UNIVERSIDADE FEDERAL DO PAMPA

ETIELY KARNOPP

USO DE HABITAT POR ESCORPIÕES DO BIOMA PAMPA: EFEITO DO USO DO SOLO E DO RELEVO

São Gabriel
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Trabalho de Conclusão de Curso apresentado ao Curso de Ciências Biológicas da Universidade Federal do Pampa, como requisito parcial para obtenção do Título de Bacharel em Ciências Biológicas.

Orientadora: Márcia Regina Spies

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“O mundo e o universo são lugares extremamente belos, e quanto mais os compreendemos mais belos eles parecem.”

Richard Dawkins

TEXTO EXPLICATIVO DA ESTRUTURA DO TRABALHO

Este Trabalho de Conclusão de Curso se estrutura em torno de um artigo científico escrito em língua inglesa pela autora durante o desenvolvimento do curso. Assim, sua estrutura segue as normas de publicação da revista Studies on Neotropical Fauna and Environment.

SUMÁRIO

HABITAT USE BY SCORPIONS IN PAMPA BIOME: THE EFFECT OF LAND USE AND RELIEF SLOPE

ABSTRACT

Many factors influence the habitat choice by scorpions, as soil properties, microclimate of shelters or hunting territories, extent and nature of vegetation, prey abundance, and the animal's age. The objective of this study was to describe the habitat use (native grassland with livestock x *Eucalyptus* afforestation; upland x lowland) by the scorpion community in the Pampa biome, as well as to teste the relationship of environmental variables with the scorpion abundance. We recorded 608 scorpions: 528 individuals from *Tityus uruguayensis*, and 80 from *Bothriurus bonariensis*. Both species were found in all areas searched, and they co-occurred in transects of all areas using all habitat sampled. However, *T. uruguayensis* was especially abundant in the transects of upland in *Eucalyptus* croplands, while *B. bonariensis* was more abundant in uplands from native grasslands with livestock, presenting low occurrence in *Eucalyptus* afforestation. Our results show the importance of preserving the Pampa biome to maintain the structure of scorpion assemblage conserved, since *Eucalyptus* afforestations were not a habitat successfully used by *Bothriurus bonariensis* and favored the occurrence of *Tityus uruguayensis,* allowing them to reach very high abundance, which may cause several environmental imbalances.

Key words: *Bothriurus bonariensis, Eucalyptus* afforestation, native grasslands, *Tityus uruguayensis.*

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RESUMO

Muitos fatores influenciam a escolha de habitat por escorpiões, as propriedades do solo, o microclima de abrigos ou territórios de caça, a extensão e a natureza da vegetação, a abundância de presas e a idade do animal. O objetivo deste estudo foi descrever o uso de habitat (campo nativo com pecuária x cultivo de *Eucalyptus*; coxilha x baixada) pela comunidade de escorpiões do bioma Pampa, bem como testar a relação de variáveis ambientais com a abundância de escorpiões. Os escorpiões foram localizados por busca ativa, com o auxílio de uma lanterna ultravioleta. Em cada período de amostragem registramos dados de temperatura média (ºC) e umidade do ar (%). Foram registrados 608 escorpiões: 528 indivíduos de *Tityus uruguayensis* e 80 de *Bothriurus bonariensis*. Ambas as espécies foram encontradas em todas as áreas pesquisadas e coocorreram em transectos de todas as áreas usando todo o habitat amostrado. No entanto, *T. uruguayensis* foi especialmente abundante nos transectos de coxilha em áreas de cultivo de *Eucalyptus*, enquanto *B. bonariensis* foi mais abundante em coxilha de campo nativo com pecuária, apresentando baixa ocorrência no cultivo de *Eucalyptus*. Nossos resultados mostram a importância de preservar o bioma Pampa para manter conservada a estrutura das comunidades de escorpiões, uma vez que as áreas de cultivo de *Eucalyptus* não foram um habitat usado com sucesso por *Bothriurus bonariensis* e favoreceram a ocorrência de *Tityus uruguayensis*, permitindo atingir uma abundância muito alta, o que pode causar vários desequilíbrios ambientais.

