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Mite fauna associated with different environments of Brazilian Pampa and a case of predatory mite impairing the control of invasive plant

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Thesis defense in the postgraduate program in Ambiente e Desenvolvimento (PPGAD), at Doctoral level, of the University of Vale do Taquari - Univates, in the research line of Technology and Environment.

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ABSTRACT

The Brazilian Pampa is predominantly a natural pasture region with gallery forest formations. It conserves a rich biodiversity and provides vast biological resources for economic use. Pampa suffers from the replacement of fields by grain crops and tree planting to obtain cellulose. The objectives of this work were to evaluate the composition of the mites community comparing the different environments; to relate mite fauna to microclimatic factors and to know the interactions between mites/plants; description of new species and evaluate the biology of *Euseius stipulatus* (Athias-Henriot) and *Typhlodromus* (*Typhlodromus*) *exhilaratus* Ragusa feeding on *Aculus mosoniensis* Ripka (Eriophyidae) from an invasive plant *Ailanthus altissima* (Mill.) Swingle (Simaroubaceae). Mites were collected from the most representative plant species in the Grassland, Grazing Exclusions and Riparian Forest environments of Pampa biome. To evaluate the diversity of mites between enviroments, the ANOSIM test was used at a significance level of 5%. Four new species of eriophyoid mites were described and illustrated. Regarding the predators Phytoseiidae and Stigmaeidae, the Riparian Forest environment had the highest abundance (72%), followed by Grazing Exclusions (20%) and Pastures (8%). One hundred seventeen species of mites were collected in the three environments, being the majority phytophagous (57%). The Riparian Forest environment forms an independent grouping from Grazing Exclusions and Grassland. The species responsible for the similarity of the Grassland and Grazing Exclusions environments was *Tydeus californicus* (Banks), while to the Riparian Forest, *Brachytydeus formosa* (Cooreman) was the most significant. Air humidity and soil correlated with Riparian Forest indicators, while active photosynthetic effects, temperature and higher wind velocity correlated with Grassland and Grazing Exclusions. *Euseius stipulatus* may make hinder the biological control of invasive plant *A. altissima*. The three environments presented mite species with a high number of plant interactions, greater than ten. Mites were indicators responding to environmental differences evaluated Grassland, Grazing Exclusions and Riparian Forest.

Palavras-chave: Biological Control, Degraded Area, *Euseius stipulatus*, *Typhlodromus* (*Typhlodromus*) *exhilaratus*, Grassland, Riparian Forest.

SUMMARY

1 INTRODUCTION

The planting of *Eucalyptus* spp. (Myrtaceae) is expanding in the Pampa biome due to increasing demand for cellulose (Pillar *et al*. 2002; Santana *et al*. 2008; Overbeck *et al*. 2009). Monoculture affects both the diversity and distribution of organisms in the environment (Rieff *et al*. 2010; Rieiff *et al.* 2016). Changes in plant variety and vegetation patterns can create gradients that also shape the composition and distribution of microarthropods (Rodrigues *et al*. 2010; Rieff *et al*. 2016).

In the fields with sand of the Pampa biome the greatest plant richness is from the families Asteraceae, Poaceae, Fabaceae, Euphorbiaceae, Cyperaceae, Rubiaceae and Myrtaceae (Freitas *et al*. 2010; Pinto *et al*. 2013). In the fields coastal plain, Boldrini *et al*. (2008) observed Poaceae, Asteraceae, Cyperaceae and Fabaceae as the most representative families of the Biome. The same author states that there are floristic variations in the plant formations of Rio Grande do Sul, influenced by local and regional environmental characteristics (Boldrini *et al*. 2008). The landscape matrix plays a critical role in recovery processes (Chazdon 2003).

Disturbances of direct soil impact, compaction and fertility can significantly slow the recovery of natural environments. In these extreme cases, intervention is required to recover species composition (Chazdon 2003; Andrade *et al*. 2015). Proximity to centennial forests is one of the main factors that positively influence vascular plants, due to the limitations of seed dispersal of the species (Coote *et al*. 2013; Derroire *et al*. 2016). Rigorous analyzes are needed to allow adequate comparisons with different mature forest areas to evaluate changes in species composition and regeneration of degraded areas (Chazdon 2003). There are several ecological indicators of environmental problems. These must be scientifically and convincingly grounded for use in management and policy decisions on environmental recovery and conservation (Niemi & McDonald 2004).

In regions with high biodiversity value, such as the Pampa biome, a complete environmental evaluation is rarely possible, in which case bioindicators are used as an alternative (Kimberling *et al*. 2001; Coote *et al*. 2013). Studies are being conducted to develop bioindicators on national and international scales, such as the European indicator system used for Sustainable

Forest Management (CMPFE) (Coote *et al*. 2013). Some of the commonly used bioindicators are vascular plants, bryophytes, spiders and bird diversity (Coote *et al*. 2013). There are studies indicating that soil mites can be environmental bioindicators in *Eucalyptus spp*. (Koehler *et al*. 1999; Rieff *et al*. 2016). Despite the abundance of mites in plants, there are few works that study their potential as environmental conservation bioindicators.

The diversity of mites in plants in the state of Rio Grande do Sul is studied in agricultural crops such as soybean, strawberry and also in associated native plants (Ferla & Moraes 2002; Ferla *et al*. 2007; Reichert *et al*. 2014; Rocha *et al*. 2015). The main mite species found in spontaneously occurring plants near strawberry crops in Rio Grande do Sul are *Typhlodromalus aripo* DeLeon, *Phytoseiulus macropilis* Banks, and *Typhlodromips mangleae* DeLeon, *Neoseiulus tunus* DeLeon and *Phytoseius guianensis* DeLeon (Ferla *et al*. 2007). In soybean-associated plants are *Tetranychus ludeni* Zacher, *Tetranychus urticae* Koch, *Neoseiulus idaeus* Denmark & Muma, *Agistemus riograndensis* Johann & Ferla, *Brachytydeus scutatus* Silva, *Brachytydeus sigthori* Baker, *Brevipalpus phoenicus* Deijus Geijus Leon (Guedes *et al*. 2007; Reichert *et al*. 2014).

The Pampa biome has high species richness, but is not adequately protected in conservation policies. It is necessary to create more conservation units and develop management strategies and conduct research on biodiversity and ecological processes (Overbeck *et al.* 2007; Carvalho *et al*. 2009; Santos & Silva 2011). The objective of this work is to identify the mite species indicating environmental conservation in permanent preservation areas exposed to *E. saligna* cultivation located in the central depression in the Pampa biome, in the state of Rio Grande do Sul. In addition, it aims to develop a monitoring approach that validates biological integrity indices, providing inputs that guide the management of environmental restorations in degraded areas.

The survey of acarine species from the Pampa biome may reveal new environmental services that this biome conserves and can offer. The biodiversity of the Pampa biome underpins ecosystem stability, provides vast biological resources for economic use, and is also culturally important (Roesch *et al*. 2009; Diehl *et al*. 2012; Jaurena *et al*. 2016). It is necessary to preserve native areas, as they support a rich biodiversity of mites (Demite *et al*. 2013; Rocha *et al*. 2015). Maintaining these areas, in addition to favoring the conservation of mite species, is very important to increase the diversity of neighboring agricultural ecosystems (Demite *et al*. 2015).

The Pampa biome suffers from the replacement of fields by grain crops and commercial eucalyptus planting (Pillar *et al*. 2009). The study of the structure of acarine communities in permanent preservation areas can help in understanding the dynamics of populations at different recovery levels, optimizing the efficiency of recovery measures.

Less than 0.5% of the total area of the pampa biome is in protected areas. Immediate actions for the recovery and conservation of the Biome's biodiversity are required (Santos & Silva 2011). Another threat is the introduction of exotic species, mainly associated with forestry (*Acacia* sp., *Eucalyptus* sp. and *Pinus* sp.), and the introduction of grasses such as *Eragrostis plana* Nees (Poaceae) (Medeiros & Focht, 2007).

There are few studies that present the mite fauna of the Brazilian Pampa. Thus, the objectives of this work were to evaluate the composition of the plant mite community comparing the different environments (Grassland, Grazing Exclusions and Riparian Forest); to relate acarofauna to microclimatic factors and to know the interactions between mite/plant and description of new species. Specific objectives: To identify acarofauna associated with species of the central depression plant families in Pampa; To evaluate the environmental indexes of acarofauna associated with different environments of the Pampa biome; To verify the acarine species associated (considered indicators) to the different formations of the vegetation cover in preservation areas; to identify plant species that may be reservoirs of predatory mites efficient in biological control applied in different crops.

The thesis is part of the research line of Technology and Environment of the Ambiente e Desenvolvimento program. This is part of a large project of Bioecology and Mite Control in Agroecosystems and Natural Environment in the state of Rio Grande do Sul. The thesis presents the articles as chapters. Chapters 3 and 4 refer to ecology in the survey of mites in the plants of the Brazilian Pampa. Chapters 5 and 6 refer to taxonomic descriptions of new species found in this survey. Chapter 7, on the other hand, refers to a work that was proposed because of the opportunity for exchange carried out in Italy and with species native to Europe.The chapter 5 was published at the Systematic and Applied Acarology, A2. Chapter 4 was submitted to the International Journal of Acarology, B2 both in environmental sciences. The other 11 papers were published during the doctoral period and are described in chapter 9.

1.1 Exchange

During the months of April and August 2018 an exchange was held at the University of Bari in Italy for the Global Doc program. In this exchange some research activities were carried out to complement the results of the thesis. In the laboratory visited, it was possible to identify and describe four new species of Eriophyidae collected in the Brazilian Pampa. Another work was the investigation of predators that would have the potential to impair the biological control of the invasive plant *Ailanthus altissima* (Mill.) Swingle (Simaroubaceae) by the mite *Aculus mosoniensis* Ripka (Eriophyidae).

2 THEORETICAL FOUNDATION

2.1 Pampa

The Pampa biome located in Uruguay, Argentina, Brazil and Paraguay is characterized by vegetation composed mainly of grasses and other herbaceous species (Carvalho *et al*. 2009). There are differences in plant species within Pampa biome regions due to changes in soil composition and adaptations to climate factors. In addition, there are anthropogenic factors that contribute to the selection of some plant species, such as cattle trampling and grazing (Carvalho *et al*. 2009; Freitas *et al*. 2010; Pinto *et al*. 2013).

South Brazilian native pastures occupy about 13.7 million hectares and support very high levels of biodiversity. The richest species of plant families are Asteraceae (about 600 species), Poaceae (about 400-500), Leguminosae (about 250) and Cyperaceae (about 200) (Overbeck *et al*. 2007). This biome also houses a high diversity of fauna and flora. It provides several important environmental services for the conservation of water resources, pollinators, genetic resources and forage potential for livestock (Pillar *et al*. 2009).

The Brazilian Pampa is predominantly a natural pasture region, with plant formations such as gallery forests, capons (tree islands within the pasture) and shrub forests (Behling *et al*. 2005; Roesch *et al*. 2009). It is accepted that human-induced activities such as grazing and burning are the most important factors preventing the expansion of forest formations on these pastures. Pasture fields should be conserved even if these ecosystems do not belong to the current climate and depend on human disturbances (Behling *et al*. 2007; Carvalho *et al*. 2009).

2.2 Eucalyptus forest gardens and preservation areas

There are a significant number of species of the genus Eucalyptus from different habitats.

Eucalyptus saligna is native to temperate regions (Kahmen *et al*. 2008). For this reason, it adapts well to the climate of Rio Grande do Sul. The traditional forest gardens are of large-scale monoculture. These areas with plantations of *Eucalyptus* spp. has less richness and diversity of organisms than in areas kept with natural vegetation (Rieff *et al*. 2010). So far, the focus of restoration ecology has been mainly on forest vegetation while open vegetation types. They were largely neglected. Although most of Brazil's natural vegetation is forested, different vegetation types cover substantial parts of the country, such as the Cerrado, Pantanal, parts of the Atlantic Forest, and the Southern Fields (Southern Brazilian Pastures). There is inefficiency on the part of the competent bodies in the recovery of natural field areas (Overbeck *et al*. 2013). In this sense, conservation studies and reduction of impacts generated by anthropic action are necessary, making the environments maintain their biodiversity (Laurence *et al.* 2001).

Areas of Permanent Preservation (APP) within forest gardens are governed by federal law (Federal Law 12,651/12) and Normative Instruction (ICMBIO No. 11). This legislation establishes procedures for the elaboration, analysis, approval and monitoring of the execution of the Degraded or Disturbed Area Recovery Project - PRAD, for the purposes of compliance with environmental legislation. It is the professionals' responsibility to define recovery or restoration measures, depending on the state of the PPA area. This should be based on its similar functioning to that of the region's ecosystems and phytophysiognomy. The techniques that can be used are:: planting native species by seedlings or direct seeding; transposition of organic soil or litter with propagules; vegetative propagation of native species; conduction of natural regeneration. The area should also be protected against any degradation action such as invasive species, cattle, fire and erosion. To measure the success of the restoration / recovery, quantitative variables should be monitored and these data obtained on a sample basis. The enforcement of these measures is supervised by the city hall and the State Foundation for Environmental Protection (FEPAM), in the state of Rio Grande do Sul.

2.3 Mite groups reported in Rio Grande do Sul State

The families already found under plants in the state were Ascidae, Cunaxidae, Erythraeidae, Parasitidae, Phytoseiidae, Stigmaeidae, Tydeidae, Anystidae, Cheyletidae, Iolinidae, Tarsonemidae, Tenuipalpidae, Tetranychidae and Suborder Oribatida (Ferla & Moraes 2002; Ferla *et al*. 2007; Reichert *et al*.2014; Rocha *et al.* 2015). These families have different eating habits (Moraes & Flechtmann 2008).

