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Collembola (Hexapoda) of Malaise traps from two localities in Tlaxcala, Mexico

Collembola (Hexapoda) de trampas Malaise de dos localidades en Tlaxcala, México

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ABSTRACT

Springtails (Collembola) from two temperate forest sites, "Ejido Los Búfalos" (LB) and "Bosque Mágico de Piedra Canteada, Santuario de la Luciérnaga" (PC), in Tlaxcala state, central Mexico, were sampled monthly with Malaise traps. More than 38,000 specimens from 10 species were collected and studied. Although abundance values between both study sites were highly different, no significant effect of locality and sampling date was found according to a two-way ANOVA unbalanced test because of the high variance between recorded data. Shannon's diversity index for LB was H'=0.61 and for PC was H'=1.08, with a significant difference in diversity between both sites. The most abundant springtails were Entomobryidae: *Seira* sp. (n=21,091 individuals in LB and n=6,424 in PC), *Americabrya arida* (6,214 and 2,860), and *Willowsia mexicana* (19 and 852); few Neanuridae: *Pseudachorutes* (241 and 169); Isotomidae: *Isotoma* (2 and 3); Katiannidae: *Polykatianna* (36 and 28); and Tomoceridae: *Pogonognathellus flavescens* (45 and 93). Two genera were only found at PC, Dicyrtomidae: *Ptenothrix* (31) and Entomobryidae: *Lepidocyrtus* (4). This is the first study to record abundance values of these high magnitudes for springtails captured with Malaise traps. Results are compared with previous studies at Mexican and French localities, respectively; Entomobryidae was the most abundant family captured with this method.

Key words: Springtails/ Malaise traps/ variation/ abundance/ Mexico.

RESUMEN

Se muestrearon mensualmente con trampas Malaise los colémbolos (Collembola) de dos sitios de bosque templado, "Ejido Los Búfalos" (LB) y "Bosque Mágico de Piedra Canteada, Santuario de la Luciérnaga" (PC), en el estado de Tlaxcala, centro de México. Se recolectaron y estudiaron más de 38.000 especímenes de 10 especies. Aunque los valores de abundancia entre ambos sitios de estudio fueron muy diferentes, no se encontró un efecto significativo de la localidad y la fecha de muestreo de acuerdo con una prueba no balanceada de ANOVA de dos vías debido a la alta varianza entre los datos registrados. El índice de diversidad de Shannon para LB fue H'=0.61 y para PC fue H'=1.08, existiendo una diferencia significativa en la diversidad entre ambos sitios. Los colémbolos más abundantes fueron Entomobryidae: *Seira* sp. (n=21 091 individuos en LB y n=6 424 en PC), *Americabrya arida* (6 214 y 2 860) y *Willowsia mexicana* (19 y 852); pocos Neanuridae: *Pseudachorutes* (241 y 169); Isotomidae: *Isotoma* (2 y 3); Katiannidae: *Polykatianna* (36 y 28); y Tomoceridae: *Pogonognathellus flavescens* (45 y 93). Solo se encontraron dos géneros en PC, Dicyrtomidae: *Ptenothrix* (31) y Entomobryidae: Lepidocyrtus (4). Este es el primer estudio que registra valores de abundancia de estas magnitudes para colémbolos capturados con trampas Malaise. Los resultados se comparan con estudios previos en localidades mexicanas y francesas, respectivamente; Entomobryidae fue la familia más abundante capturada con este método.

Palabras clave: Springtails/ Malaise traps/ variation/ abundance/ Mexico.

Springtails or Collembola are present in large amounts in different environments and were considered the most abundant "insects" on earth by Hopkin (1998); this is true in many different terrestrial and semiaquatic biotopes under special circumstances, which denotes their importance in terrestrial ecosystems ecology. Some sampling techniques, such as Malaise traps are convenient for catching flying insects, but their efficacy for springtails has not been well evaluated and was not typically intended for their capture. Those traps are generally aimed for collecting Diptera, Hymenoptera, Hemiptera, and Lepidoptera (Mattheus & Mattheus 1971). Malaise (1937) designed them because most flying insects hitting an obstacle respond by flying or crawling upwards, trying to escape, and they finally fall into a collecting jar. Few studies using those traps for collecting springtails have been done.

