory for identification, and then stored in 70% ethanol. Aquatic insects were collected with an aquatic hand net (mesh size 300 μm , diameter 30 cm) by dragging 2 m.

Species were identified following Trémouilles *et al* (1995), Fernández & Bachmann (1998), Oliva *et al* (2002), and Archangelsky *et al* (2009). The material studied was deposited in the collection of the Centro de Ecología Aplicada del Litoral (CECOAL-CONICET), Corrientes.

Spatial distribution

We considered only samples with at least five individuals to determine their spatial distribution. The spatial distribution was determined in two different ways: firstly, by using the dispersion index (DI) and spatial pattern according to chi-squared distribution (Elliot 1971), and secondly, by applying distribution models as Poisson series, positive binomial, and

negative binomial (Pielou 1977), and checking the difference between expected (obtained through the models of best fit) and observed data with a chi-squared test (Southwood 1978). When the data did not conform to the theoretical models applied, we considered the aggregation type obtained by the DI for that sample.

In sampling units with aggregate spatial distribution and adjusted to the binomial negative model, we calculated the mean number of individuals in the aggregation by using the formula proposed by Arbous & Kerrich as described in Southwood (1978):

$$\lambda = \frac{\mu}{2K} \nu$$

where λ is the number of individuals in the aggregation for the probability level allocated to ν , μ is the mean, K is the aggregation index of the probability function of the negative