Palavras chave: *Bothriurus bonariensis,* campo nativo, cultivo de *Eucalyptus*, *Tityus uruguayensis.*

INTRODUCTION

Vegetation structure influences the amount of sunlight exposure of the soil surface, habitat moisture, temperature, and physical structure and heterogeneity of the leaf litter (Medianero et al., 2007). Conversions of grasslands in forests currently affect some of the world's most productive regions and have the potential to modify many soil properties, soils under *Eucalyptus* croplands in Pampa biome had substantially higher acidity than native grassland soils (Jobbágy and Jackson, 2002). Brazilian grasslands have undergone an extensive transformation and large areas of native grasslands have been converted to afforestation of conifers, *Eucalyptus* (Bond and Par, 2010) and soybean cultivation (Castanheira and Freire, 2013). The density of scorpion populations is highly variable and depends on environmental factors. These factors influence their choice of microhabitat, some of them are the texture of substrate, the microclimate of shelters or hunting territories, extent and nature of vegetation, prey abundance and the animal's age (Stockmann, 2015; Stockmann and Ythier, 2010). The processes involved in habitat selection usually involve responses to environmental conditions that promote growth, survival and reproductive success (Polis, 1981; Lira et al., 2013).

Not well known outside and even within Brazil, the Pampa grassland is among the most species-rich pastures in the world (Overbeck et al., 2007). In addition to its unique biodiversity, it provides essential ecosystem services including water supply, livestock forage production, and carbon storage. Livestock grazing is one of the few land uses that can be reconciled with the biodiversity of non-forested areas. For example, natural grasslands in the Pampa biome allow sustainable large-scale livestock production (Overbeck et al., 2007). Grasslands constitute the main habitat of a significant portion of the fauna of southern Brazil and, especially, of Rio Grande do Sul, where this ecosystem occupies 69% of the Rio Grande do Sul state surface (Behling, 2009; IBGE, 2019). The general level of knowledge about the terrestrial invertebrates of southern grasslands is precarious, being below the "bad", according to the judgment of experts consulted to assess the state of knowledge of Brazilian biodiversity (Lewinsohn, 2006). As a result, very little can be said about the richness, composition and peculiarity of the several groups of southern Brazilian invertebrates, although such groups

represent most of the biodiversity of the southern grasslands, as well as any other terrestrial ecosystem on the planet (Benke, 2009).

In southern Brazil, tree afforestation on native grasslands totaled 1.9 million hectares in 1996 (IBGE, 2005), and in the state of Rio Grande do Sul *Eucalyptus* croplands alone currently occupy 284,701 ha (ABRAF, 2013). Tree plantations will eventually create a new pattern of landscape, where open grasslands are represented by patches of native vegetation in an array of shaded areas composed largely of exotic trees (Audino et al., 2011). The negative impacts of forestry on biota have been reported since *Eucalyptus* croplands are described as 'biological deserts' because they have little or no value to native species (Kanowski et al., 2005). Thus, monocultures are expected to have direct and indirect effects on biota due to environmental changes, such as physicochemical conditions of the substrate, availability of resources, allelopathic effects, and shading (Machado et al., 2012; Saccol et al., 2017; Toscano-Gadea, 2013).

Anthropogenic land use was a determinant key on scorpion abundance and diversity, but not on the species richness, independent of the type of non-natural habitats (Lira et al., 2019). In this way, differences in species habitat requirements also play a role in scorpion distribution in a heterogeneous environment (Lira et al., 2019). These arachnids are primarily solitary and sedentary arthropods and the spatial distribution in each microhabitat dependent on the scorpion species. Information on microhabitat distribution is crucial for understanding the processes of species coexistence (Lira et al., 2013). Therefore, the presence of conspecifics and heterospecifics in the environment will most likely result in substantial competition for the resources of food and shelter and may decisively influence habitat selection (Polis and McCormick, 1987; Lira et al., 2014).

In the southeastern Rio Grande do Sul, Brazil, two species of scorpions coexist, *Bothriurus bonariensis* (Koch, 1843) and *Tityus uruguayensis* Borelli, 1901 (Olivero et al., 2012; Stockmann & Ythier, 2010). *Bothriurus bonariensis* is a black or reddish-brown scorpion of the Bothriuridae family that can measure up to six centimeters in length (Lourenço, 2002). *Tityus uruguayensis* is a small yellowish scorpion of the Buthidae family, facultative parthenogenetic, which measures approximately four or five centimeters in length (Toscano-Gadea and Costa, 2006; Francke, 2008). These species inhabit a large part of the Pampa biome, from central Argentina, Uruguay and southern