For instance, during three months of Malaise trap sampling in an Albany mesic forest, east of New York state,

Mattheus & Mattheus (1971) obtained 564 collembolans, which represented 1.4% of the total 40,348 insects captured; traps were placed in four locations 500 m from each other. Cepeda-Pizarro et al. (2013) compared the efficiency of Barber and Malaise traps to study the biodiversity of Hexapoda of three Andean peatlands in the Region of Coquimbo (from 3,100 to 3,800 m a.s.l.), Chile. The total Hexapoda collected by Barber traps was 65,015 specimens, half of them were Collembola (32,581) but they did not identify them to family or species. They also collected 23,202 insects with Malaise traps, however springtails were not counted in this part of the study, and Diptera yielded 84% of Hexapoda.

Pernin & Langlois (2013) studied the Collembola collected with thee traps in the natural national reserve of Valbois, France from May through October 2009 and from April through September 2010. Samples were taken every 15 days. For comparison, they took litter samples only in April of 2010 and processed them in Berlese funnels. Their identification (to species of most specimens) was 17 species in Malaise traps (versus 45 in soil samples), with a total abundance of 1,204 springtails. Haenni & Matthey (1984), in Switzerland, captured 542 springtails, without any further identification. Greenslade & Florentine (2013) captured 2,458 springtails of 20 taxa in Tasmania, with most of them identified as morphospecies. It should be noted that some new species, mainly from Entomobryomorpha (Plumachaetas, Salina) have been described thanks to this collecting method (Oliveira et al. 2019, Oliveira & Cipola 2016).

The only previous Mexican contribution on Collembola captured with Malaise traps was in a tropical dry forest at Chamela Biological Station, Jalisco (Palacios-Vargas et al. 1999). Sampling was done every two weeks for two years (from August 1991 to July 1992) in two watersheds. They collected 2,633 springtails (among 41,443 arthropods) from 14 species, versus 62 morphospecies living in the soil and litter in the same locality (PalaciosVargas & GómezAnaya 1993).

Malaise traps are suitable for collecting climbing springtails, which due to their small size and agility are very difficult to collect, except if fogging techniques are used (Palacios-Vargas 2017; Palacios-Vargas et al. 1998, 1999; Palacios-Vargas & Mejía-Recamier 2017). Their ecological importance on the tree canopy has to do with the degradation and transformation of organic matter in situ, and that they are prey for many different groups including spiders, mites, and some small vertebrates such as lizards and birds.

In order to study the Collembola found in the Malaise traps used in a project to study the taxonomy and ecology of Neuroptera (Marquez López, 2017), with the objective to evaluate and compare Collembola communities of two localities from Tlaxcala using these traps; this is a Mexican state close to Mexico City, yet an area where this group of Hexapoda is almost unknown (Palacios-Vargas 2014).

MATERIALS AND METHODS

Study area

The state of Tlaxcala is in east-central Mexico (Fig. 1); after Mexico City, it is the second smallest state, with only 4,016 Km² (0.2% of the country's territory). It belongs to the Neo-Volcanic Axis physiographic region and has a temperate subhumid climate with rains during summer, mean annual temperature of 14.8°C, mean annual precipitation of 1,728.51 mm, and the lowest elevation in the state of 2,200 m, while Nanacamilpa's municipality (where sampling sites are located), has a highest daily temperature average of 22°C during the temperate season, late-March through early June (INEGI 2014). Sampling took place at two sites, "Ejido Los Búfalos" (LB; 19°28'14.8" N, 98°35'17.1" W; 2,896 m a.s.l.) and "Bosque Mágico de Piedra Canteada, Santuario de la Luciérnaga" (PC; 19°27'35.2" N, 98°35'46.9" W; 2,837 m a.s.l.), both managed forested areas aimed to preserve firefly populations through ecotourism. Both sites hold pine-oak forest, with scattered areas of sacred fir (Abies religiosa) and other tree species such as Texas madrone (Arbutus xalapensis) (Ramírez-Albores et al. 2014).

Collecting techniques

Three Malaise traps were placed at each collecting locality (Fig. 2). Sampling started at LB on September 25, 2015 (Fig. 3), while in PC traps were set up on December 08, 2015 (two traps) and February 06, 2016 (one trap) (Fig. 4), with a total of 51 samples obtained from LB and 43 from PC, as sampling ended in March 2017 (Table 1). The traps operated in the field continuously, with samples picked up and ethyl alcohol (96%) replaced every month. Specimens were sorted out at Instituto de Biología-UNAM in small jars and then transported to Laboratorio de Ecología y Sistemática de Microartrópodos, Facultad de Ciencias-UNAM for slide preparations, morphospecies identification, counting, and spreadsheet recording.