Phytoseiidae receive great attention from the scientific community due to their use as applied biological control agents. Some species of this family are commercially available for use in pest control in agriculture. Thus, they are widely studied to identify the food preference and habitat of the species (McMurtry *et al*. 2013). In 2012, the first version of an electronic database on Phytoseiidae was launched with 2,521 species described (Demite *et al*. 2019). In the state of Rio Grande do Sul, 29 species of Phytoseiidae belonging to 18 genera of the subfamilies Amblyseiinae, Typhlodrominae and Phytoseiinae were recorded in plants (Rocha *et al*. 2015). In this work, Phytoseiidae species related to preserved environments were *Typhlodromips japi* Lofego, Demite & Feres, *Phytoseius litoralis* Silva, Rocha & Ferla, *Iphiseiodes saopaulus* Denmark & Muma and *Iphiseiodes zuluagai* Denmark & Muma. Also found were mites that are generally present in environments with greater human intervention such as *Euseius mesembrinus* (Dean), *Euseius alatus* De Leon and *Amblyseius operculatus* De Leon. Of economic importance and as an applied biological control agent was found *Typhlodromalus aripo* De Leon (Rocha *et al*. 2015).

Stigmaeidae mites live in plants and soil, being considered the most diverse among the Raphignathoidea (Walter *et al*. 2009), with 34 genera and 577 described species (Spongoski 2009; Fan *et al*. 2016). In the state of Rio Grande do Sul, they are observed in several agroecosystems and considered the most abundant predators after Phytoseiidae (Ferla & Moraes 2002; Johann *et al*. 2013). Cunaxidae are also predators that inhabit several environments, common in forest systems, pastures, agricultural fields and areas with anthropological disturbances. Little is known about the region of occurrence of Cunaxidae and its habitats. The number of described species reaches 400 today (Skvarla 2014).

Ascidae sensu Lindquist and Evans (1965) are part of the order Mesostigmata. They have morphological and biological similarities with Phytoseiidae. Krantz & Walter (2009) updated Ascidae so that the Melicharini and Blattisociini tribes of Lindquist and Evans (1965) were elevated to family level. The groups were named Melicharidae and Blattisociidae due to their female structure for sperm storage. Thus, the Blattisociidae family moved from the Ascoidea superfamily, to which Ascidae and Melicharidae belong, to the Phytoseoiidea superfamily (Krantz & Walter 2009).

Tetranychidae are associated with numerous plant species, where they feed on the parenchyma by extracting the cell contents, causing a reduction in the photosynthetic capacity of the plant (Bolland *et al*. 1998; Moraes & Flechtmann 2008). They comprise a large number of important species for national and world agriculture. In Brazil, the occurrence of about 100 species of tetraniquids has already been reported among about 1250 of the family (Bolland *et al*. 1998; Migeon *et al*. 2011). They are concentrated in crops such as soy, strawberry, yerba mate and grapevine. The most important tetraniquids are *Panonychus ulmi* (Koch), *Mononychellus planki* McGregor, *Tetranychus ludeni* Zacher, *Tetranychus urticae* Koch and *Oligonychus yothersi* McGregor (Ferla *et al*. 2005; 2009; Ferla & Botton, 2008; Reichert *et al*. 2014).

Among the Tarsonemidae, approximately 545 species belonging to 45 genera are known. At least seven of them are considered pests of different cultures around the world. The most important species is *Polyphagotarsonemus latus* (Banks), which is widely distributed and attacks dozens of plant species (Moraes & Flechtmann 2008). Species of this group have very varied eating habits and can feed on fungi, algae and vascular plants, as well as predatory species and insect parasites. Species of agricultural importance are mostly polyphagous (Moraes & Flechtmann 2008).

The Tenuipalpidae gather about 875 species grouped in 32 genera. These mites are strictly phytophagous. Five species are considered pests of different cultures worldwide, specifically in Brazil. Even though the number of species that reach the pest level is low, they are still considered of great relevance. Among the best-known species, some are found in several plant species and others are very specific (Moraes & Flechtmann 2008). The number of species described within the Tydeidae group is currently 374, grouped into 58 genera. They are often found on plants and soil, feeding on animals and vegetables. Even having a phytophagous habit, it is not considered a plant pest mite. Some Tydeidae still have mycophagous habits (Moraes & Flechtmann 2008).

Eriophyoidea have vermiform, fusiform or elongated body and two pairs of legs in all stages. Its size is from 0.13 to 0.20 mm in length and 0.5 mm in width in adulthood (Moraes & Flechtmann 2008). This family is made up of strictly phytophagous individuals. Its economic importance is surpassed only by tetraniquids, being considered pest mites in various parts of the world. In Brazil, approximately six species are considered of major economic importance. Some species in this group attack grasses and are less specialized feeding on plants of various genera (Moraes & Flechtmann 2008). Amrine *et al*. (2003) cite the existence of 3,442 species described in 301 genera divided into Eriophyidae, Diptilomiopidae and Phytoptidae.

Suborder Oribatida mites are abundant in soil and litter, and can also be found on plants when environmental conditions are not appropriate. In forests the density is approximately one million individuals per square meter. In 20 years, the number of genera increased from 700 to

1000 and 6000 species were recorded (Moraes & Flechtmann, 2008). Recently, a total of 9.356 oribatid species have been reported in 1.316 genera (Moraes & Flechtmann, 2008).

2.4 Mites as Environmental Indicators

There are few studies that suggest panicle mites as bioindicators. However, some mite species may have greater potential as bioindicators, as they are found in more than one plant species. In Rio Grande do Sul the mites *Iphiseiodes saopaulus* (Denmark and Muma), *Euseius mesembrinus* (Dean) and *Typhlodromalus aripo* DeLeon were recorded in more than five plant species (Rocha *et al*. 2015). Just as there are plants with greater species richness, such as *Psidium guajava* L. (Myrtaceae) recorded with 11 species of mites and *Eugenia cumini* (L.) (Myrtaceae) with four species (Rocha *et al*. 2015). These authors also relate *Typhlodromips japi* Lofego, Demite & Feres and *I. saopaulus* species to more conserved environments and *T. aripo* to more unstable environments.

The diversity of plant mites in conserved environments is influenced by the type of nearby agricultural environment. Phytoseiidae were more abundant in neighboring pasture fragments, while sugarcane cultivation favored the occurrence of phytophagous mites in neighboring forest fragments (Demite *et al*. 2015). McMurtry *et al*. (2013) classifies phytoseiids according to some behavioral and dietary characteristics, so the presence of a group may indicate some characteristics of the environment. In Type I are predators of mites specialized in a particular species, feeding on species of Tetranychidae and Tydeoidea, such as *Phytoseiulus*, *Paraseiulus* and *Typhlodromina*. Type II includes selective predators of tetraniquid mites, most often associated with dense web production, *Neoseiulus* and *Galendromus*. Type III includes species that feed on and reproduce on a wide variety of prey, *Paraphytoseius* and *Phytoseius*. In Type IV there are pollinator and generalist predators that can feed only on pollen, *Euseius*, *Iphiseius* and *Iphiseiodes*. The presence of each type may indicate some environmental conditions of the site, such as plant diversity presence of plants with pollen.

3 ACAROFAUNA ASSOCIATED TO DIFFERENT ENVIRONMENTS IN THE SOUTHERN PAMPA, BRAZIL

ABSTRACT

The brazilian Pampa is predominantly composed by native grasslands with riparian forest formations. It conserves a rich biodiversity and provides vast biological resources for economical uses. The objectives of this work are evaluate the composition of the acarofauna in plants comparing grassland, grazing exclusions and riparian forest environments, relate the mite to microclimatic factors and evaluate the interactions with plants. The mites were collected in the most representative vegetal species in the sampled environments. ANOSIM test at the 5% was used to evaluate the mite diversity among de sites. A total of 117 acarine species were collected from the evaluated environmental, most of them were phytophagous (57%). The air and soil humidity correlate to the Riparian Forest indicators while active photosynthetic effects, as temperature and wind speed, correlate to the Grassland and Grazing Exclusion environments. Concerning the composition of the species, Riparian Forest environment forms an independent group comparing to Grazing Exclusions and Grassland. The species responsible for the similarity of the Grassland and Grazing Exclusions environments are *Brevipalpus* sp.2 and *Tydeus californicus* (Banks). In Riparian Forest, *Brachytydeus formosa* (Cooreman) is more significant. In conclusion, the mites consist of environmental indicators of microclimatic factors and landscape. All the sampled sites showed great interaction between mite and plants.

Keywords: Natural Environments; Indicator; Grasslands; Riparian Forest

3.1 INTRODUCTION

The Pampa biome occurs through the Argentina, south of Brazil, Uruguay and Paraguay (Carvalho *et al*. 2009). In the brazilian Pampa dominates native grassland with riparian forest, tree islands within grasslands and shrub forests (Behling *et al*. 2005; Roesch *et al*. 2009). There are floristic variations of Pampa plant formations in the Rio Grande do Sul state, influenced by local and regional environmental characteristics (Boldrini *et al.* 2008). The vegetal species contrast within the Pampa regions that is justified by the soil composition changes and adaptation to climatic factors. Moreover, there are anthropogenic factors that contribute to the selection of some plant species, such as cattle trampling and grazing (Carvalho *et al*. 2009; Freitas *et al*. 2010; Pinto *et al*. 2013).

The microclimatic gradient that forms in the environments with different phytophysiognomies in natural or anthropized matrices can influence the richness, composition and the life cycle of the associated biota. These gradient triggers directly and indirectly affect the biotic and abiotic changes (Murcia 1995). Thus, as the landscape gradually or abruptly changes, variations in radiation, wind speed and humidity percentages occur (Murcia 1995).

Immediate actions for recovery and conservation of the Pampa's biodiversity are necessary, as only 3.3% of the whole area is protected by conservational units (Santos & Silva 2011; MAPA 2019). One of the threats is the introduction of exotic species, mainly associated to forestry (*Acacia* sp., *Eucalyptus* sp. and *Pinus* sp.), and of grasses, like *Eragrostis plana* Ness (Medeiros & Focht 2007). The *Eucalyptus* sp. is expanding in the Pampa due to the increase of celluloses demand (Pillar *et al*. 2002; Santana *et al*. 2008). This monoculture affects the diversity as much as the distribution of different organisms in the environment (Rieff *et al*. 2010; Rieiff *et al.* 2016). Thus, changes in the variety of vegetations patterns can create gradients that can also influence the composition and distribution of microarthropods (Rodrigues *et al*. 2010; Rieff *et al*. 2016). In Brazil, the focus of the restoration has been mainly in forest vegetation while the grasslands vegetations types have been largely neglected (Overbeck *et al*. 2013). Regarding that, conservation studies and reduction of impacts resulting from anthropic action are necessary to promote the environmental biodiversity preservation (Laurence *et al*. 2001).

Direct impact disturbances in soil, compaction and fertility can significantly slow the recovery of natural environments. In these extreme cases, an intervention is required to restore the species composition (Chazdon 2003; Andrade *et al*. 2015). The landscape matrix plays a critical role in the recovery processes (Chazdon 2003). Rigorous analysis are required to allow adequate comparisons with different areas to assess changes in species composition and regeneration of degraded areas (Chazdon 2003).

Ecological indicators of environmental problems can be used in the management and policy decisions on environmental restoration and conservation (Niemi & McDonald 2004). The

Pampa biodiversity supports the ecosystem stability, and provides a vast biological resources for economical use (Roesch *et al*. 2009; Diehl *et al*. 2012; Jaurena *et al*. 2016). The native areas preservation, in addition to favoring the conservation of predatory mite species, is very important to increase the diversity of neighboring agricultural ecosystems (Demite *et al*. 2015). Native areas support a rich biodiversity of mites (Demite *et al*. 2013; Rocha *et al*. 2015), however, there are a few studies that suggest plant mites as indicators. Some mite species are related to more than one plant species and, thus, may have an increased potential as environmental indicators (Rocha *et al*. 2015).

Acarine species previously found on plants of the Rio Grande do Sul state belongs to the Ascidae, Cunaxidae, Erythraeidae, Parasitidae, Phytoseiidae, Stigmaeidae, Tydeidae, Anystidae, Cheyletidae, Iolinidae, Tarsonemidae, Tenuipalpidae and Tetranychidae families and to the Oribatida suborder (Ferla & Moraes, 2002; Ferla *et al*. 2007; Reichert *et al*. 2014; Rocha *et al*. 2015). These families present the most diversified feeding habits (Moraes & Flechtmann, 2008) such as the phytophagism, predatorism, polynivorism, saprophtism and parasitism (Krantz & Walter 2009).

The most important phytophagous belongs to the Tetranychidae, Tarsonemidae, Tenuipalpidae and Eriophyidae and have economical importance for reaching the pest status in the most diversified agricultural crops worldwide (Bolland *et al*. 1998; Ferla *et al.* 2005; Ferla & Botton 2008; Johann *et al*. 2009; Migeon *et al*. 2011; Reichert *et al*. 2014;). The predaceous acarine species associated to plants in the southern of Brazil belongs to the Cunaxidae, Phytoseiidae and Stigmaeidae (Ferla & Moraes 2002; Ferla *et al*. 2007; Walter *et al*. 2009; Spongoski 2009; Reichert *et al*. 2014; Rocha *et al*. 2015; Fan *et al*. 2016). Besides the phytophagous and predaceous mites, some acarine species may present some diversified feeding habits. For example, the Iolinidae, Tydeidae and Triophtydeidae can be predaceous, phytophagous, saprophytes, micophages and can also feed with arthropods in decomposition (Moraes & Flechtmann 2008; Krantz & Walter 2009). Other families with different feeding habits are the Histiostomatidae, Acaridae, Winterschmidtiidae and Suidasiidae that are generally considered mycophages and saprophytes (Moraes & Flechtmann 2008; Krantz & Walter 2009).

Researches on the Pampa's mite species may reveal new environmental services that this biome can provide. There is no study on acarine of Pampa fields native areas in Brazil. Few studies relate the occurrence of mites to microclimatic factors that change in forest and country environments (Tixer, 2018). Thus, the objectives of this work are evaluate the composition of the acarofauna in plants comparing grassland, grazing exclusions and riparian forest environments and evaluating the interactions with plants.