Brazil (Mattoni and Acosta, 2005). The influence of specific microclimates, as well as soil composition, rock types, extreme temperature seasonality and food availability are limiting factors in the distribution of these animals, as well as the reproduction type of species (Marcussi et al., 2011). The objective of this study was to describe the habitat use by a scorpion assemblage in the Pampa biome, testing the effects of land use (native grassland with livestock x *Eucalyptus* croplands) and relief slope (upland x lowland), as well as to test the relationship of environmental variables (temperature and relative air humidity) on the scorpion abundance. We hypothesize that is that *Eucalyptus* croplands would be less used by scorpion assemblage than native grasslands since several species are sensitive to anthropogenic land use (Lira, 2019).

MATERIALS AND METHODS

Study area

The study was performed in the physiographic region known as Serra do Sudeste, which is characterized by an undulated relief, with altitudes varying from 200 to 500 m, it sits on an old geological formation, the Precambrian granitic shield (Boldrini, 1997). Covering the northern part of the Pampa biome, the Serra do Sudeste is in the southeastern portion of the state of Rio Grande do Sul. This physiographic region covers 15% of the total surface of the state (Rambo, 2005) and present climate as humid subtropical, Cfa type, according to the Köppen classification (Moreno, 1961). Annual rainfall ranges from 1,200–1,600 mm and seasonal variations can occur, concentrating the rainfall in winter (Porto, 2002; Overbeck et al., 2007). The temperatures can range from <15°C, in the winter, to 40°C, in the summer (Porto, 2002).

Sampling

We sampled scorpions in a private property and in a logging company (CMPC) of Santa Margarida do Sul, Rio Grande do Sul state. We explored uplands and lowlands reliefs in native grassland with livestock and *Eucalyptus* afforestation, totaling four areas (grassland/upland - Gup; grassland/lowland - Glow; cropland/upland - Cup; cropland/lowland - Clow). In each area we sampled 10 transects of 100m length and 2m width each, 8m apart of each other (Figure 1). Each area was explored for 1h with a sampling effort of one person per hour. The areas were explored from 10h p.m. to 2h

a.m. The sampling sequence of the areas was drawn in each sampling period, to alternate the sampling time and avoid temporal bias.

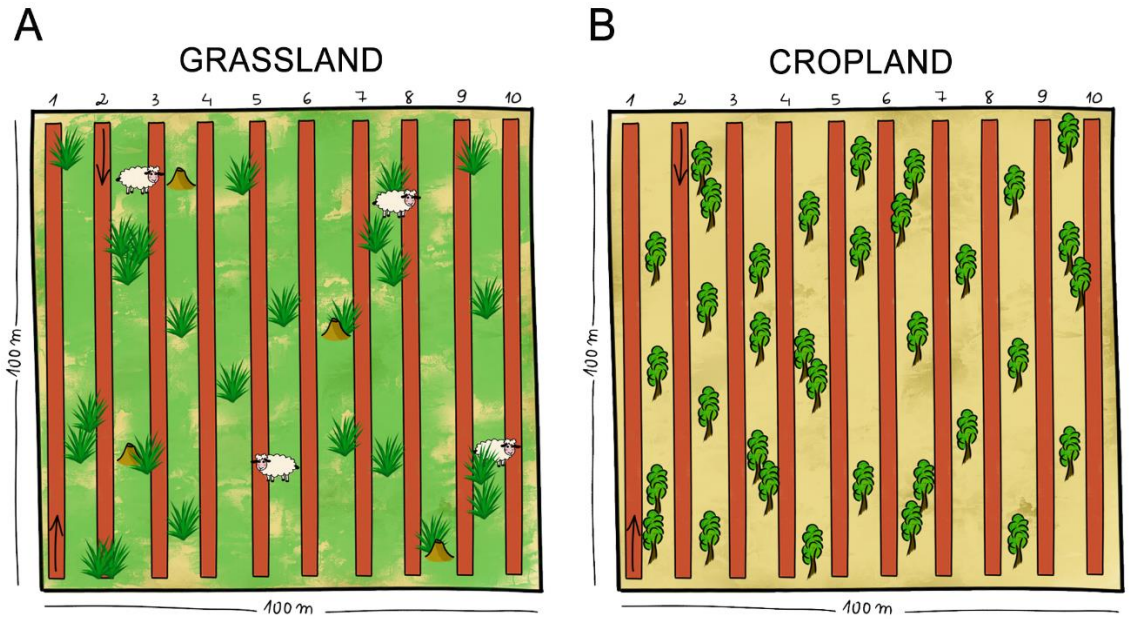

Figure 1. Sampling scheme for native grasslands with livestock (A) and *Eucalyptus* afforestation (B). The red lines represent the transects and the arrows represent the directions. For each land use area there were doubles for the upland/lowland reliefs, totaling four sample areas (grassland/upland; grassland/lowland; afforestation/upland; afforestation/lowland).