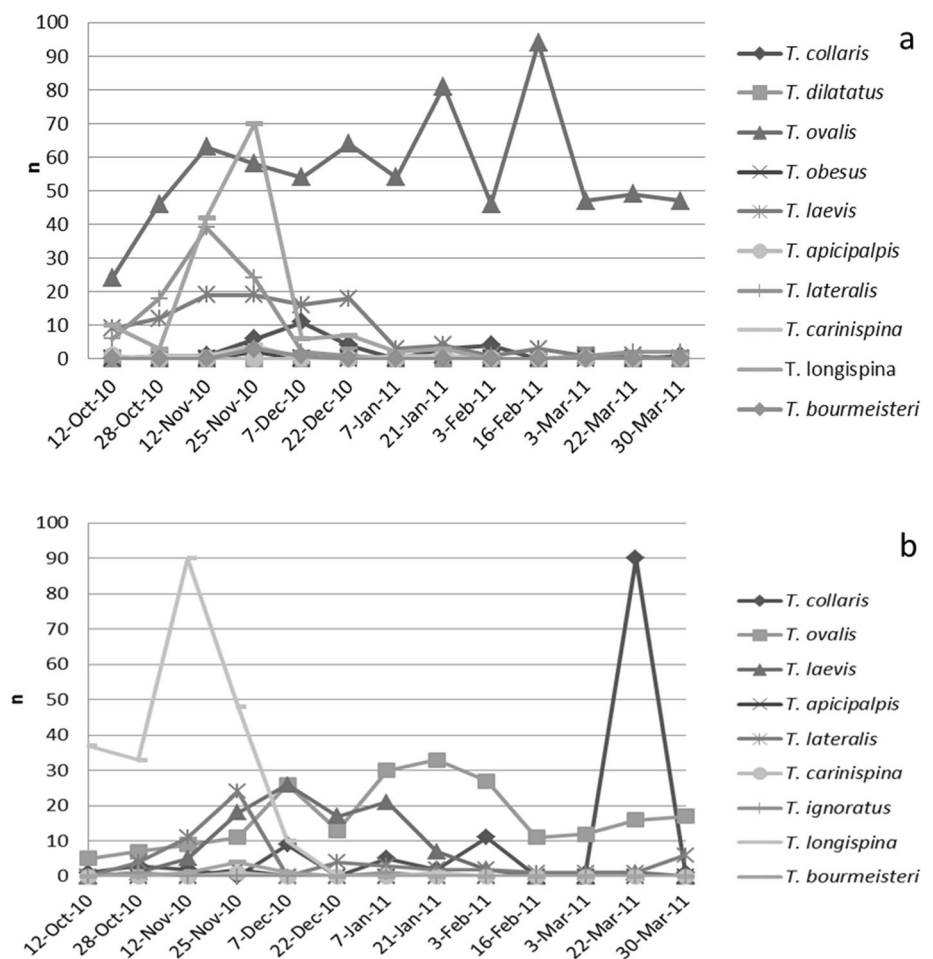


Fig 2 Abundance of *Tropisternus* species present in the **a** Tendalero pond and in the **b** Don Luis pond.

binomial, and ν is the function with a χ^2 distribution with 2 K degrees of freedom. When the mean number of individuals in the aggregation is lower than the critical value of 2, the aggregation seems to be caused by some environmental effect and not by an active process. Aggregations higher than the critical value of 2 can be either caused by environmental factors or be a result of an active process.

Microhabitat specificity and preference

We used nonparametric Kruskal-Wallis test and then the Dunn's test with the Bonferroni's correction to evaluate whether the microhabitats were differentially used by the *Tropisternus* species.

The use of the three microhabitats of Tendalero pond by each species was tested through a K proportion test, and when it was significant, a two proportion Z test was applied between pairs of microhabitats.

Results

Tendalero pond

Relative abundance and temporal variation. Ten species of coleopterans were recorded: *Tropisternus collaris* (Fabricius), *Tropisternus ovalis* Castelnau, *Tropisternus laevis* (Sturm), *Tropisternus lateralis limbatus* (Brullé), *Tropisternus*

Table 1 Species of *Tropisternus* sampled in the Tendalero pond and sampling dates.

Species	Dates	Total	μ	S^2	DI	Prob.	Dist.	K	df	Prob. χ^2	λ^a
<i>Tropisternus collaris</i>	25 Nov. 10	6	0.85	1.48	10.33	0.11	R	—	2	>0.05	—
	7 Dec. 10	11	1.57	13.95	53.27	0.00	A	0.20	4	>0.05	0.71
<i>Tropisternus ovalis</i>	12 Oct. 10	24	3.43	5.62	9.83	0.13	R	—	5	>0.05	—
	28 Oct. 10	46	6.57	3.29	3.00	0.81	R	—	8	>0.05	—
	12 Nov. 10	63	9.00	19.00	12.67	0.05	R	—	14	<0.05	—
	25 Nov. 10	58	8.29	35.90	26.00	0.00	A	2.49	4	<0.05	5.59
	7 Dec. 10	54	7.71	40.24	31.30	0.00	A	1.83	4	>0.05	4.99
	22 Dec. 10	64	9.14	67.81	44.50	0.00	A	1.42	4	>0.05	4.45
	7 Jan. 11	54	7.71	40.24	31.30	0.00	A	1.83	4	>0.05	4.99
	21 Jan. 11	81	11.57	94.95	49.23	0.00	A	1.61	4	>0.05	8.52
	3 Feb. 11	46	6.57	19.95	18.22	0.01	A	3.23	4	>0.05	5.45
	16 Feb. 11	94	13.43	105.62	47.19	0.00	A	1.96	4	<0.05	8.12
	3 Mar. 11	47	6.71	64.24	57.40	0.00	A	0.78	4	>0.05	1.95
	22 Mar. 11	49	7.00	26.33	22.57	0.00	A	2.53	4	>0.05	6.01
<i>Tropisternus laevis</i>	30 Mar. 11	47	6.71	64.24	57.40	0.00	A	0.78	4	>0.05	1.95
	12 Oct. 10	9	1.29	2.57	12.00	0.06	R	—	2	<0.05	—
	28 Oct. 10	12	1.71	7.57	26.50	0.00	A	0.50	4	>0.05	0.78
	12 Nov. 10	19	2.71	25.90	57.26	0.00	A	0.32	4	>0.05	1.23
	25 Nov. 10	19	2.71	31.57	69.79	0.00	A	0.26	4	>0.05	1.23
	7 Dec. 10	16	2.29	14.90	39.13	0.00	A	0.41	4	>0.05	1.04
<i>Tropisternus lateralis</i>	22 Dec. 10	18	2.57	15.95	37.22	0.00	A	0.49	4	>0.05	1.17
	12 Oct. 10	6	0.86	0.81	5.67	0.46	R	—	1	>0.05	—
	28 Oct. 10	18	2.57	4.62	10.78	0.10	R	—	5	>0.05	—
	12 Nov. 10	39	5.57	16.95	18.26	0.01	A	2.73	4	>0.05	4.44
	25 Nov. 10	24	3.43	18.95	33.17	0.00	A	0.76	4	>0.05	1.03
<i>Tropisternus longispina</i>	12 Oct. 10	10	1.43	2.62	11.00	0.09	R	—	3	>0.05	—
	12 Nov. 10	42	6.00	69.33	69.33	0.00	A	0.57	4	>0.05	2.40
	25 Nov. 10	70	10.00	237.33	142.40	0.00	A	0.44	4	>0.05	4.55
	7 Dec. 10	6	0.86	2.14	15.00	0.02	A	0.57	4	>0.05	0.34
	22 Dec. 10	7	1.00	1.67	10.00	0.12	R	—	2	>0.05	—