3.2 MATERIALS AND METHODS

3.2.1 Study area

The sampled units are located in permanent preservation areas with comercial eucalyptus forest in Pantano Grande municipality, Rio Grande do Sul state, Brazil (30 ° 19'36,59 "S e 52 ° 29'24,83" W) (Figure 3.1). These areas belong to a phytophysiognomies shrub fields in the central depression of Pampa and are classified as Cfa, thus with well defined summer and winter seasons and fluctuations of annual precipitation that range from 1.600 to 1.900 mm/m² (Peel *et al*. 2007). The soil is a red dystrophic argisol (Streck *et al*. 2018). These areas were used for agriculture and cattle breeding up to 2006 when the eucalyptus plantation was defined as an environmental recovery tool for the area.

Nine sites were defined as conservation units since 12 years ago and areas greater then 0.5 ha were selected and split into three environments: **Riparian Forest** – with tree vegetation occurring in the border of water bodies within the site. It comprises a phytophysiognomic element, named Seasonal Semideciduous Forest, which has high leaf fall in some periods of the year and litter accumulation; **Grazing Exclusion** – with environmental recovery actions, enclousured since six years ago, with shrubby vegetation of spontaneous growing; **Grassland** – with compacted soil and herbaceous vegetation, without enclosure or recovery actions and free access for cattle pasture.

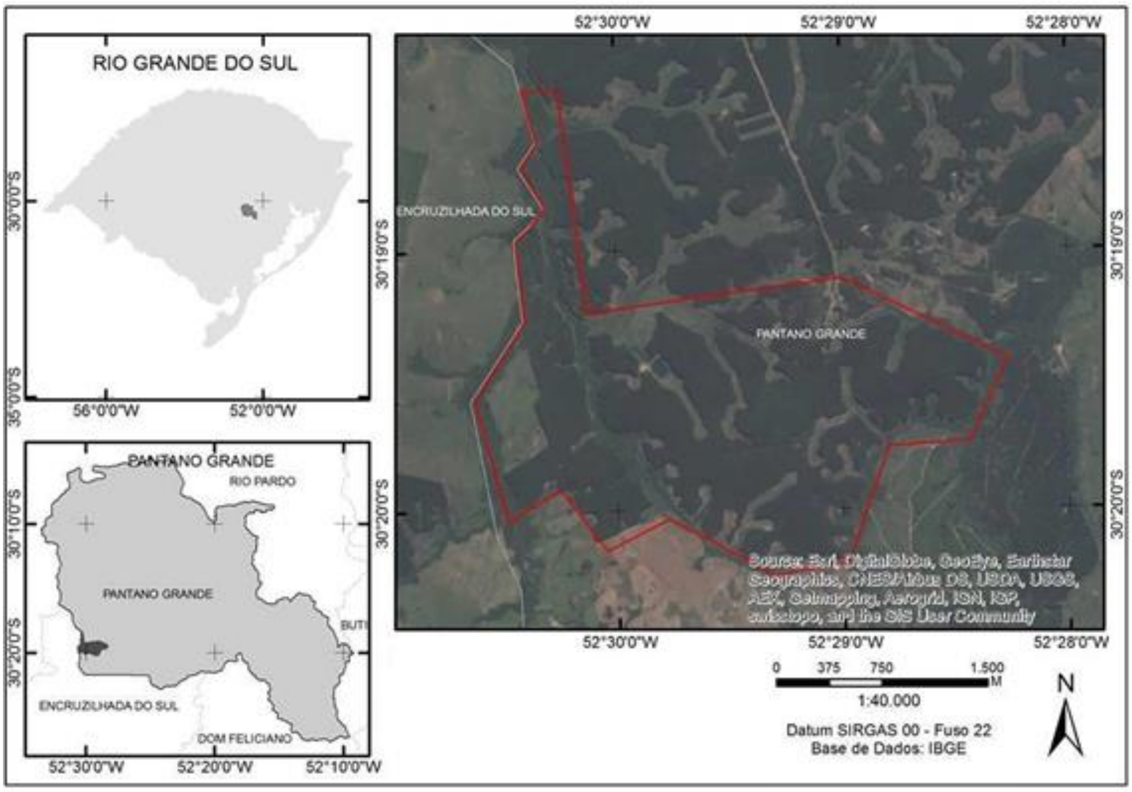

Figure 3.1. Sampled area in Pantano Grande municipality, Rio Grande do Sul state, Brazil.

3.2.2 Plants collect

From the phytosociological studies of vegetal communities, 56 native plant species were selected, 12 per site (Table 3.1). These plant species show the higher indexes of importance value in each sample unit, which were calculated through the sum average of Relative Coverage and Relative Frequency values. These data were provided by the Horticulture and Forest Department of the Agronomy School of the Federal University of Rio Grande do Sul (UFRGS) and by the Botany Laboratory of the University of the Taquari Valley (UNIVATES). The sampling was executed between 2017 and 2018. In addition to the field identification, plants exsicatas for laboratorial level verification were prepared. A total of 324 plant samples, equally distributed into the environments, were collected.

Table 3.1. Plant species sampled into Grassland (G), Grazing Exclusion (GE) and Riparian Forest (RF) environments of Brazilian Pampa.

	Collected environments		
Plant species	G	GE	RF
Adianthopsis clorophyll (Sw.) Fée	■	■	

Agrostis montevidensis Spreng. ex Nees.	■		
Andropogon lateralis Nees	■	■	
Axonopus affinis Chase	■		
Baccharis dracunculifolia DC.	■	■	
Baccharis trimera (Less)	■	■	
Bromelia antiacantha Bertol.			■
Casearia sylvestris Sw.			■
Centella asiatica (L.) Urb.	■	■	
Chaetogastra gracilis (Bonpl.)	■	■	
Chrysophyllum marginatum (Hook. & Arn.)			■
Chromolaena laevigata (Lam.) R.M.King & H.Rob.	■	■	
Citronella gongonha			■
Cyathea atrovirens (Langsd. & Fisch.)			■
Daphnopsis racemosa Griseb.			■
Desmodium incanum DC.	■	■	
Desmodium adscendens (Sw.) DC.	■	■	
Dichondra sericea SW	■	■	
Dichanthelium sabulorum (Lam.) Gould & C.A.Clark	■	■	
Elephantopus mollis Kunth	■	■	
Eryngium horridum Malme	■	■	
Eugenia hiemalis Cambess.			■
Eugenia uruguayensis Cambess.			■
Gymnanthes klotzschiana Mü ll. Arg.			■
Ilex brevicuspis Reissek			■
Ilex dumosa Reissek			■
Hypoxis decumbens L.	■		
Lithraea brasiliensis Marchand			■
Miconia hyemalis A. St.-Hil. & Naud.			■
Myrrhinium atropurpureum Schott			■
Myrcia palustres DC.			■
Myrcianthes pungens (Berg)			■
Myrciaria tenella (DC.)			■
Myrsine gardneriana DC.			■
Ocotea pulchella (Nees)			■
Paspalum umbrosum Trin.	■		
Paspalum conjugatum P.J. Bergius		■	
Piptochaetium montevidense (Spreng.)	■	■	
Paspalum notatum Fluegge	■	■	
Paspalum umbrosun Trin.	■	■	
Paspalum urvillei Steud.		■	
Psidium cattleianum Sabine			■
Psychotria carthagenensis Jacq.			■
Ruellia angustiflora (Ness)			■
Saccharum angustifolium (Nees) Trin.	■		
Scutellaria racemosa Pers.	■		

Sebastiania brasiliensis Spreng.			■
Setaria parviflora (Poir.) Kerguélen	■		
Sida rhombifolia L.	■	■	
Schizachyrium microstachyum (Desv. ex Ham.) Roseng	■		
Syagrus romanzoffiana (Cham.) Glassman			■
Symplocos uniflora (Pohl)			■
Trichilia elegans A. Juss.			■
Vernonanthura nudiflora (Less)	■	■	
Vitex megapotamica (Spreng.)			■

The amount of the plants parts of each sample allowed one-hour of mite seeking. The samples were conditioned in plastic bags in a Styrofoam box at controlled temperature of around 5 ºC and then transported to the laboratory. The lab storage was also at a controlled temperature of 5 ºC. The triage was run within seven days of storage.

3.2.3 Mites mounting and identification

Mites were mounted in microscopic slides with Hoyer medium (Jeppson *et al*. 1975) under 60 ºC for seven days to distention and clarification (Moraes & Flechtmann 2008). The slides were labelled and conditioned into a collection room at < 40% of relative humidity. The mites were identified with dichotomic keys (Krantz & Walter 2009; Ferla *et al*. 2011; Johann *et al*. 2013; McMurtry *et al*. 2013; Fan *et al*. 2016) through a phase contrast optical microscope (Zeiss Axio Scope A1).

3.2.4 Microclimatic factors

Temperature, air humidity, soil moisture, wind speed and photosynthetically active radiation data were collected in each environment. These data were registered throughout 2018 and collected every two hours for eight consecutive hours, starting at 10 a.m. and ending at 6 p.m. Wind speed, temperature and air humidity were measured by a Portable Digital Thermo-Hygrocanemometer Luxmeter (Instrutherm Thal 300, model 0211). Photosynthetically active radiation (PAR) data was simultaneously acquired through a LI-COR light sensor (LI-190 Line Quantum Sensor) coupled to a LI-COR DataLogger (LI-1400). Soil moisture was determined by a moisture meter (type HH2 - AT Delta-TDevices, version 4.0) which allows direct measurement of the data.

3.2.5 Data analysis

The sampling ordination and its respective mite abundances from the three different environments were performed through a non-metric multidimensional scaling analysis (NMDS). The stress values of NMDS graphs indicate the best fit of the multidimensionality on the 2-dimensional plots. Outliers were removed to improve data graphing and square root was applied to flatten the data into a Bray-Curtis similarity matrix. A similarity analysis (ANOSIM) was run to detect significant changes in mite communities among the environments and a statistical test with 999 randomized permutations on p/a data was performed to calculate the statistical global R, with their corresponding probability values. The similarity percentual was obtained by Similarity percentage breakdown (SIMPER) analysis. The analysis was performed in the software PRIMER-E (Clarcke & Gorley 2002) 5.2.9 version. Plants were considered the sampling units.

A Principal Component Analysis (PCA) was performed to investigate the correlation among abiotic parameters at each environment. It were run 108 measurements to select the three best correlations of the analyzed environment. PCAs were performed through the PAST software (Hammer *et al*. 2001), 3.14 version.

The interaction network analysis was performed by the Bipartite package method (Dormann *et al*. 2008, R News) in the R program (R Development Core Team 2010). Some parameters included in the networklevel function were estimated and a bipartite network graph of mite-plant interactions was generated. In addition, two indexes (robustness and connectance) were chosen. The overall topology of this network was used to determine its alignment to the NODF (Nestedness based on Overlap and Decreasing Fill).

3.3 RESULTS

3.3.1 Acarofauna's composition

It was collected 1741 mites with 117 species within 22 families (Table 3.2). The phytophagous species is dominant (57%), with higher abundance of Tenuipalpidae (323) followed by Tetranychidae (293). The more abundant species of Tetranychidae are *Tetranychus urticae* Koch and *Tetranychus armiphenis* Flechtmann & Baker that were found in Grasslands and Grazing Exclusion but were not found in Riparian Forest. The most abundant predaceous is Phytoseiidae with 16.6%, mostly in Riparian Forest. *Amblyseius tamatavensis* Bloomers and *Amblyseius herbicolus* (Chant) are the most abundant only in the Riparian Forest. The abundance in Grassland was 582 individuals distributed into 48 species, 513 in Grazing Exclusions with 50 species and 646 in Riparian Forest with 70 species.

Table 3.2. Mite abundances in Grassland (G), Grazing Exclusion (GE) and Riparian Forest (RF) environments of the brazilian Pampa Rio Grande do Sul, Brazil.

	environments				
Family/Species	G	GE	RF	Total	%
Phytophagous groups	**458**	**329**	**217**	**1004**	**57.67**
Eriophyidae	**81**	**38**	**88**	**207**	11.89
Aculus **sp. nov.1**	6			6	
Aculus **sp. nov.2**		4		4	
Aculus **sp. nov.3**		12		12	
Aculus **sp. nov.4**	13			13	
Aculus **sp. nov.5**			12	12	
Aculus **sp.6**		3		3	
Aculus panpae Toldi, de Lillo & Ferla	9			9	
Aculops **sp.**		2		2	
Leipotrix **sp.1**		4		4	
Vittacus **sp.1**		4		4	
Tegunotus **sp. nov. 1**	4	8		12	
Metaculus **sp. nov. 1**			6	6	
Disella ilicicola Navia e Flescthmann			39	39	
Rhynacus synflori Toldi, de Lillo & Ferla			7	7	
Porcinocus lanceolatus Toldi, de Lillo & Ferla			8	8	
Criotacus sidae Keifer	49			49	
Dichopelmus sigfeminae Toldi, de Lillo & Ferla			9	9	
Pseudocolopodacus **sp. nov. 1**			7	7	
Shevtchenkella **sp.1**		1		1	
Tetranychidae	**161**	**127**	**5**	**293**	16.83
Tetranychus **sp.1**		2		2	
Tetranychus **sp.2**		8		8	
Tetranychus urticae (Koch)	28	46		74	
Tetranychus ludeni Zacher	17	13		30	
Tetranychus mexicanus (McGregor)			1	1	
T. armiphenis Flechtmann & Baker	46	8		54	
Oligonychus **sp.1**	1			1	
Oligonychus **sp.2**	5	3		8	
Neotetranychus **sp.1**			1	1	
Eotetranychus **sp.1**	4	7		11	
Eotetranychus **sp.2**	4			4	
Eotetranychus **sp.3**		2		2	
Mononychellus planki (McGregor)	10	3		13	
Schizotetranychus **sp.1**	1			1	
Schizotetranychus oryzae Rossi de Simons	9	23		32	
Atrichoproctus uncinatus Flechtmann	36	12	3	51	
Tarsonemidae	**27**	**45**	**109**	**181**	10.40
Xenotarsonemus **sp. nov.1**	14	27	21	62	
Xenotarsonemus **sp. nov.2**	3	5		8	
Xenotarsonemus **sp. nov.3**	5	1	9	15	
Tarsonemus **sp.1**	2	8	6	16	
Tarsonemus **sp.2**	2		5	7	
Fungitarsonemus **sp.1**		1	36	37	
Fungitarsonemus **sp.2**		1	8	9	
Excelsotarsonemus **sp. nov.**			23	23	