Data Analysis

The effect of locality and sampling date on springtail abundance was evaluated by a two-way ANOVA test for unbalanced designs (Shaw & Mitchell-Olds 1993). Normality and homocedasticity in data were present. The analyses were performed using Statistica 8.0 software (StatSoft 2007). Also, Shannon's diversity index was calculated for the community at each locality and the difference between diversity index values was analyzed by a modified t-test (Magurran 1988), with PAST software (Hammer et al. 2001). In order to evaluate the springtails community composition in localities and sampling months a cluster analysis was performed using single linkage as amalgamation method and Euclidian distances as distance measuring, using also Statistica software (StatSoft 2007).

Results were compared with a study carried out in Chamela, Jalisco, Mexico (Palacios-Vargas et al. 1998), and with another from the national reserve of Valbois, France (Pernin & Langlois 2013).

RESULTS

A total of 38,115 springtails of 10 morphospecies were obtained from 94 Malaise trap samples. Most specimens were members of the family Entomobryidae, with 70% belonging to the genus *Seira*. To our knowledge, this study is the first one to record such high abundance values of springtails through this kind of traps (Table 2).

Springtail species obtained were *Americabrya arida* (n=6,214 in LB and 2,860 in PC), *Seira* sp. (21,091 and 6,424) and *Willowsia mexicana* (19 and 852), *Lepidocyrtus* sp. (0 and 4); *Pseudachorutes* sp. (241 and 169), *Isotoma* sp. (2 and 3), *Ptenothrix* sp. (0 and 31), and *Polykatianna* spp. (36 and 28). Combined, the abundance of *A. arida* and *Seira* sp. represented 96% of the total amount of Collembola captured, with *Seira* sp. outnumbering all, while other genera represented less than 4% of the total abundance.

Although the specimen's abundance was very high (27,651 at LB and 10,464 at PC), the species richness was low (10 morphospecies). Nevertheless, the total abundance value recorded at each site had no significant effect from locality and sampling date, according to the two-way ANOVA unbalanced test (Locality: $F_{1,7,3} = 0.64$, p>0.05; date: $F_{17,73} = 1.23$, p>0.05), because of the high variance of recorded data, mean of individuals by trap, and standard deviation, which for LB was 536.59±1679.90, and for PC was 243.35±564.22 (Fig. 5). Shannon's diversity index for LB was H'=0.61, and for PC was H'=1.08, so a significant difference was found in the diversity of both sites ($t_{15933} = 39.76$, p<0.05).

The abundance value of springtails for each month varied highly (Fig. 6), even if numbers are expressed in a logarithmic scale. When a similarity dendrogram is done for each month, there is not a clear pattern, nevertheless at low scale, it seems that most of the close relations are between samples of the same localities and the same season of the year, thus most months of dry season of LB locality are grouping and are close to dry months in PC locality (Fig. 7).

DISCUSSION

Few studies are known using Malaise traps as a sampling technique to explore springtail populations or diversity. Most of them have been done only for few months, for reasons such as season preference or authors' selection of time of highest biological activity yet results from more than a year of collecting may depict a broader scenario. At both localities, we found a very high abundance of Americabrya arida and Seira sp., both recorded for the first time through Malaise trap collecting. Willowsia mexicana, Isotoma sp., Ptenothrix sp., Polykatianna sp., Lepidocyrtus sp., Pogonognathellus flavescens (Tomoceridae), and Pseudachorutes sp. (Neanuridae) were represented with relatively few specimens. The difference in species abundance documented in both localities, even with no positive statistical significance, might be due to factors such as climate, and microenvironmental differences and

requirements of the springtail's species.

In addition, a monthly variation in abundance values is evident, although there were no significant differences between both study sites. It may be possible that the variation in abundance of Collembola is dependent on other biological factors, such as moss or lichens living on the bark of trees. According to Gressitt & Gressitt (1962), Malaise trap results are influenced by local circumstances of topography, density of vegetation, relation with wind, water, as well as temperature, precipitation, and location of traps in shade or sun. In addition, the time of the year might be reflected in taxa composition and abundances, as many groups have seasonal occurrences.

When comparing springtail abundance values from the two study sites at Tlaxcala with that from the tropical dry forest of Chamela, Jalisco (Palacios-Vargas et al. 1999), where only 2,633 specimens were collected, there is a huge difference. Also, if they are compared with results at Valbois, France, where 1,204 total Collembola were captured, the difference is even bigger (Table 2). As a general pattern, Entomobryidae seems to be the most common family in many places across geography, however, represented by a different dominant genus in each locale. In Tlaxcala, Seira sp. was the most abundant (n=27,515), while in Chamela Lepidocyrtus spp. (397), in Valbois Entomobrya (1020), and Tasmania Lepidocyrtoides (1,010) were the most abundant genera. Members of Pseudachorutes (Neanuridae) can climb trees and might be an important element of the springtail community in these forested environments because they have specialized stylet-like mouthparts, which enable them to feed on fungi or moss.