The surveys were performed in seven sampling dates: 31 January; 16 February; 01 and 29 March; 15 April; 02 May and 08 June of 2019 (with sampling failure on Clow 16/February; Gup 01/March; Glow 15/April and Gup transects 1,2,3 15/April). Scorpions were located by active search (Nime et al., 2014) along 2m wide transect scanning with the aid of an ultraviolet flashlight (Toscano-Gadea et al., 2015), since scorpions have higher activity at night and a fluorescence property at ultraviolet light (Lowe et al., 2003). All individuals found were counted, marked (by white face paint) to avoid recounting during the same sampling period.

In each sampling period, we obtain average temperature (°C) and air humidity (%) data (obtained from Instituto Nacional de Meteorologia in each collet by hour: http://www.inmet.gov.br/portal/index.php?r=bdmep/bdmep). To characterized the soil properties we also performed analyzes to determine the moisture content present in the soil, where the collected samples were weighed and transferred to a kiln at 105-

110ºC, leaving this condition for 24 hours, after withdrawals were placed in desiccator, cooled and weighed again (EMBRAPA, 1997). The particle size analysis was based on the velocity of the fall of the particles that compose the soil, for normal soils (EMBRAPA, 1997).

Statistical analysis

We analyzed the effect of temperature and relative air humidity on the abundance of scorpions *Bothriurus bonariensis* and *Tityus uruguayensis,* separately, through adjusted a generalized linear mixed model in the *brms* package (Bürkner, 2017; 2018) of R software (R Core Team, 2019). To consider the high amount of zero in the abundance data, in both models, the zero-inflated-poisson family (with log link function) was used. For both models, the response variable was the abundance of scorpions (count data). As predictor variables, were used the relative air humidity and the temperature, and the relief slope (lowland and upland) and the land use (grassland and cropland) were included as variables to control that effects. The identity of each sampling dates (1-7) and the identity of each transect (1-40) was included as random effects. The relationship between habitat use and soil properties was performed by comparing sampled areas with soil texture and moisture information using signed-rank Wilcoxon tests.

A two factor permutational multivariate analysis of variance - Permanova (Anderson, 2017) was performed to assess variance in habitat use by the scorpion assemblage, adopting a fixed/fixed model (i.e. land use x relief slope). Scorpion abundance by transect sampling were square root transformed to decrease the abundance effect of dominant species. Analyses were performed using Primer-E 7 (Anderson et al., 2008; Clarke and Gorley, 2015). Next, we represented the sampling spatial data of the scorpion assemblage through metric-Multi-Dimensional Scaling (mMDS) using the Bray-Curtis similarity coefficient to explore spatial ordination of transects according to the land use and relief slope. We analyzed the spatial distribution of *Bothriurus bonarienesis* and *Tityus uruguayensis* along land use and relief by bubble plot of mMDS (Clarke et al., 2014).

RESULTS

We recorded a total of 608 scorpions: 528 individuals from *Tityus uruguayensis* and 80 from *Bothriurus bonariensis*. All *T. uruguayensis* individuals were adults and only one female had newborn scorpions (not accounted). On the other hand, *B. bonariensis* population was composed of 13 young and 67 adults (31 males and 36 females).

All sampling dates presented records of scorpions, *B. bonariensis* and *T. uruguayensis*. Both species were found in all areas searched, native grassland with livestock and *Eucalyptus* afforestation, as well as in upland and lowland reliefs. However, *T. uruguayensis* was especially abundant in the transects in croplands of *Eucalyptus* in upland, 472 individuals, while only three individuals of *B. bonariensis* were recorded in this area (Table 1, Figure 2). *B. bonariensis* was more abundant in native grasslands in upland (41 individuals X only two *T. uruguayensis*) and lowland (27), presenting low occurrence in *Eucalyptus* croplands (Table 1, Figure 2). Soil moisture and texture do not present significative differences among areas sampled (all signed-rank Wilcoxon tests presented $p>0.05$).