Neotarsonemoides **sp.**		1		1	
Polyphagotarsonemus latus (Banks)			1	1	
Daleonia **sp.**		1		1	
Phytonemus **sp.**	1			1	
Tenuipalpidae	**189**	**119**	**15**	**323**	18.55
Tenuipalpus apichai			7	7	
Tenuipalpus **sp.**			2	2	
Brevipalpus **sp. nov.1**	17		4	21	
Brevipalpus **sp. nov.2**	150	94	2	246	
Brevipalpus **sp.3**	22	25		47	
Predatory groups	**28**	**83**	**284**	**395**	**22.69**
Ascidae			**11**	**11**	0.63
Asca **sp.1**			8	8	
Asca **sp.2**			2	2	
Asca **sp.3**			1	1	
Blattisociidae			**1**	**1**	0.06
Blattisocius **sp.**			1	1	
Cunaxidae		**1**	**13**	**14**	0.80
Cunaxa ***sp.*** von Heyden			2	2	
Cunaxatricha tarsospinosa Castro & Den Heyer			5	5	
Cunaxoides lajeadensis Wurlitzer & Monjarás-Barrera			6	6	
Cunaxoides **sp.1**		1		1	
Iolinidae	**1**	**18**	**22**	**41**	2.35
Pronematus **sp.**			2	2	
Pseudopronematulus **sp.**		13	10	23	
Metapronematus **sp.**	1	5	6	12	
Parapronematus **sp.**			4	4	
Phytoseiidae	**23**	**55**	**211**	**289**	16.60
Amblyseius **sp.1**			1	1	
Amblyseius **sp.2**			3	3	
Amblyseius **sp.3**		1		1	
Amblyseius herbicolus (Chant)			44	44	
Iphiseiodes moraesi Ferla & Silva			18	18	
Neoseiulus tunus (De Leon)		33	31	64	
Typhlodromips japi Lofego, Demite & Feres			9	9	
Typhlodromips manglae De Leon			6	6	
Amblydromalus limonicus Garman & McGregor			30	30	
Amblyseius tamatavensis Bloomers			54	54	
Euseius inouei (Ehara & Moraes)	2			2	
Euseius mesembrinus (Dean)	1			1	
Typhlodromalus aripo De Leon	4	10	1	15	
Phytoseiulus macropilis (Banks)	3	6		9	
Arrenoseius gaucho Ferla, Silva & Moraes	9	4		13	
Phytoseius guianensis De Leon			6	6	
Graminaseius **sp**		1		1	
Amblyseius euterpes Gondim & Moraes			8	8	
Arrenoseius **sp.**	1			1	
Chelaseius **sp.**	1			1	
Neoseiulus californicus (McGregor)	1			1	
Neoseiulus anonymus (Chant & Baker)	1			1	
Stigmaeidae	**4**	**9**	**26**	**39**	2.24
Agistemus **sp.nov.1**		1	3	4	
Agistemus floridanus Gonzales		5	21	26	
Agistemus brasiliensis Matioli		2	1	3	
Eustigmaeus **sp.1**			1	1	
Zetzellia malvinae Matioli, Ueckermann & Oliveira	4	1		5	
Other feeding habits	**96**	**101**	**145**	**342**	**19.64**
Alycidae	**1**			**1**	0.06
Alycius **sp.**	1			1	
Bdellidae			**3**	**3**	0.17
Bdella ueckermanni Hernandes, Daud & Feres			3	3	
Laelapidae	**1**			**1**	0.06
Cosmolaelaps **sp.**	1			1	

Eupodidae			**3**	**3**	0.17
Eupodes **sp.**			2	2	
Benoinyssus **sp.**			1	1	
Tydeidae	**79**	**63**	**102**	**244**	14.01
Afrotydeus kenyensis (Baker)			6	6	
Tydeus californicus (Banks)	75	56	18	149	
Tydeus costensis Baker	2			2	
Brachytydeus formosa (Cooreman)	1	1	43	45	
Brachytydeus podocarpa (Baker)		5	5	10	
Brachytydeus amazonensis (Mondim)	1	1	21	23	
Pretydeus curiosa (Ueckermann & Meyer)			5	5	
Pretydeus henriandrei Kaźmierski			2	2	
Metalorrya delicata (Kuznetzov)			2	2	
Triophtydeidae		**1**	**4**	**5**	0.29
Triophtydeus **sp.**		1	4	5	
Acaridae			**6**	**6**	0.34
Acarus **sp.1**			5	5	
Tyrophagus **sp.**			1	1	
Suidasiidae			**11**	**11**	0,63
Suidasia **sp.**			11	11	
Winterschimidtidae	**3**	**3**	**13**	**19**	1.09
Czenspinskia **sp.**	3	3	4	10	
Acalvolia squamata (Oudemans)			6	6	
Sphexicozela **sp.**			3	3	
Cheyletidae			**3**	**3**	0.17
Cheletomimus **sp.**			2	2	
Cheletomimus duosetosus Muma			1	1	
Pyemotidae	**1**			**1**	0.06
Pyemotes **sp.**	1			1	
Histiostomatidae	**11**	**34**		**45**	2.58
Histiostoma **sp.**	10	30		40	
Glyphanoetus **sp.**	1	4		5	
Abundance	**582**	**513**	**646**	**1741**	**100**
Richness	**47**	**50**	**70**	**117**	

3.3.2 Comparative of different environments

Regarding the abiotic variables in the PCA analysis, the three environments present contrasting information according to the different analytic factors (Figure 3.2). The air humidity and soil moisture correlate to the Riparian Forest indicators while higher active photosynthetic effects, temperature and wind speed, correlate to the Grassland and Grazingl Exclusion environments. The component 1 (43.78%) shows the best correlation to the photosynthetic radiation activity (0.56) and secondly to the wind speed (0.50). The component 2 (25.02%) presents the best correlation to the temperature (-0.63) and then to the soil moisture (0.57).

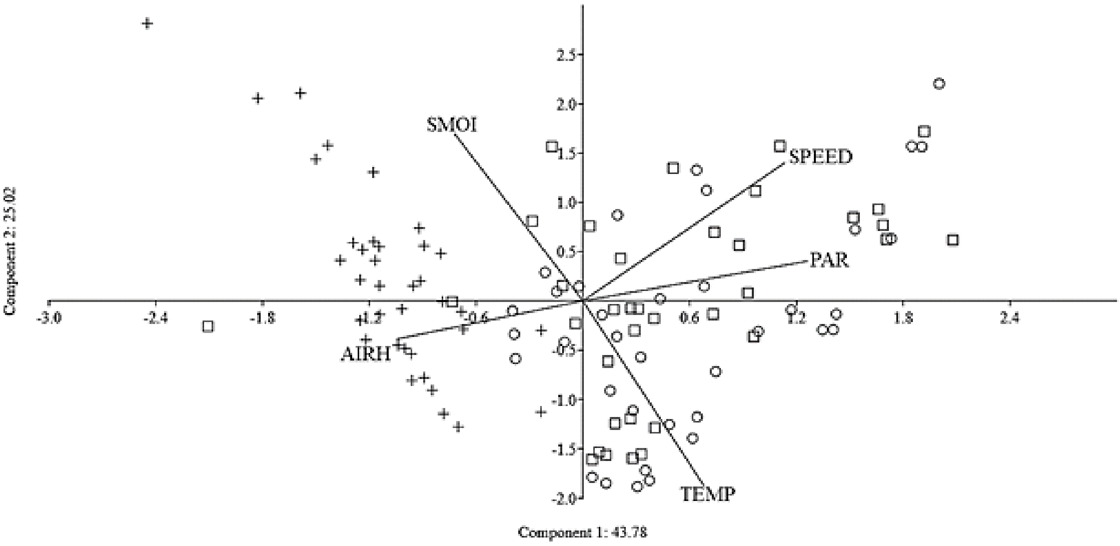

Figure 3.2. Principal component analysis (PCA). TEMP: Temperature (°C); SPEED: Wind Speed (km/h); SMOI: Soil moisture (%); AIRH: Air humidity (%); PAR: photosynthetic activity radiation (µmol s-1 m-2). ○: Grassland, □: Grazing Exclusions: + Riparian Forest.

Samples of the three environments were ordered by NMDS to evaluate the species composition (Figure 3.3). The disturbance, stress values = 0.13, indicates a proper graphic ordination. ANOSIM analysis (R global = 0.286 p = 0.001) attests that the environments form distinct groupings. Riparian Forest differs from Grazing Exclusions (R = 0.392 $p < 0.01$) and from Grassland (R = 0.494 $p < 0.01$). However, there is no distinction between Grazing Exclusions and Grassland (R = 0.012 $p > 0.05$). Riparian Forest formsan independent group.

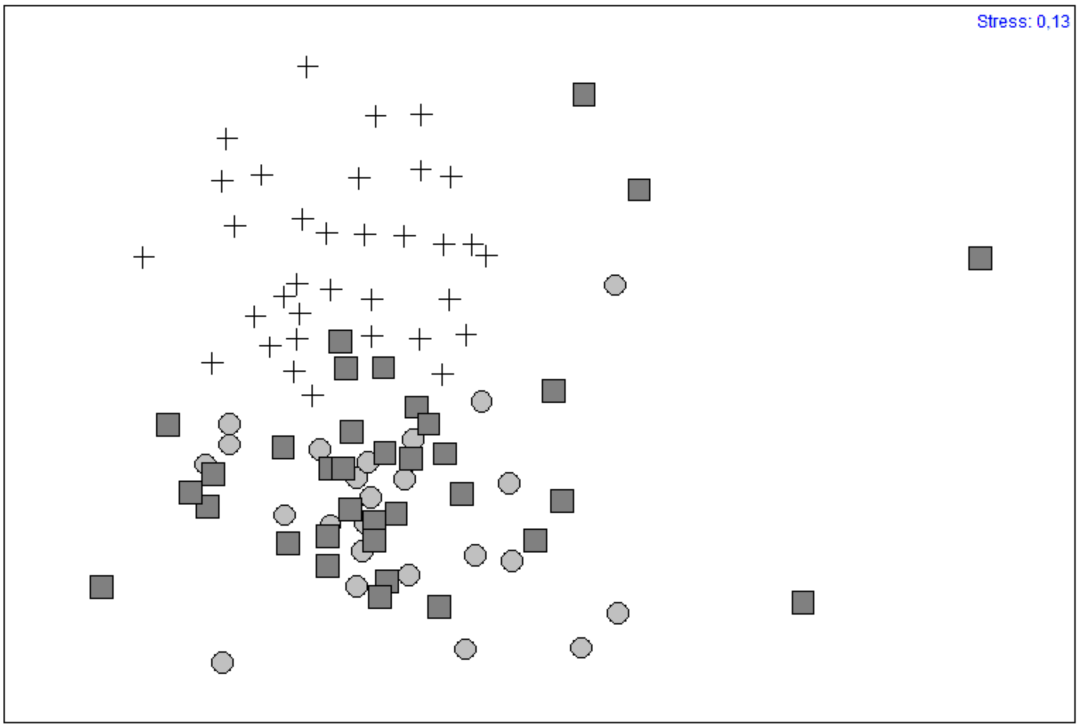

Figura 3.3: Abundance ordination of the mite species in the three environments (NMDS). ○: Grassland; □: Grazing Exclusions; +: Riparian Forest. The low stress value (= 0.13) indicates a positive fit of the two-dimensional representation of the distances among the samples.

Through the Simper analysis, the samples similarity of Grassland is of 15.24%, the Grazing Exclusion with of 13.0% and 14.02% in the Riparian Forest. The main accountable species for the sample similarity of Grassland are *Brevipalpus* sp.2 (66% of contribution), *Tydeus californicus* (Banks) (11.0%) and *Atrichoproctus uncinatus* Flechtmann (4.98%). In the Grazing Exclusion environment, the accountable species for the similarity are *Brevipalpus* sp.2 (45.69%), *T. californicus* (21.3%) and *Xenotarsonemus* sp.1 (13.12%). The similarity contributions in the Riparian Forest are the *Brachytydeus formosa* (Cooreman) (17.4%), *Fungitarsonemus* sp.1 (17.4%) and *A. tamatavensis* (11.37%).

3.3.3. Mite/plant interactions

Considering the network interactions between mite and plant in the Grasslands, the *Baccharis dracunculifolia* DC stands out presenting 17 interactions (Robustness = 0.796; Connectance = 0.127) (Figure 3.4). *Brevipalpus* sp.2 show 11 interactions with different plants.