μ arithmetic mean, S^2 variance, DI Dispersion Index, $Prob.$ probability according to χ^2 distribution, $Dist.$ distribution type, A aggregate distribution (the best fit model is the negative binomial), R random distribution (the best fit model is the Poisson series), K Aggregation Index, df degrees of freedom, $Prob. \chi^2$ probability of χ^2 , λ mean aggregation, calculated with the formula proposed by Arbous and Kerrich (1951)].

^a Critical value 2. Higher than 2, aggregations by active processes; lower than 2, aggregation by environmental effects.

longispina Fernández & Bachmann, *Tropisternus carinispina* Orchymont, *Tropisternus bourmeisteri* Fernández & Bachmann, *Tropisternus dilatatus* Bruch, *Tropisternus obesus* Bruch, and *Tropisternus apicipalpis* (Chevrolat). *Tropisternus ovalis* was recorded in all samples with high relative abundance in most samples. *Tropisternus longispina*, *T. lateralis*, and *T. laevis* showed high abundance in October, November, and December. The other species showed low abundance during the entire study period (Fig 2a).

Spatial distribution. The abundance of only five out of the ten species recorded in this pond was appropriate (see “Materials and Methods”) for spatial distribution analysis (Table 1). For *T. collaris* and *T. lateralis*, 50% of the samples were randomly distributed and the other 50% were aggregated. The aggregation of individuals in *T. collaris* was due to external factors, whereas that in *T. lateralis* was due to external and active processes. The aggregate distribution was predominant in *T. ovalis* (77% of samples), *T. laevis* (83%), and *T. longispina* (60%). In *T. ovalis*, the K values of negative binomial were higher (>0.78), showing aggregations that were not too strong, being most of them due to active processes. Although the K values were <0.58 for *T. laevis* and *T. longispina*, the aggregation was very strong, but the aggregation in the former was due to external factors, while in the latter, it was mostly due to active processes (Table 1).

Specificity and preference of microhabitats. Three microhabitats were differentially used by *Tropisternus* species (Kruskal-Wallis test $K-W=24.37$; $df=2$; $p<0.0001$). An a posteriori test (Dunn’s test) with Bonferroni’s correction ($\alpha=0.016$) indicated significant differences between the microhabitats with *L. laevigatum* and *Hydrocleys nymphoides* with respect to the open area. A total of 222 and 165 individuals of *Tropisternus* were respectively recorded in the microhabitats with each plant species, whereas only 49 individuals were collected in the open area.