Table 1. Abundance of a scorpion assemblage in uplands (Gup) and lowlands (Glow) of native grassland with livestock, and in uplands (Cup) and lowlands (Clow) *Eucalyptus* afforestation in Brazilian Pampa biome.

	Gup	**Glow**	**Cup**	**Clow**	**Total**
Bothriurus bonariensis	41	27	3	9	80
Tityus uruguayensis	2	37	472	44	528

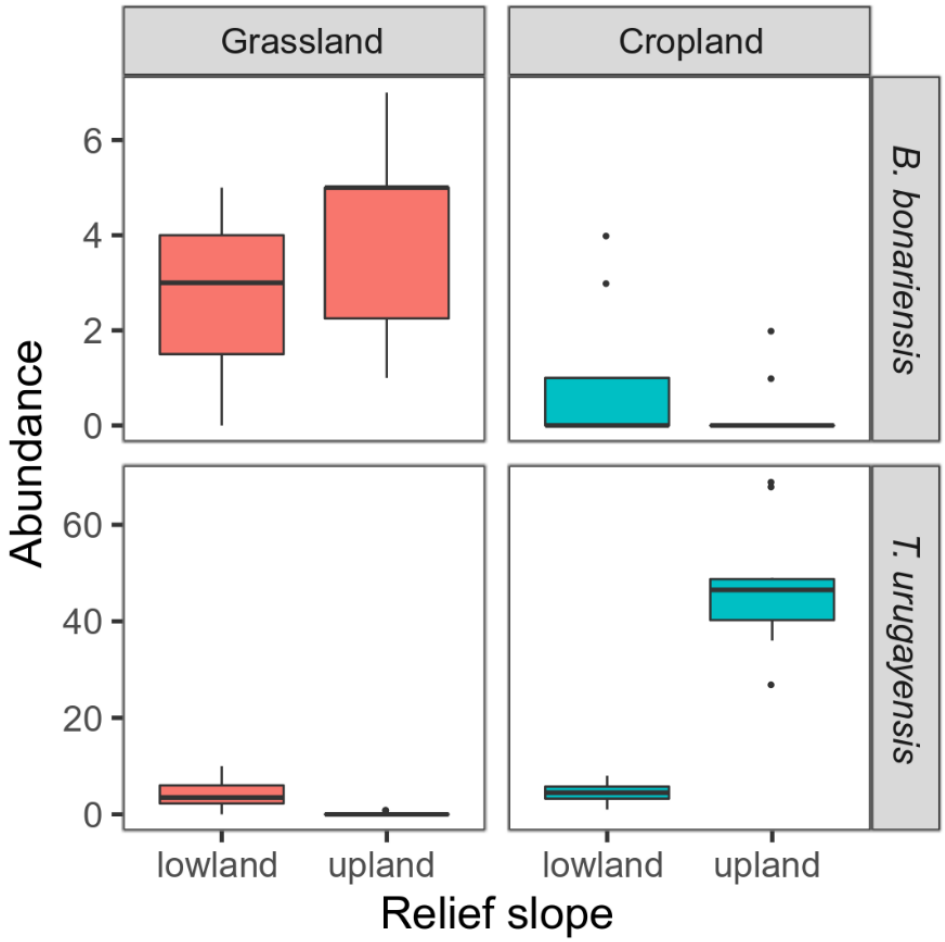

Figure 2. Abundance of *Bothriurus bonariensis* and *Tityus uruguayensis* per sampling area data, in native grasslands with livestock (red boxes) in upland and lowland and croplands of *Eucalyptus* (blue boxes) in upland and lowland in Brazilian Pampa biome. Bold lines represent the median, boxes represent the standard error and the up and low line represent 95% of data.

The temperature positively influences the abundance of *T. uruguayensis* (Estimate: 0.31; SE: 0.10; CI 95%: lower 0.14, upper 0.55; figure 4a), but not influence the abundance of *B. bonariensis* (Estimate: 0.23; SE: 0.20; CI 95%: lower -0.16, upper 0.61; figure 4a). The relative air humidity has not significant effect on the abundance of *B. bonariensis* (Estimate: -0.08; SE: 0.03; CI 95%: lower -0.15, upper -0.01; figure 4b), no either the abundance of *T. uruguayensis* (Estimate: 0.03; SE: 0.02; CI 95%: lower 0.00, upper 0.07, figure 4b).