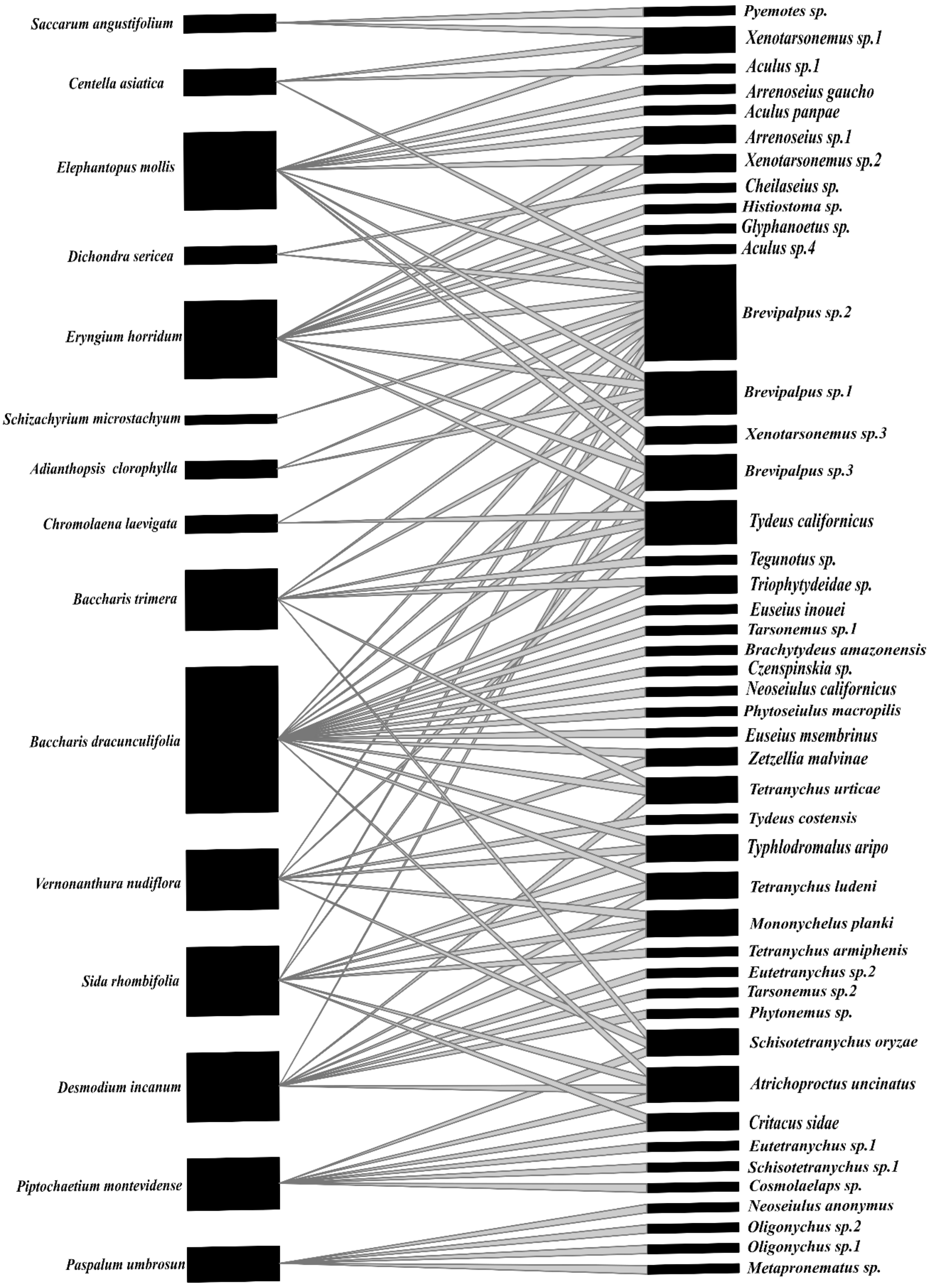

Figure 3.4: Interaction bipartite network of the Grassland site. The left side bars represent host plant species and the right-side bars represent mite species.

In the mite/plant network of the Grazing Exclusion site, *Elephantophus mollis* Kunth presents 16 interaction; *B. dracunculifolia* with 12 interactions (Robustness = 0.784; Connectance = 0.113) (Figure 3.5). and 11 interactions of *T. californicus* and *Brevipalpus* sp.2.

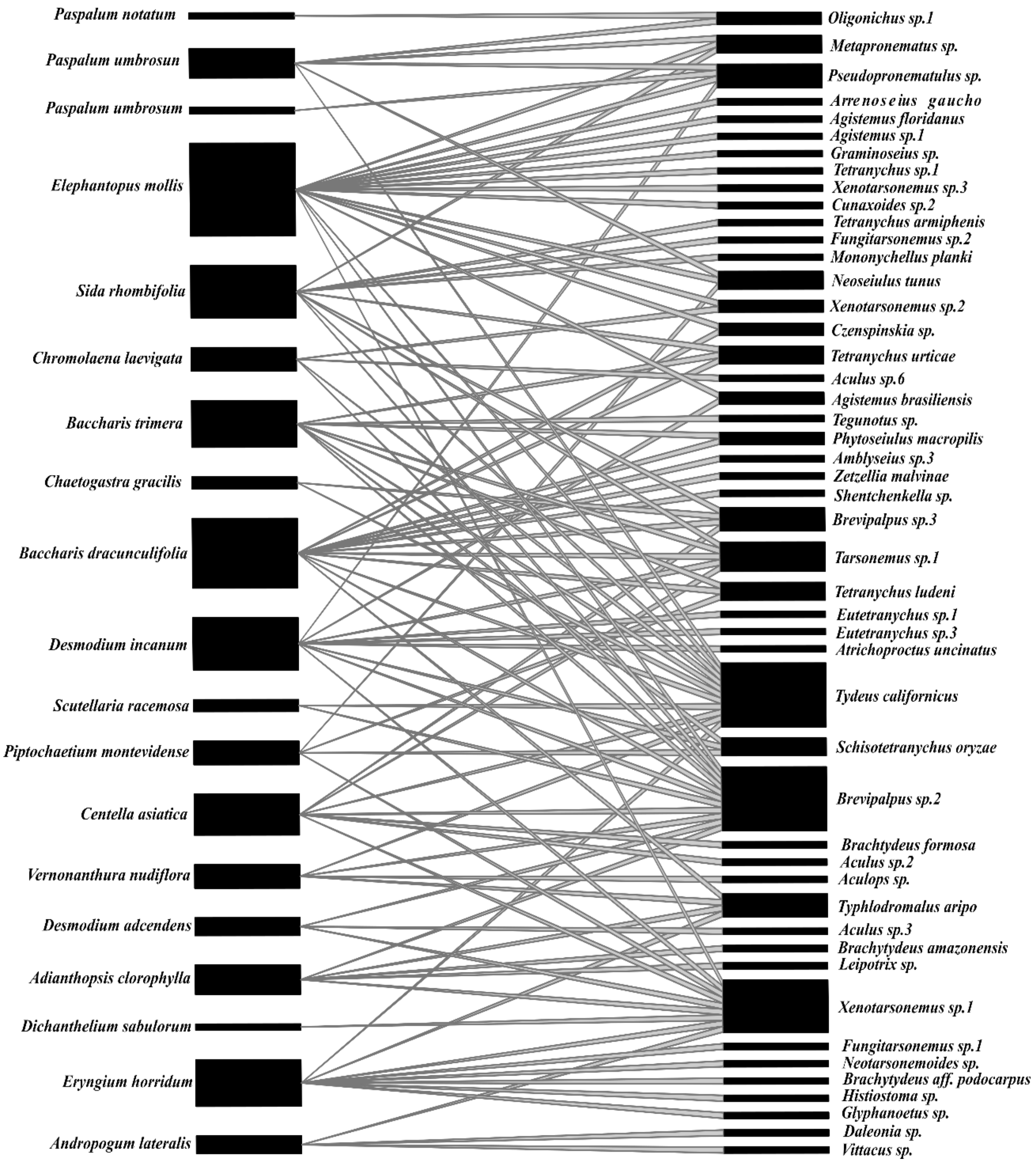

Figure 3.5: Interaction bipartite network of Grazing Exclusions site. The left side bars represent host plant species and the right-side bars represent mite species.

The mite/plant network in the Riparian Forest site indicate more interactions of *Ruellia angustiflora* (Ness) and *Myrcianthes pungens* (Berg), 17 and 16 respectively (Robustness = 0.856; Connectance = 0.116). *A. tamatavensis* presents13 interactions (13) (Figure 3.6).

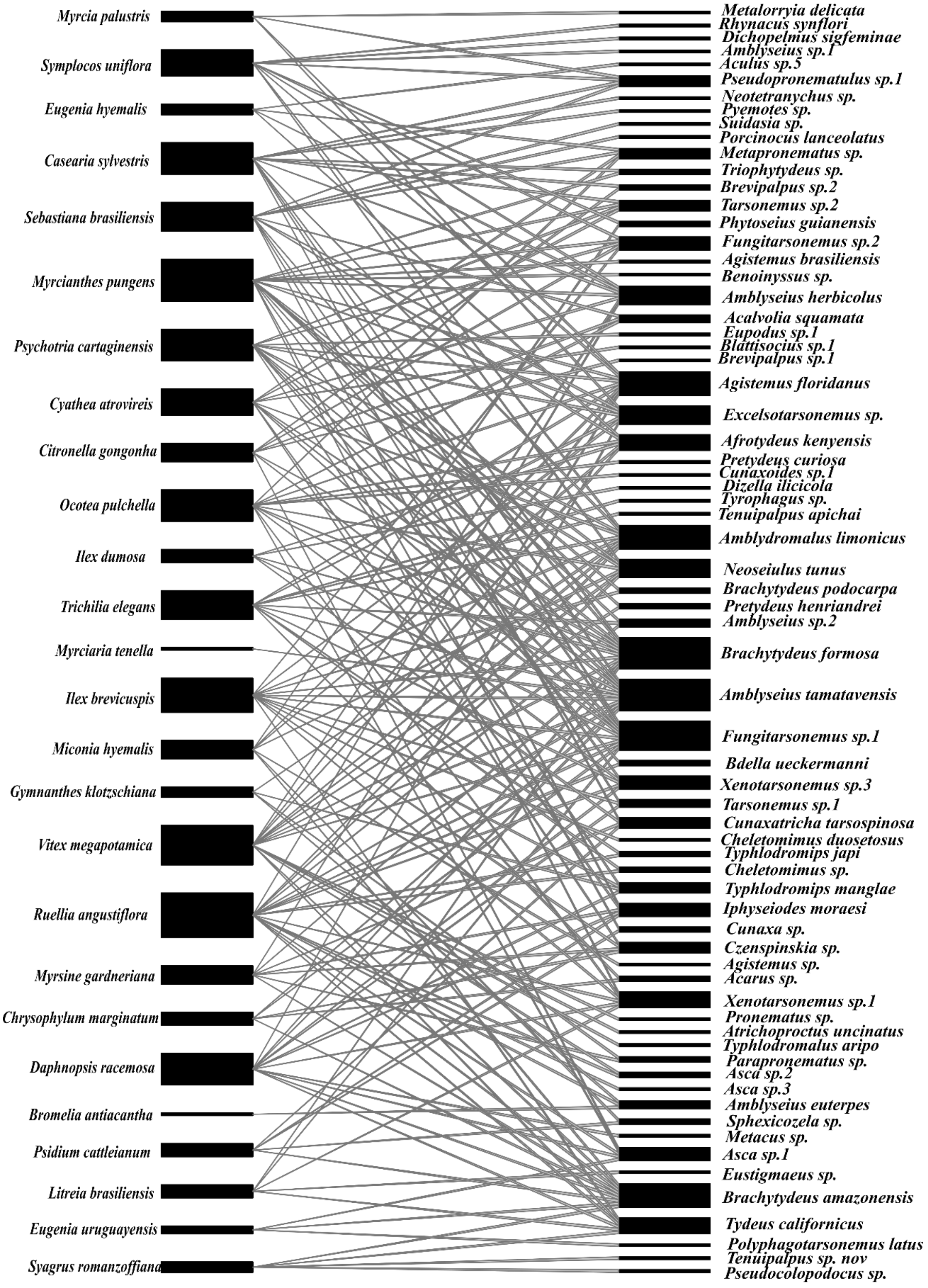

Figure 3.6: Bipartite network of Riparian Forest site. The left side bars represent host plant species and the right-side bars represent mite species.

3.4 DISCUSSION

The composition of the acarofauna of Riparian Forest contrasts with Grassland and Grazing Exclusions for presenting higher humidity and lower luminosity and wind. Thus, it may be suggested that the acarofauna is sensitive to these microclimatic factors. These results corroborate with other studies that used arthropods to compare different environments. Demite *et al*. (2015) found higher amount of mites in adjacent areas to sugarcane and pasture crops compared to neighboring areas of orange culture. Rocha *et al*. (2015) found higher richness of predatory mites in preserved areas compared to anthropized areas. Thus, the acarofauna in plants can also be considered a meaningful indicator of environmental characteristics in the brazilian Pampa.

Regarding the community composition, it is possible to notice that some species are unique of the Riparian Forest, Grassland and Grazing Exclusions. *Tetranychus urticae*, an economically important pest (Moraes & Flechtmann 2008), is unique and abundant in Grassland and Grazing Exclusions. Native plants that form mite reservoirs are *B. dracunculifolia*, *Baccharis trimera* (Less), *Desmodium incanum* DC. and *Sida rhombifolia* L. These may also be natural reservoirs of predators. Two predator species commercialized in Brazil for the biological control of this phytophagous are *Neoseiulus californicus* (McGregor) and *Phytoseiulus macropilis* (Banks) (MAPA 2019). These were also found in *B. dracunculifolia* and *B. trimera*.

The predaceous *A. tamatavensis* and *A. herbicolus* were abundant in the Riparian Forest and may associate to the preys of this site *Disella ilicola* Navia and *Fungitarsonemus* sp.1 or relate to the higher air humidity. Phytoseiidae depends on relative air humidity for oviposition and are abundant in the Riparian Forest (Demite & Feres 2005; Devis *et al*. 2006). The relation of this microclimatic factor with the acarofauna is poorly known (Tixer 2018) and further studies that may confirm the suggested relation of this work are needed.