Only *T. collaris* similarly used the three microhabitats (K proportion test, $\chi^2=0.37$; $df=2$; $p>0.05$). *Tropisternus ovalis* and *T. laevis* differently occupied the three microhabitats (*T. ovalis*: $\chi^2=82.87$; $df=2$; $p<0.001$; *T. laevis*: $\chi^2=30.68$; $df=2$; $p<0.001$). The microhabitat preference order for these species was *L. laevigatum*, *Hydrocleys nymphoides*, and open area. *Tropisternus lateralis* and *T. longispina* preferentially used the two vegetated microhabitats in comparison to the open area (*T. lateralis*, $\chi^2=24.61$; $df=2$; $p<0.001$; *T. longispina*, $\chi^2=37.03$; $df=2$; $p<0.001$) (Table 2).

Don Luis pond

Relative abundance and temporal variation. Nine species were recorded: *T. collaris*, *T. ovalis*, *T. laevis*, *T. lateralis* *limbatus*, *T. apicipalpis*, *T. carinispina*, *T. ignoratus* Knisch,

T. longispina, and *T. bourmeisteri*. *Tropisternus longispina* and *T. collaris* showed different peaks of abundance during the study period. While *T. longispina* was evident during the first 3 months (October to December), *T. collaris* was evident during the last month (March). *Tropisternus ovalis* and *T. laevis* were also well represented but in lower proportion. These species showed no evident abundance peaks but were the most abundant in January and February. During the study period, *T. ovalis* was the only species recorded on every sampling occasion (Fig 2b).

Spatial distribution. The abundance of only five out of the nine species recorded in this pond was appropriate for spatial distribution analysis (see “Materials and Methods”) (Table 3). The random spatial distribution was predominant in *T. ovalis* (62% of the samples), *T. lateralis*, and *T. laevis* (67% of the samples). *Tropisternus collaris* showed both random (50% of samples) and aggregate (50%) spatial distribution. *Tropisternus longispina* showed mainly aggregate spatial distribution (80% of samples). In this last species, the K value ranged between 0.95 and 1.46, demonstrating a weak aggregation pattern due to active processes ($\lambda>2$). In the other four species with aggregate distribution, the K values ranged from 0.33 to 3.78, indicating lax aggregations.

Specificity and preference of microhabitats. In Don Luis pond, we did not perform these analyses due to the homogeneity of the site. Unlike Tendalero pond, this pond had no different microhabitats.

Discussion

Although the two ponds studied were structurally different, eight of the 11 species were common to both. While

Table 2 Relation between species of *Tropisternus* and microhabitats in the Tendalero pond obtained from K proportions test and Z test for two proportions.

Species	Microhabitats		
	(A)	(B)	(C)
<i>Tropisternus collaris</i>	a	a	a
<i>Tropisternus ovalis</i>	150 ^b	110 ^b	45 ^b
<i>Tropisternus laevis</i>	23 ^b	12 ^b	1 ^b
<i>Tropisternus lateralis</i>	20 ^b	14 ^b	1 ^b
<i>Tropisternus longispina</i>	26 ^b	28 ^b	1 ^b

The different microhabitats: (A) *Limnabium laevigatum*, (B) *Hydrocleys nymphoides*, (C) water without floating vegetation.

^a No significant differences according to the test of K proportions in the use of microhabitats.

^b Significant differences between groups or of microhabitats analyzed from Z test for two proportions.

Table 3 Species in the Don Luis pond with dates.