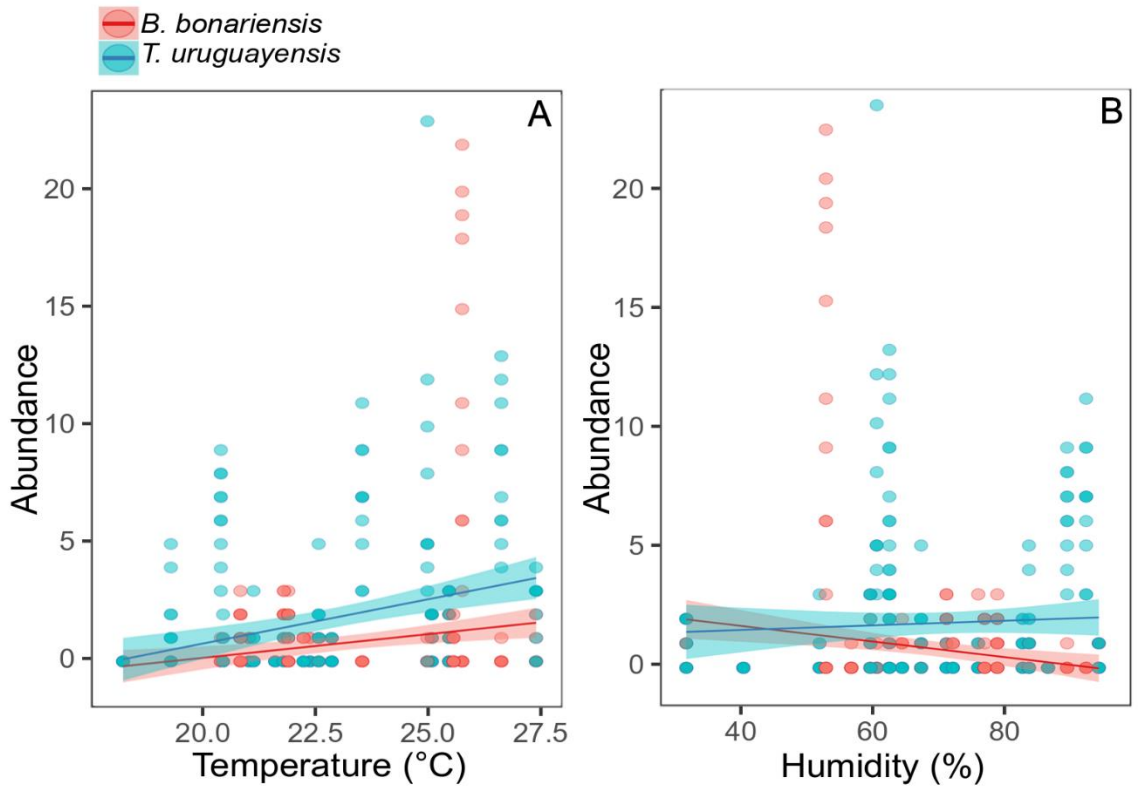

Figure 4. Influence of temperature (A) and air relative humidity (B) on the abundance of scorpions *Bothriurus bonariensis* (red points and lines) and *Tityus uruguayensis* (blue points and lines) in Brazilian Pampa biome.

The evaluation of variance in spatial habitat use by scorpion assemblage showed that both factors tested (land use and relief slope) were significant and that there was interaction among the factors (Table 3). The partitioning of the variance evidenced that land use represented 24% of variance, while relief slope only 9% and the interaction of the factors 33% (Table 3). In fact, the ordination of similarity yielded three groups for the scorpion assemblage (Figure 5): 1) transects where only *B. bonariensis* occurred, mostly characterized by native grassland with livestock upland; 2) transects where both species co-occurred, mostly characterized by native grassland with livestock lowland; 3) transects where only *T. uruguayensis* occurred, mostly characterized by *Eucalyptus* afforestation upland.