The comparative of the different environments demonstrates that the Grassland and Grazing Exclusion are similar, one of the most relevant occurrence is due to the presence 244 specimens of *Brevipalpus* sp.2 and both contrast with the Riparian Forest, where only two occurrences are identified. Besides the association to lower air humidity, soil moisture, higher temperatures and luminosity, this phytophagus suprasses the defenses of a large plant diversity (Peterson *et al*. 2016). These results corroborate the assumption of a polyphagous habit of the genus *Brevipalpus* individuals (Moraes & Flechtmann 2008; Krantz & Walter 2009). The diversified feeding like pollen, phytophagous or even mycophagous characteritics of the *Tydeus californicus*, justify its abundance in several samples of Grassland and Grazing Exclusions (da Silva *et al*. 2014).

Regarding the Riparian Forest similarity, the *B. formosa* plays a major role due to its abundance in several plants. The capacity of association to various several host plants possibly relates to the characteristic varied diet of the Tydeidae (Krantz & Walter 2009).

The data point out to the plant species that can be sampled in further studies on mites interactions. The plants with higher interactions in Grassland, Grazing Exclusions and Riparian Forest are *B. dracunculifolia*, *E. molis* and *R. angustiflora*, respectively. These plants spreadout through several South American countries (Freire *et al*. 2007; Borges & Forzza 2008; Cruz *et al*. 2016) and present trichomes, that associate to higher mite populations (Tixer 2018). Thus, the mite interaction networks observed in plants of the Brazilian Pampa are similar to other sites in South American countries (Calatayud *et al*. 2016). The robustness values are similar to the Walter & Araujo (2019) work, however, the higher connectivity value suggests a lower specialization of these networks, as expected in species of contrasting feeding habits.

The six year enclosure recovery action in Grazing Exclusion do not promote changes in the acarine community, suggesting similarity between the Grazing Exclusion and the Grassland. The practice of enclosure as a recovery action in the Pampa should be reevaluated, reagrding the subsistence of native fields. The four acarine species described by Toldi *et al*. (2019) and the identification of thirteen unprecedent species in the brazilian Pampa highlight the need of further acarofauna surveys. The consolidation of the technique of identification of mites in plants as an index to environmental characteristics depends on new studies focused on attesting the efficiency of the sampling methodologies and on the recognition of mites species common to different environments. According to Ruf & Beck (2005), soil mites can be indicators of environmental quality, since the soils are rich in species diversity and quantity, that can reach 20,000 individuals/m^2.

In this study, the mites demonstrate to be an efficient indicator of the different characteristics of the evaluated environments. Future work relating mites to microclimatic factors and landscapes may attest the hypothesis that plant mites are effective indicators of environmental characteristics. Plant interactions of the mite species are high in the three environments.

4 ASSEMBLAGES OF PREDATORY MITES IN DIFFERENT ENVIRONMENTS OF THE BRAZILIAN PAMPA

Changes required by the journal to accept in International Journal of acarology. B2 in environmental sciences.

Abstract

The Brazilian Pampa houses a high diversity of fauna and flora, which maintain ecosystem stability. The aim of this study was to evaluate mite community composition in different environments that belong to the Pampa of the South of Brazil. The environments selected were Grassland, Grazing Exclusions and Riparian Forest. Community composition was evaluated using ANOSIM and IndVal analyses. A total of twenty-two Phytoseiidae species and six Stigmaeidae species were recognized in this study. Riparian Forest was the environment with the highest percentage of total predatory mite abundance (72%), followed by Grazing Exclusions (20%) and Grassland (8%). Comparisons between Grassland and Grazing Exclusions did not show any differences (R = 0.092, $p < 0.05$). There was difference between Grassland and Riparian Forest (R = 0.45, $p = 0.001$), and between Grazing Exclusions and Riparian Forest (R = 0.46, $p = 0.001$). The highest number of indicator mite species (10) were recognized in Riparian Forest and had correlation with humidity, *Amblyseius tamatavensis* Blommers (0.78), *Amblyseius herbicolus* (Chant) (0.67) and *Agistemus floridanus* González-Rodríguez (0.62) having the highest IndVal values. *Arrenoseius gaucho* Ferla, Silva & Moraes (0.31) and *Zetzellia malvinae* Matioli, Ueckermann & Oliveira (0.27) were defined as indicators of Grassland, while *Typholodromalus aripo* DeLeon (0.44) was the only species associated with Grazing Exclusions. Grassland and Grazing Exclusions were the hottest, sunniest, most wind-exposed environments. It was evident that microclimatic variations negatively changed community composition of predatory mites in grasslands compared to forests.

Keywords: Biological Control; Natural Environment; Phytoseiidae; Stigmaeidae.

4.1 Introduction

In Brazil, the Pampa is limited to the state of Rio Grande do Sul, occupying approximately 63% of the state's area, and currently less than 40% of its original native vegetation remains (MMA, 2019). Vegetation typical of this phytophysiognomy is comprised of a mosaic of grasslands with dominance of grasslands and riparian forests (Carvalho *et al*. 2009; Behling *et al*. 2005; Roesch *et al*. 2009).

The high diversity of fauna and flora in the Pampa provides environmental services, conserves water resources, pollinators, genetic resources, and foraging potential for the livestock chain (Pillar *et al*. 2009). Some human activities favor the maintenance of the grassland environment, e.g. grazing activities and fire that prevent the expansion of forest formations over grasslands (Behling *et al*. 2007; Carvalho *et al*. 2009). Grasslands have been recently replaced by grain crops and *Eucalyptus saligna* Sm. (Myrtaceae) plantations to obtain cellulose (Pillar *et al*. 2002; Santana *et al*. 2008; Overbeck *et al*. 2007; Pillar *et al*. 2009). These monocultures affect both the diversity and distribution of organisms in this environment (Rieff *et al*. 2010; Rieff *et al*. 2016).

Several factors might affect environmental recovery and analyses are required to compare different mature forest areas and assess changes in species composition and regeneration of degraded areas (Chazdon 2003; Coote *et al*. 2013; Derroire *et al*. 2016). Since it is not possible to analyze the environment completely in megadiverse regions such as the Pampa, the use of ecological indicators is an alternative for the diagnosis of environmental quality (Kimberling *et al*. 2001; Coote *et al*. 2013).

Studies have been conducted to recommend indicators of environmental and ecological changes at national and international scales, such as the European System of Indicators, used for the Sustainable Management of Forests (CMPFE) (Coote *et al*. 2013). Plant (Silva *et al*. 2018), spider, and bird species (Coote *et al*. 2013) are some of the most frequently used indicators. Studies indicate that soil mites might be environmental indicators of both native areas and areas of exotic monocultures (Rieff *et al*. 2016). These ecological indicators are used in decision-making processes regarding management and public policies for environmental recovery and conservation (Niemi and McDonald 2004).

Plant-dwelling mites represent one of the most diverse and abundant groups in terrestrial ecosystems (Walter and Proctor 2013). In spite of that, studies that analyze their potential as indicators of both anthropized environments and preserved environments are scarce (Rocha *et al*. 2015). Several studies provide information on the occurrence of predatory mites on plants. However, little is known about the parameters that explain this occurrence (Tixier 2018).

Reports of mite species of predatory families are commonly associated to plants in the south of Brazil, especially Phytoseiidae and Stigmaeidae (Ferla & Moraes 2002; Ferla *et al*. 2007; Reichert *et al*. 2014; Rocha *et al*. 2015), which are the most frequent ones. These two families have different feeding habits (Moraes & Flechtmann 2008; Krantz & Walter 2009), predominantly predatory mites, which are abundant and have significant richness in the vegetation of the South of Brazil (Johann *et al*. 2013).

Phytoseiidae are largely employed in applied biological control (Gerson *et al*. 2003; Tixier 2018), and their feeding preference, biological potential, and habitat are frequently studied. It is currently the most studied family in the Neotropical region (McMurtry *et al*. 2013; Araújo and Daud 2018a; 2018b), with 2,521 species described (Demit *et al* 2014; Demite *et al*. 2019). Stigmaeidae live on plants and soil, and have 577 species described (Fan *et al*. 2016).

In the state of Rio Grande do Sul, stigmateiids have been observed in several agroecosystems, and they are considered the second most abundant predatory family after Phytoseiidae (Ferla & Moraes 2002; Johann *et al*. 2013). Several studies provide information on the occurrence of predatory mites on plants. However, little is known about the parameters that explain this occurrence (Tixier 2018).

This study corroborates the hypothesis that microclimatic variations negatively affect community composition of predatory mites in grasslands compared to forests. Therefore, the aim of this study was to evaluate mite community composition in different environments that belong to the Pampa of the South of Brazil.

4.2 Material and methods

4.2.1 Study area

The sampling units (sites) are located in permanent preservation areas within a eucalyptus forest garden in the municipality of Pântano Grande, state of Rio Grande do Sul, Brazil (30°19'36.59"S and 52°29'24.83"W). These areas belong to the Phytophysiognomy of Shrub Field in the Central Depression of the Pampa. This area is classified as Cfa, i.e., with quite defined summer and winter, and annual rainfall varying from 1,600 to 1,900 mm (Peel *et al*. 2007). Soil is Red dystrophic Argisol (Streck *et al*. 2018). These areas were used for agriculture and cattle breeding until 2006, when eucalyptus plantations started and environmental recovery areas were defined.

Nine sites were selected and divided into three environments with areas larger than 0.5 ha (Fig. 4.1): **Riparian Forest** - with tree vegetation occurring on the margins of existing water resources within the site. Riparian forests are comprised of a phytophysiognomic element called Seasonal Semideciduous Forest, which has high fall of leaves in a certain period of the year and accumulation of litter; **Grazing Exclusions** – with environmental recovery measures, fenced for six years, shrub vegetation, which occurs spontaneously, and shrub species; **Grassland -** with compacted soil and herbaceous vegetation, with no fencing or recovery measures, and access to cattle for grazing.

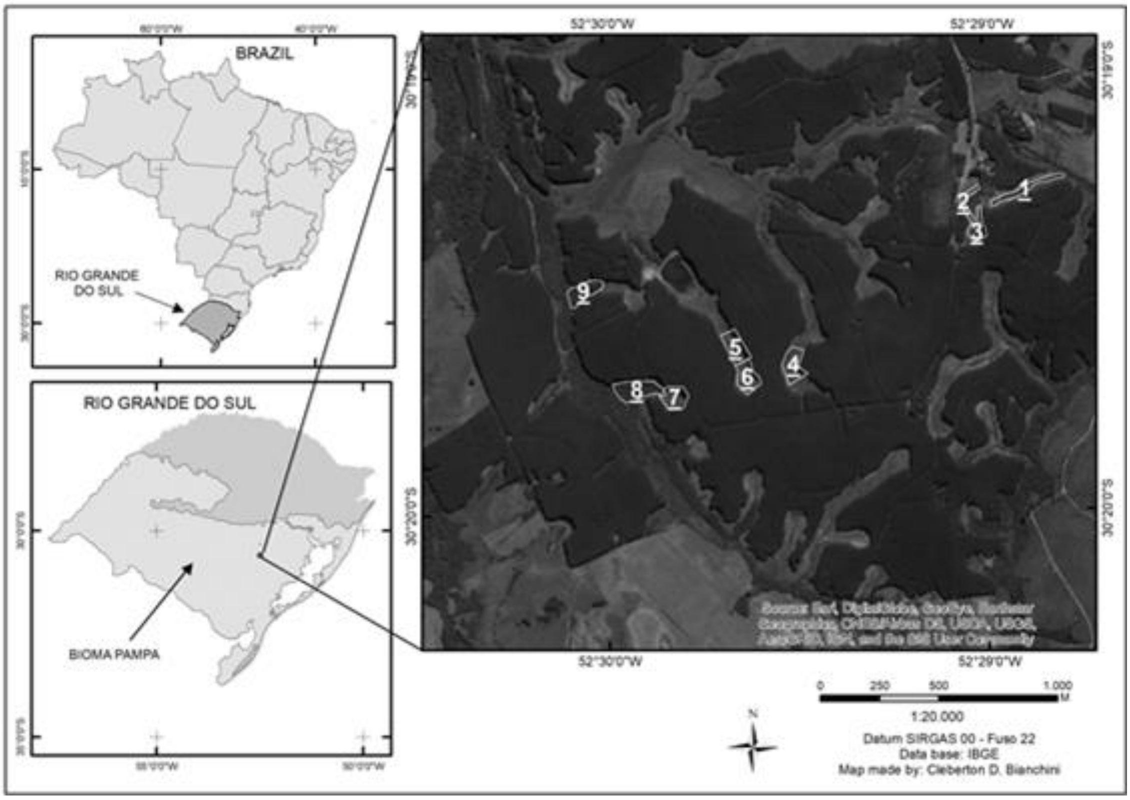

Figure 4.1. Areas assessed in the Pampa, municipality of Pântano Grande, Rio Grande do Sul,

Brazil. Environments: Grassland (2, 4, 5), Grazing exclusions (3, 6, 7) and Riparian Forest (1, 8, 9).

The samplings were conducted between September 2017 and March 2018 starting 20 meters away from the edge of the area, to avoid the edge effect (Rodrigues *et al*. 2010). A total of 324 plant samples were collected. Fifty-five native plant species were selected based on phytosociological studies of plant communities in the same areas. The species selected for the present study were those that had the highest Importance Value Indices at each sampling unit, calculated using the mean of the sum of Relative Cover and Relative Frequency values. These data were provided by the Department of Horticulture and Forestry of the Agronomy School of UFRGS, Porto Alegre, RS and by the Laboratory of Botany of Univates, Lajeado, RS.

The amount of the plants parts of each sample allowed one-hour of mite seeking. The samples were conditioned in plastic bags in a Styrofoam box at controlled temperature of around 5 ºC and then transported to the laboratory. The lab storage was also at a controlled temperature of 5 ºC. The triage was run within seven days of storage. Mite screening sampling effort on each plant species was of one hour, thus standardizing the sampling effort in the three environments. Mites were mounted on microscope slides in Hoyer's medium, kept in a furnace (60°C) for a period of seven days for distension and clarification of the specimens (Moraes & Flechtmann 2008). Slides were tagged and kept in a room with relative humidity lower than 40%. Species were determined with the help of dichotomous keys (Ferla *et al*. 2011, Johann *et al*. 2013; McMurtry *et al*. 2013; Fan *et al*. 2016) and a phase-contrast optical microscope (Zeizz Axio Scope A1). Only females of each species were considered.