When studying the abundance of arboreal arthropod taxa at two localities, Rivière Bleue and Pindaï, New Calédonie, Guilbert (1997) found that Collembola captured through fogging represented 37% and 13.2%, with 23,652 and 14,557 specimens, respectively (but they were not identified to family or genus), and their trophic guild at both sites was epiphyte grazers. Such arboreal species depend on many factors such as weather, wind, and humidity, which allow moss, lichens, and other epiphytic plants to create a rich environment for their development and reproduction and support them to have very big communities of a few species.

CONCLUSIONS

Members of 10 species of Collembola were collected in two study sites of Tlaxcala through 17 months of sampling using Malaise traps, with the largest relative abundance presented by members of Entomobryidae (96%), while the remaining value (less than 4%) was held by other families. The most abundant taxa were *Seira* sp. and *Americabrya arida*, which are epiedaphic and usually live on the litter surface and epiphytic habitats, including bushes and the tree canopy. The abundance values obtained appear to indicate populations of the same community because the statistical analysis did not find a significant difference among study sites, located ca. 1.5 km from each other.

In this study, sampling was continuous for 17 months, and when the total abundance of Collembola (n=38,115) captured by Malaise traps at Tlaxcala, is compared with numbers from other localities, this appears to be the highest number of springtails ever collected through this technique. These results provide good evidence that springtails that climb trees might be a highly abundant component of terrestrial arthropod communities, and so they may be responsible for a relevant ecological role.

Even when species richness of the springtail community was low, which generally agrees with a high elevation and temperate climatic conditions, important abundance values were recorded at both study sites, which may be explained through a comparative study of substrates, such as coverage by moss, lichens, and other epiphytic plants at each site. Further studies comparing soil- versus arboreal springtail communities might offer insight into springtail significance in forest ecosystems.

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	LB	PC
Starting date (Malaise trap set up)	25.ix.2015 (3 traps)	08.xii.2015 (2 traps) &
		06.ii.2016 (1 trap added)
Closing date (Malaise trap pick up)	02.iii.2017	03.iii.2017
Duration of sampling (months)	17	15
Total samples (<i>n</i>)	51	43

Table 1. Sampling regime of springtails with Malaise traps in Tlaxcala, Mexico. LB = Ejido Los Búfalos, PC = Piedra Canteada.

GENERA	MEX Jalisco	MEX T	MEX Tlaxcala	
	CHAMELA	PC	LB	VALBOIS
Adisianus	19	0	0	0
Allacma	0	0	0	12
Americabrya	0	2,860	6,214	0
Calvatomina	388	0	0	0
Ceratophysella	5	0	0	0
Dicyrtomina	0	0	0	3
Entomobrya	0	0	0	1,020
Isotoma	2	3	2	0
Polykatianna	0	28	36	0
Lepidocyrtus	397	4	0	40
Orchesella	0	0	0	125
Pogonognatellus	0	93	45	0
Proisotoma	0	0	2	0
Pseudachorutes	0	169	241	0
Ptenothrix	0	31	0	0
Rapoportella	8	0	0	0
Salina sp.	225	0	0	0
Seira	268	6,424	21,091	5
Sminthurus	275	0	0	0
Tomocerus	0	0	0	8
Willowsia	0	852	19	3
TOTALS	1,587	10,464	27,651	1,216



Figure 1. Location of study sites in Nanacapilma, Tlaxcala, Mexico.



Figure 2. Malaise trap setting in Los Búfalos, Tlaxcala, Mexico.



Figure 3. Mixed vegetation habitat at Los Búfalos, Tlaxcala, Mexico.



Figure 4. Mixed vegetation habitat at Piedra Canteada, Tlaxcala, Mexico.



Figure 5. Abundance average and standard deviation of Collembola collected with Malaise traps in two localities of Tlaxcala, Mexico. Ejido Los Búfalos, left; Piedra Canteada, right.



Figure 6. Monthly variation of abundance and standard deviation of Collembola collected with Malaise traps in two localities of Tlaxcala, Mexico. LB = Ejido Los Búfalos, PC = Piedra Canteada.



Figure 7. Dendrogram of cluster analysis from Collembola community composition collected with Malaise traps in two localities of Tlaxcala, México.