<i>Tropisternus</i>	Dates	Total	μ	S^2	DI	Prob.	Dist.	K	df	Prob. χ^2	λ^a
<i>Tropisternus collaris</i>	7 Dec. 10	9	2.25	2.92	3.89	0.27	R	—	3	>0.05	—
	7 Jan. 11	5	1.25	1.58	3.80	0.28	R	—	2	>0.05	—
	3 Feb. 11	11	2.75	23.58	25.73	0.00	A	0.36	1	>0.05	1.25
	22 Mar. 11	90	22.5	459.67	61.29	0.00	A	1.16	1	<0.05	13.47
<i>Tropisternus ovalis</i>	12 Oct. 10	5	1.25	3.58	8.60	0.04	R	—	3	<0.05	—
	28 Oct. 10	7	1.75	2.25	3.86	0.28	R	—	2	>0.05	—
	12 Nov. 10	9	2.25	2.92	3.89	0.27	R	—	3	>0.05	—
	25 Nov. 10	11	2.75	3.58	3.91	0.27	R	—	3	>0.05	—
	7 Dec. 10	26	6.5	23.00	10.62	0.01	A	2.56	1	>0.05	5.52
	22 Dec. 10	13	3.25	6.25	5.77	0.12	R	—	5	>0.05	—
	7 Jan. 11	30	7.50	33.67	13.47	0.00	A	2.15	1	>0.05	5.86
	21 Jan. 11	33	8.25	26.25	9.55	0.02	A	3.78	1	>0.05	6.92
	3 Feb. 11	27	6.75	20.92	9.30	0.03	R	—	9	<0.05	—
	16 Feb. 11	11	2.75	2.92	3.18	0.36	R	—	4	>0.05	—
	3 Mar. 11	12	3.00	7.33	7.33	0.06	R	—	6	<0.05	—
	22 Mar. 11	16	4.00	12.67	9.50	0.02	A	1.85	1	>0.05	2.56
	30 Mar. 11	17	4.25	59.20	41.79	0.00	A	0.33	1	>0.05	1.93
	12 Nov. 10	5	1.25	1.58	3.80	0.28	R	—	2	>0.05	—
	25 Nov. 10	18	4.50	4.33	2.89	0.41	R	—	6	>0.05	—
<i>Tropisternus laevis</i>	7 Dec. 10	26	6.50	27.67	12.77	0.01	A	2.00	1	>0.05	3.85
	22 Dec. 10	17	4.25	10.92	7.71	0.05	R	—	8	<0.05	—
	7 Jan. 11	21	5.25	6.25	3.57	0.31	R	—	7	>0.05	—
	21 Jan. 11	7	1.75	8.25	14.14	0.00	A	0.47	1	>0.05	0.80
	12 Nov. 10	11	2.75	23.58	25.73	0.00	A	0.36	1	>0.05	1.25
<i>Tropisternus lateralis</i>	25 Nov. 10	24	6.00	16.67	8.33	0.04	R	—	10	<0.05	—
	30 Mar. 11	6	1.50	4.30	8.60	0.04	R	—	1	>0.05	—
<i>Tropisternus longispina</i>	12 Oct. 10	37	9.25	99.58	32.30	0.00	A	0.95	1	>0.05	2.22
	28 Oct. 10	33	8.25	54.92	19.97	0.00	A	1.46	1	>0.05	3.92
	12 Nov. 10	90	22.5	459.67	61.29	0.00	A	1.16	1	<0.05	13.47
	25 Nov. 10	48	12.00	139.33	34.83	0.00	A	1.13	1	>0.05	7.36
	7 Dec. 10	10	2.50	3.67	4.40	0.22	R	—	4	>0.05	—

μ arithmetic mean, S^2 variance, DI Dispersion Index, $Prob.$ probability according to χ^2 distribution, $Dist.$ distribution type, A aggregate distribution (the best fit model is the negative binomial), R random distribution (the best fit model is the Poisson series), K Aggregation Index, df degrees of freedom, $Prob. \chi^2$ probability of χ^2 , λ mean aggregation, calculated with the formula proposed by Arbous and Kerrich (1951).

^a Critical value 2. Higher than 2, aggregations by active processes; lower than 2, aggregation by environmental effects.