Data referring to temperature, relative air humidity, wind speed, photosynthetically active radiation, and soil moist were collected within each site. These data were recorded throughout 2018 in three field trips on cloudless days, and were collected every two hours, during eight consecutive hours, starting at 10 AM and ending at 6 PM. For the analysis of wind, temperature, and air humidity, we used a Digital Portable Thermo-Hygro-Anemometer Luxmeter (Instrutherm Thal 300, model 0211). We measured photosynthetically active radiation (PAR) on the same days and at the same times using a LI-COR luminosity sensor (Line Quantum Sensor LI-190) coupled to a LI-COR DataLogger (LI-1400). Soil moisture was measured using a Moisture Meter (HH2 type – AT Delta-TDevices, version 4.0), which allows for directly measuring the data, following the same methodology of the other microclimatic measurements.

4.2.2 Data analyses

Ordination of samplings with their respective predatory mite abundances from the three different sites was performed using a non-metric multidimensional scaling analysis (NMDS). The stress values of NMDS graphs indicate the fit of multidimensionality on the 2-dimensional plots, using three geographical replicates and three temporal replicates, totaling nine samples at each site. Abundance data were not transformed and were used in a Bray-Curtis similarity matrix. To detect significant changes in mite communities between forest sites, we ran a similarity analysis (ANOSIM), which is similar to a standard univariate ANOVA, and performs a statistical test with 999 randomized permutations on p/a data to calculate the statistical global R, with their corresponding probability *p*-values performed in the software PRIMER-E (Clarke and Gorley 2002) version 5.2.9.

A Principal Component Analysis (PCA) was performed using a software to investigate the correlation between abiotic parameters at each site, which used 36 measurements per site to separated best the three sites analyzed. PCAs were performed using the PAST software (Hammer *et al*. 2001), version 3.14.

IndVal values were calculated for each species and site using the ‘labdsv’ package v1.8-

0 in R (R Development Core Team, 2010). Species were considered indicators when their maximum IndVal value was significant ($P \leq 0.05$).

Aiming at determining the predator mites species as potential ecological indicators of different conditions, we applied the Indicator Value method (IndVal) (Dufrene and Legendre 1997). The IndVal method is based on exclusivity for a habitat and frequency of occurrence within the same habitat. Calculations of Indval were performed for each species and site with the package ‘labdsv’ v1.8-0 in R (R Development Core Team 2010). Statistical significance of indicator values was measured using random iterative methods by relocating 10,000 samples within sample groups. Species were considered as indicators if their maximum value of indval was significant ($p \leq 0.05$).

An interaction network analysis was performed using the method Bipartite package (Dormann *et al*. 2008, R News) in the R program (R Development Core Team 2010). Some parameters included in the networklevel function were estimated and a bipartite network graph of mite-plant interactions was generated. In addition, two indices (robustness and connectance) were chosen. The overall topology of this network was used to determine its alignment with NODF (Nestedness based on Overlap and Decreasing Fill).

4.3 Results

Of the 55 plants analyzed in the three environments, 60% had predatory mites. Most of the predatory mite species were phytoseiids (22), followed by stigmateiids (6). Riparian Forest was the environment with the highest percentage of total predatory mite abundance (72%), followed by Grazing Exclusions (20%) and Grassland (8%) (Tab. 4.1). Approximately 70% of the abundance found in the environments were represented by only five species: *Neoseiulus tunus* (De Leon) (64), *Amblyseius tamatavensis* Blommers (55), *Amblyseius herbicolus* (Chant) *(*48), *Amblydromalus limonicus* (Garman & McGregor) (30) e *Agistemus floridanus* Gonzalez-Rodriguez (25).

Baccharis dracunculifolia, DC., which has trichomes, had the highest predator richness (7; *Amblyseius* sp.3, *Euseius inouei* Ehara & Moraes *Euseius mesenbrinus* (Dean), *Neoseiulus californicus* (McGregor), *Phytoseiulus macropilis* (Banks), *Typhlodromalus aripo* De Leon and *Zetzellia malvinae* Matioli, Ueckermann & Oliveira). Other plants with higher richness were: *Elephantopus mollis* Kunth, *Symplocos uniflora* (Pohl), *Ruellia angustiflora* (Ness) which have trichomes; *Trichilia elegans*. A. Juss. and *Vitex megapotamica* (Spreng.), which are glabrous and have domatia formed by hairs of trichomes; and *Psychotria carthagenensis* Jacq. e *Myrcianthes pungens* (Berg), which are totally glabrous (Tab. 4.1).

Table 4.1 Environments assessed in the municipality of Pântano Grande, State of Rio Grande do Sul, Brazil.

Plant species	**Environments**	Predatory mite species	**N**
Adianthopsis clorophyll (Sw.) Fée	GE	*Typholodromalus aripo* DeLeon	2
Baccharis dracunculifolia DC.	G e GE	*Amblyseius* sp.3	1
		Neoseiulus californicus (McGregor)	1
		Euseius inouei Ehara & Moraes	2
		Euseius mesenbrinus (Dean)	1
		Phytoseiulus macropilis (Banks)	4
		Typhlodromalus aripo	7
		Zetzellia malvinae Matioli, Ueckermann & Oliveira	4
Baccharis trimera (Less)	G	*Phytoseiulus macropilis*	5
Bromelia antiacantha Bertol.	RF	*Amblyseius* sp.4	1
Casearia sylvestris Sw.	RF	*Amblyseius herbicolus* (Chant)	5

		Amblydromalus limonicus (Garman & McGregor)	6
		Amblyseius tamatavensis Blommers	1
Chrysophyllum marginatum (Hook. & Arn.)		*Amblyseius tamatavensis*	1
Cyathea atrovirens (Langsd. & Fisch.)	RF	*Amblyseius herbicolus*	25
		Amblyseius tamatavensis	1
		Iphiseiodes moraesi Ferla & Silva	3
Daphnopsis racemosa Griseb.	RF	*Typhlodromips mangleae* De Leon	1
		Iphiseiodes moraesi	1
		Amblyseius tamatavensis	2
		Amblydromalus limonicus	1
Desmodium incanum DC.	GE	*Agistemus brasiliensis* Matioli, Ueckerma	1
Dichondra sericea SW	G	*Chelaseius* sp.	1
Elephantopus mollis Kunth	G e GE	*Neoseiulus tunus* (DeLeon)	27
		Agistemus sp.	1
		Arrenoseius gaucho Ferla, Silva & Moraes	13
		Graminaseius sp.1	1
		Agistemus floridanus Gonzalez-Rodriguez	5
		Agistemus brasiliensis	1
Eryngium horridum Malme	GE	*Arrenoseius* sp.	1
		Typhlodromalus aripo	1
Eugenia hiemalis Cambess.	RF	*Amblyseius herbicolus*	2
Gymnanthes klotzschiana Mü ll. Arg.	RF	*Typhlodromips mangleae*	2
		Amblydromalus limonicus	4
		Amblyseius sp.4	1
Ilex brevicuspis Reissek		*Amblydromalus limonicus*	1
		Typhlodromips mangleae	2
		Amblyseius tamatavensis	1
Lithraea brasiliensis Marchand		*Eustigmaeus sp.*	1
Miconia hyemalis A. St.-Hil. & Naud.	RF	*Neoseiulus tunus*	1

		Agistemus floridanus	2
		Agistemus sp.	3
		Phytoseius guianensis De Leon	2
Myrcia palustres DC.	RF	*Agistemus floridanus*	5
Myrcianthes pungens (Berg)	RF	*Amblyseius herbicolus*	3
		Agistemus brasiliensis	1
		Agistemus floridanus	2
		Neoseiulus tunus	1
		Amblyseius tamatavensis	14
		Amblyseius sp.2	1
Myrciaria tenella (DC.)	RF	*Amblyseius tamatavensis*	1
Myrsine gardneriana DC.	RF	*Iphiseiodes moraesi*	5
		Agistemus floridanus	3
Ocotea pulchella (Nees)	RF	*Neoseiulus tunus*	6
		Agistemus floridanus	1
		Amblyseius tamatavensis	2
Paspalum umbrosum Trin.	G and GE	*Neoseiulus tunus*	1
		Neoseiulus anonymus (Chant & Baker)	1
Piptochaetium montevidense (Spreng.)	GE	*Neoseiulus tunus*	5
Psidium cattleianum Sabine	G and GE	*Iphiseiodes moraesi*	6
Psychotria carthagenensis Jacq.	RF	*Neoseiulus tunus*	2
		Agistemus floridanus	1
		Typhlodromips mangleae	1
		Amblydromalus limonicus	1
		Amblyseius tamatavensis	15
Ruellia angustiflora (Ness)	RF	*Amblyseius tamatavensis*	13
		Amblyseius sp.4	6
		Amblydromalus limonicus	3
		Ambliseius herbicolus	4
		Neoseiulus tunus	14

		Typhlodromips japi Lofego, Demite & Feres	5
Sebastiania brasiliensis Spreng.	RF	*Iphiseiodes moraesi*	3
Sida rhombifolia L.	G and GE	*Amblyseius tamatavensis*	1
		Typholodromalus aripo	1
Symplocos uniflora (Pohl)	RF	*Amblyseius herbicolus*	4
		Agistemus floridanus	2
		Neoseiulus tunus	5
		Amblydromalus limonicus	8
		Phytoseius guianensis	4
		Amblyseius sp.1	1
Trichilia elegans A. Juss.	RF	*Typhlodromips japi*	4
		Agistemus sp.	1
		Agistemus floridanus	1
		Amblydromalus limonicus	1
		Amblyseius tamatavensis	1
Vernonanthura nudiflora (Less)	G and GE	*Typhlodromalus aripo*	3
		Zetzellia malvinae	1
Vitex megapotamica (Spreng.)	RF	*Neoseiulus tunus*	2
		Agistemus floridanus	3
		Typholodromalus aripo	1
		Amblyseius herbicolus	5
		Amblydromalus limonicus	5
		Amblyseius tamatavensis	3

G= Grassland; GE= Grazing Exclusions; RF = Riparian Forest; N = number of specimens.

Rank-abundance curves comparing Grassland, Grazing Exclusions and Riparian Forest had a sharp reduction in predatory mite fauna and their corresponding abundances in Grassland and Grazing Exclusions, compared to the community curve in Riparian Forest, thus indicating higher ecological stability in predator community in terms of abundance and richness (Fig. 4.2).

Both environments, Grassland and Grazing Exclusions, had a very similar pattern, with curves considerably overlapping, indicating fewer changes in the community of these two sites.

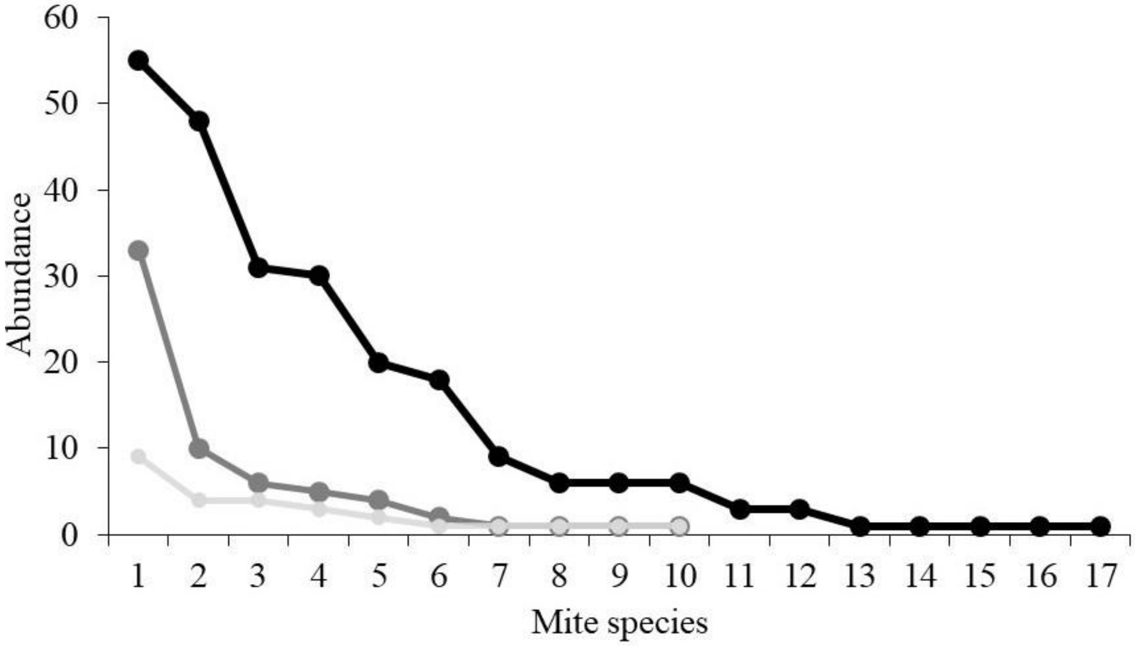

Figure 4.2: Rank-abundance curves of mite species, comparing the edge and inside areas of the three study sites. Black line Riparian Forest, light gray line Grazing Exclusions, and dark gray line Grassland.

Interrelated abiotic variables were evident at all three sites, with the first two PCA components explaining ca. 70% of the variation between environments (Fig. 4.3). However, the components were correlated differently in each environment. Component 1 (43.78%) was most correlated with photosynthetically active radiation (0.56) followed by wind speed (0.50). Component 2 (25.02%) was most correlated with temperature (-0.63) followed by soil moisture (0.57). Based on this multivariate analysis, the three study environments had differences regarding the abiotic factors analyzed, indicating that air humidity and soil moisture were more correlated in Riparian Forest, while higher photosynthetically active radiation, temperature, and wind speed were more correlated in Grassland and Grazing Exclusions.