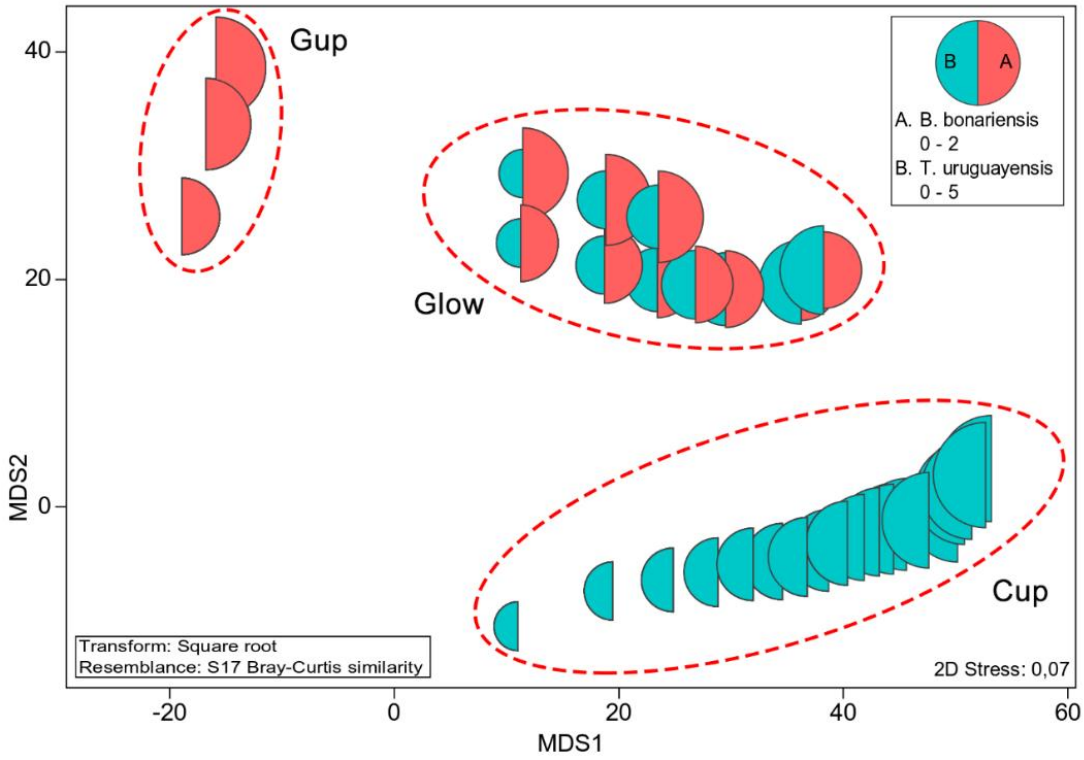

Figure 5. Bubble plot of Metric Multi-Dimensional Scaling representing the spatial habitat use by a scorpion assemblage in native grasslands with livestock and *Eucalyptus* croplands in uplands and lowlands of the Brazilian Pampa biome.

Table 3. Permanova based on Bray-Curtis similarity index for a model of 2 fixed factors (land use and relief) representing the spatial habitat use by a scorpion assemblage in Brazilian Pampa biome, including the variance components (CV%).

Source	MS	Pseudo-F	P(perm)	CV%
land use	34305	65.41	0.0001	24.21
relief slope	5133.9	9.79	0.0001	8.94
land use x relief slope	32707	62.37	0.0001	33.42
residuals	524.4			33.43
Total				100

DISCUSSION

In this study we tested the hypothesis that *Eucalyptus* afforestations were lesser used by scorpion assemblage than native grasslands. Our results showed that indeed this happens on *Bothriurus bonariensis* scorpions, while *Tityus uruguayensis* was favored

by *Eucalyptus* afforestation. Although both species were found in all areas, *T. uruguayensis* was more abundant in *Eucalyptus* afforestation, especially in the transects in upland, while *B. bonariensis* was more abundant in native grasslands, presenting low abundance in *Eucalyptus* afforestation.

The low occurrence of *Bothriurus bonariensis* in *Eucalyptus* afforestation here recorded do not corroborate the results of Toscano-Gadea (2013), that recorded this species in a wide range of subtract types, including *Eucalyptus* croplands, and suggested a low selectivity to potential shelter for this species. In other way, *B. bonariensis* is also pointed as a species with preference to grassland with livestock (San Martín, 1961; San Martín & Gambardella, 1967; Ojanguren-Affilastro, 2005).