T. dilatatus and *T. obesus* were found only in the Tendalero pond, *T. ignoratus* was present only in the Don Luis pond. These three species were observed in few samples and in low abundance. Fontanarrosa et al (2004) reported that aquatic insects of the city of Buenos Aires differed in their frequency of occurrence in different environments, suggesting that these groups would be using such environments differently according to their ecological requirements, which probably explains the presence of *T. dilatatus* and *T. obesus* in one or other pond.

The permanent vegetated habitats are considered more suitable to host some species of water beetles (Byttebier et al 2012). In this study, both ponds had the presence of

aquatic vegetation, but with different composition. The Tendalero pond was characterized by the presence of different microhabitats due to different patches of floating aquatic vegetation and had a higher abundance of individuals ($n=1121$) than the Don Luis pond, which was characterized by a generally homogeneous habitat ($n=729$). It is considered that the presence of macrophytes increases the heterogeneity of habitats (Thomaz & Ribeiro Da Cunha 2010).

The general pattern of spatial distribution observed in *T. ovalis*, *T. collaris*, *T. longispina*, *T. laevis*, and *T. lateralis limbatus*, whose abundance was appropriate for these analyses, showed that the aggregate distribution predominated in the pond with heterogeneous features, with well-

differentiated microhabitats, and that the random distribution predominated in the pond with homogeneous features. This same behavioral pattern was also observed in different populations of *Helochares talarum* Fernández in a pond in Buenos Aires, Argentina, where the distribution was randomly attributed to the environmental homogeneity (Fernández 1990). Later, Fernández & Kehr (1995) also observed a random distribution for adults of *Helochares femoratus* (Brullé) in a permanent pond located within a forest in galleria, and the surface of the pond was covered with permanent aquatic vegetation (mainly *Salvinia rotundifolia* and *Hydrocotyle ranunculoides*).

The spatial disposition that predominated in different populations of the aquatic Coleoptera studied was added. The strongest aggregations in the Tendalero pond would indicate the influence of the type of vegetation in aquatic species of coleopterans. This could be related to the places to support for scrolling (Archangelsky 1997), substrate for oviposition (Corbet 1980, Archangelsky 1997), shelter from predators and sites used as ambush predators (Schnack 1976, Corbet 1980), food availability (Taniguchi & Tokeshi 2004, Da Rocha *et al* 2006), and shelter (Burks *et al* 2001, Rennie & Jackson 2005), provided by the vegetation of different microhabitats. In addition, many other studies conducted in various groups of aquatic insects (Ephemeroptera, Diptera, Trichoptera) showed this type of spatial disposition as the most common (Glass & Bovbjerg 1969, Lamberti & Resh 1983, McAuliffe 1984, Martin & Barton 1987, Ciborowski & Craig 1989).

In general, the greater abundance observed in the Tendalero pond could be related to the heterogeneity of plant substrate (i.e. increased number of macrophytes). The microhabitat formed by *L. laevigatum* was selected by the beetle species studied. It has been documented that macrophytes increase habitat complexity or heterogeneity of aquatic ecosystems, affecting population structure and diversity (Thomaz *et al* 2008, Thomaz & Ribeiro Da Cunha 2010). The size and structure of the root and leaves of aquatic plants influence the richness and species diversity of aquatic fauna (Fontanarrosa *et al* 2013). In this study, the microhabitat formed by *L. laevigatum* in Tendalero ponds presented a more dense root structure than another dominant macrophyte (*Hydrocleys nymphoides*) and was the first microhabitats in which greater richness and abundance of species were recorded, showing the root structure influence on population structure of water beetles.

Based on our data, we conclude that the presence of more complex microhabitats would foster spatial partitioning of populations of aquatic Coleoptera. From the above, it can be inferred that the habitats and microhabitats directly influence the structure and the spatial and temporal variation of water beetles. Aquatic macrophytes are important because they favor or limit the abundance of some species of water

beetles modifying the structure thereof. In addition, macrophytes influence the spatial arrangement of aquatic Coleoptera on the site, benefiting in many cases aggregation of species, showing a marked preference for the sectors formed by plants.

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