The especially high abundance of *Tityus uruguayensis* in upland of *Eucalyptus* croplands seems to be a consequence of habitat changes caused by human interference that favored microhabitat requirements of this species. In fact, *T. uruguayensis* is pointed as a comum species in *Eucalyptus* croplands (Toscano-Gadea, 2013). In this way, our results suggest that *T. uruguayensis* shows a higher ecological plasticity than *B. bonariensis*.

The greater importance of land use evidenced by our results of variance partitioning was largely due to the low occurrence of *Bothriurus bonariensis* in *Eucalyptus* afforestation. In addition, the large data variance explained by the interaction between land use and relief slope was due to the high occurrence of *Tityus uruguayensis* in upland of *Eucalyptus* afforestation. This habitat use pattern may be related to differences in species habitat requirements (Lira et al., 2019). *T. uruguayensis* is considered errant (Stockmann and Ythier, 2010) and *B. bonariensis* fossorial scorpions (Pizarro-Araya et al., 2011). Scorpions that are considered errant animals have a great dispersion capacity (Stockmann and Ythier, 2010). In contrast, scorpions that dig burrows present a lower displacement capacity, since build burrows in new environments are highly dependent on the soil stability (Polis, 1990; Lira et al., 2019). Therefore, errant scorpions can inhabit *Eucalyptus* afforestation, where they can find shelter and food under the litter, while fossorial scorpions can be damaged by the preparation of the soil that results in the destruction of the burrows and it is more costly for the animal to have to frequently build a new burrows, making it difficult for the establishment in the area (Lira et al., 2019). In fact, Bothriuridade scorpions are typical

species from open vegetated areas and can be easily found on burrows, ground and under stones (Lira et al., 2018), being lower abundant in closed environments, such as the *Eucalyptus* croplands (Lira et al., 2019).

The temperature positively influenced the abundance of *Tityus uruguayensis.* The influence of temperature on surface activity is important for scorpions, as it impacts both hunting ability and predator avoidance through such activities as foraging due high temperatures that increase plant growth and cause higher numbers of herbivorous insects, which likely improve food availability for scorpions (Schwerdt, et al., 2016; Wise 1993).

As mentioned before, the similarity analysis recovered three groups regarding habitat use: where only *B. bonariensis* occurred, where both species co-occurred and where only *T. uruguayensis* occurred, characterized mainly by upland *Eucalyptus* croplands. The use of different substrates could reduce the possibility of contact and subsequent conflict between scorpion species (Lira et al., 2013). Smaller species and immature individuals of larger species would avoid microhabitats with larger scorpion species, since they are active at the same time (Polis and McCormick, 1986; Ramos, 2007).

The relief alone was not very important for species occurrence, except for *Tityus uruguayensis*, which had a higher preference for upland, but this was strongly associated with land use *Eucalyptus* afforestation, how our results showed. The low importance of relief slope corroborates the studied developed by Toscano-Gadea (2002) and Toscano-Gadea et al. (2015), which found a low variation on *B. bonariensis* and *T. uruguayensis* abundance among relief slope.

In conclusion, the present study describes the habitats used by two scorpions inhabit the Brazilian Pampa Biome (*Bothriurus bonariensis* and *Tityus uruguayensis*) in native grasslands with livestock and *Eucalyptus* afforestation, with differences in relief slope (uplands and lowlands) and to explore the relationship among the environment and each scorpion species. Our results show the importance of preserving the Pampa biome to maintain the structure of scorpion assemblage conserved, since *Eucalyptus* afforestations were not a habitat successfully used by *Bothriurus bonariensis* and favored the occurrence of *Tityus uruguayensis,* allowing them to reach very high abundance, which may cause other environmental imbalances.

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São Gabriel, 12 de dezembro de 2019.

À Coordenação de Trabalho de Conclusão de Curso do Curso de Ciências Biológicas Bacharelado

Assunto: Submissão de artigo de TCC

Venho por meio deste, ao cumprimentá-lo, informar que o manuscrito defendido pela aluna Etiely Karnopp como Trabalho de Conclusão de Curso será submetido a revista Studies on Neotropical Fauna and Enviroment, porém não o fizemos antes da defesa do TCC para implementar as sugestões realizadas pelos professores da Banca de avaliação do trabalho.

Atenciosamente,

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