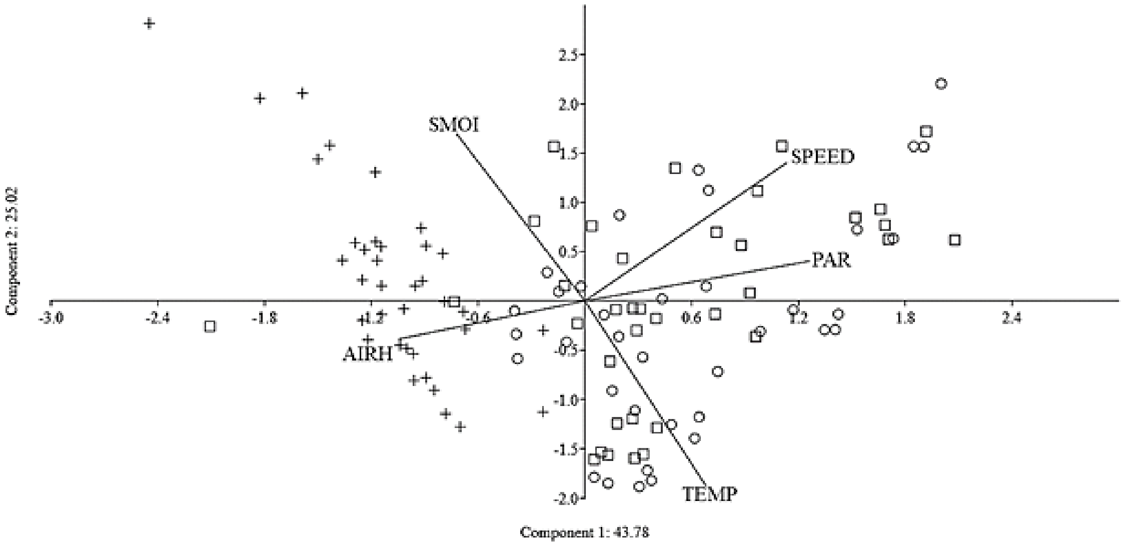

Figure 4.3: Principal component analysis (PCA). TEMP: Temperature (°C); SPEED: Wind Speed (km/h); SMOI: Soil moisture (%); AIRH: Air humidity (%); PAR: photosynthetically active radiation (µmol s-1 m-2). ○: Grassland, □: Grazing Exclusions: + Riparian Forest.

NMDS analysis ordinated the nine replicates of each environment by mite abundance and richness, thus emphasizing three groups, each one representing one sampled environment (Fig. 4.4). The disturbance value provided by the scaling (Stress value = 0.06) indicates an ordination fit for graphical representation. This result was confirmed by the ANOSIM analysis (global R = 0.462 $p = 0.001$), thus indicating the areas form different groupings. Comparisons between Grassland and Grazing Exclusions did not show any differences (R = 0.092 $p < 0.05$). On the other hand, there was difference between Grassland and Riparian Forest (R = 0.45 $p = 0.001$), and between Grazing Exclusions and Riparian Forest (R = 0.46, $p = 0.001$), thus emphasizing that Riparian Forest is the most heterogeneous environment regarding predatory mite fauna diversity and abundance, forming an independent grouping.

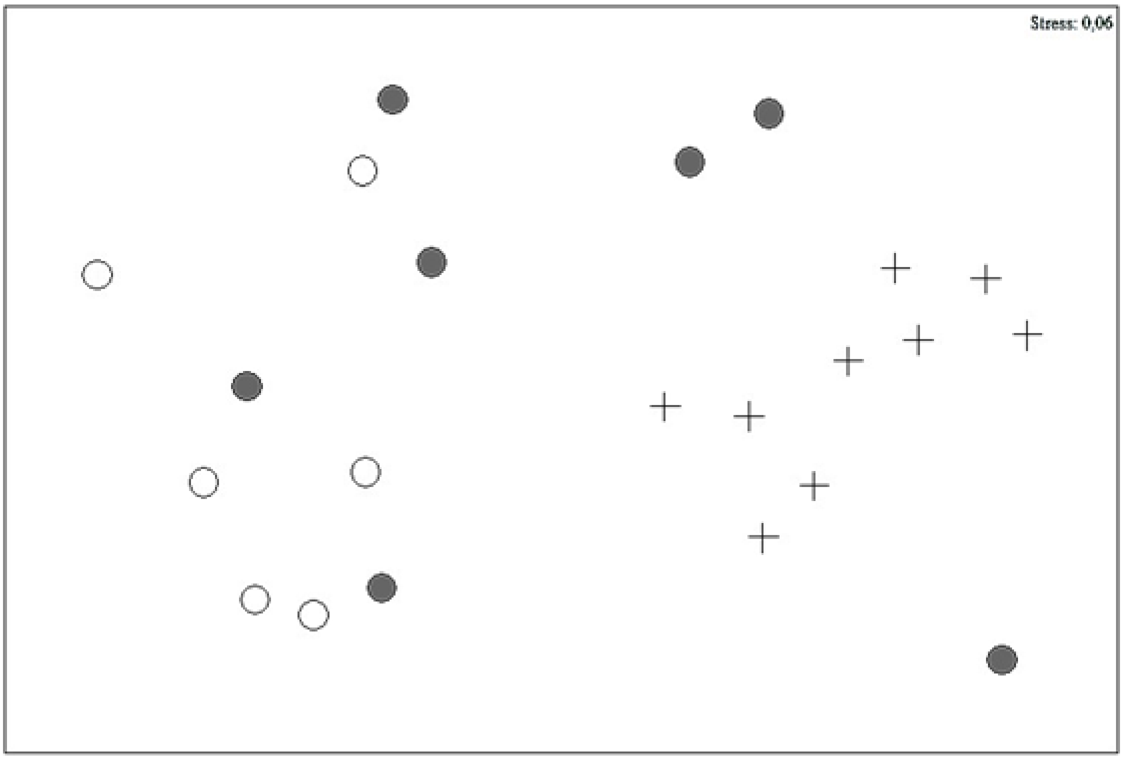

Figure 4.4: Ordination (NMDS) of mite species abundances from 3 sites x 3 samplings per environment. ○: Grassland, ●: Grazing Exclusions: + Riparian Forest. The low stress value (= 0.06) indicates good fit of the 2-dimensional representation of the distances between samplings.

Approximately 50% of mite species were environmental indicators or detectors. Riparian Forest was the environment with the highest number of mites that are environmental indicators, with emphasis on *A. tamatavensis*, *A. herbicolus*, *A. floridanus*, and *A. limonicus*, with IndVal higher than 0.50. They are related to the environment as they have higher abundance and frequency within the same habitat. There were two species, *Z. malvinae* and *Arrenoseius gaucho* Ferla, Silva & Moraes, in Grassland and only one species, *T. aripo*, in Grazing Exclusions with values lower than 0.50 (Tab. 4.2).

Table 4.2: Predatory mite species as indicators (IndVal) of Grassland (G), Grazing exclusion (GE) and Riparian Forest (RF), in decreasing order of IndVal.

IndVal	Predatory Mite Species	Environment
0.78	*Amblyseius tamatavensis*	RF
0.67	*Amblyseius herbicolus*	RF
0.62	*Agistemus floridanus*	RF
0.56	*Amblydromalus limonicus*	RF
0.44	*Typholodromalus aripo*	GE
0.44	*Iphyseiodes moraesi*	RF
0.44	*Typholodromips mangleae*	RF
0.38	*Neoseiulus tunus*	RF
0.33	*Amblyseius* aff. *euterpes*	RF
0.33	*Phytoseius guianensis*	RF
0.33	*Typholodromips japi*	RF
0.31	*Arrenoseius gaucho*	G
0.27	*Zetzellia malvinae*	G

Networks of mites and plants had 98 interactions (Robustness = 0.583; connectance = 0.090) (Fig. 4.5). *Amblyseius tamatavensis* was the Phytoseidae species with the largest number of interactions and *A. floridanus* was the Stigmeidae species with the largest number of interactions. *B. dracunculifolia* was the plant species with more interaction in the Grassland environment, and *E. molis* was the species with more interactions in Grazing Exclusions. Four plants had the largest number of interactions in Riparian Forest, *V. megapotamica*, *S. uniflora*, *R. angustiflora*, and *M. pungens.*

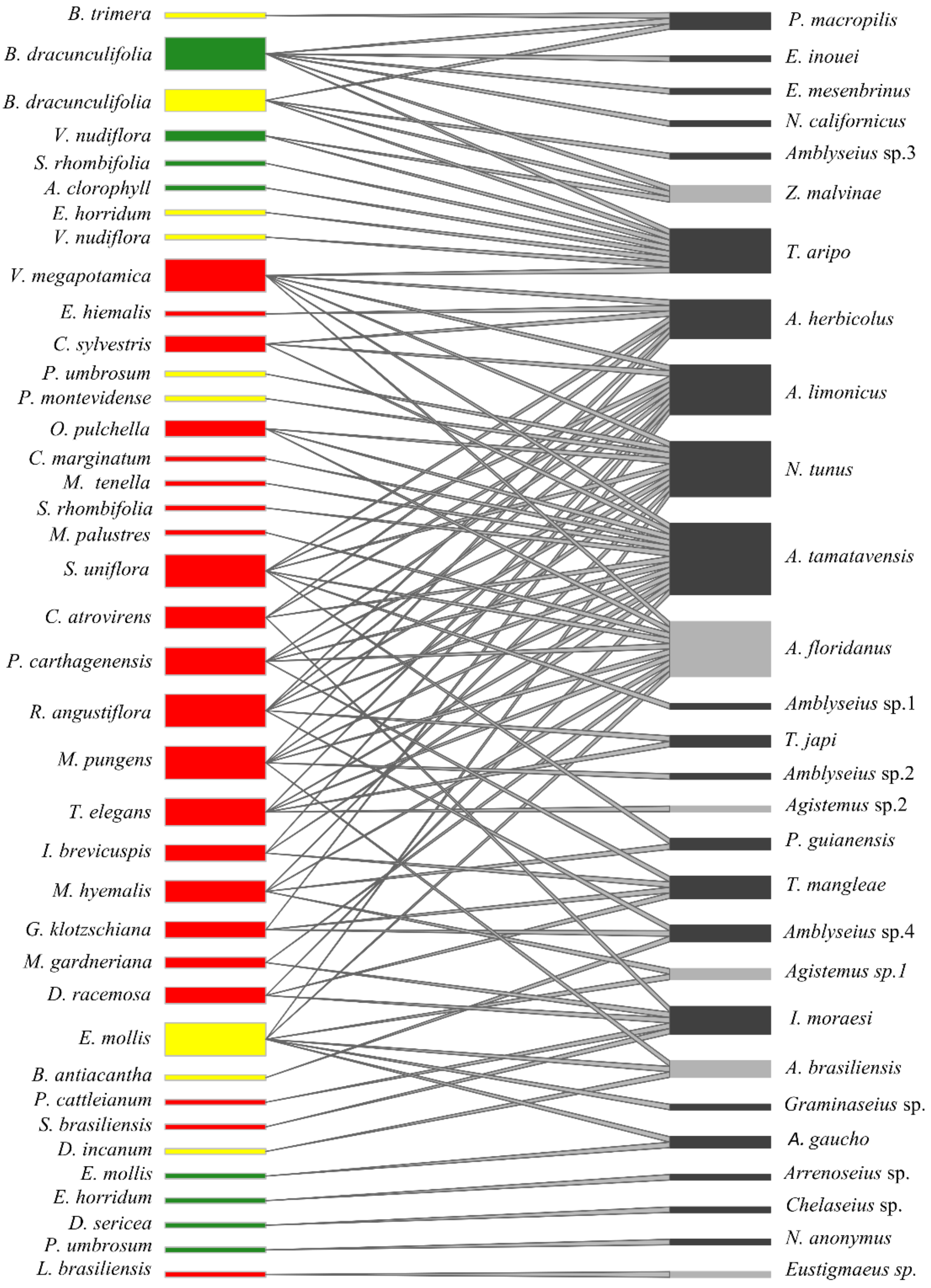

Figure 4.5: Bipartite network of mite and host plant species. Bars on the left side represent host plant species and bars on the right side represent mite species; green bars represent Grassland; yellow bars represent Grazing Exclusions; red bars represent Riparian Forest; grey bars are

Stigmeides and black bars represent Phytoseidae.

4.4 Discussion

The difference in climatic conditions and in resources in Riparian Forest provided a higher habitat variability, with reduced competition between species, which led to higher ecological stability of the associated predatory mite community (Cody *et al*. 1975). It was evident that Riparian Forest characteristics, such as higher humidity and milder winds, as well as lower incident radiation on the understory were more suitable for increased predatory mite richness and abundance. These results corroborate the findings by Tixier (2018), who observed that the major factors that increased predatory mite migration are high temperatures, light intensity, and water stress. Egg viability of predatory mites is higher in environments with higher air humidity, more than 80% (Devis *et al*. 2006). According to the same authors, trichomes and domatia on the leaves increase the relative air humidity at the foliar disk level, thus increasing egg viability. Therefore, low air humidity might be a limiting factor for predatory mites to settle in an environment.

Landscapes of surrounding environments might also affect mite diversity (Demite *et al*. 2015). Additionally, there is change in plant species composition due to the type of vegetation formation (Grassland and Riparian Forest) and presence of cattle (Grassland and Grazing Exclusions) (Boldrini *et al*. 2008; Carvalho *et al*. 2009; Freitas *et al*. 2010; Pinto *et al*. 2013). Therefore, these results agree with Cajaiba *et al*. (2017a; 2017b), who found that arthropod richness and abundance were sensitive to increased anthropogenic disturbances. Therefore, predatory mite community must be considered a valuable ecological indicator of the extent to which ecosystems have been disrupted in the Neotropical region.

Ten species were indicators of higher air humidity and soil moisture in Riparian Forest.

Amblyseius tamatavensis, *A. herbicolus*, and *A. floridanus* might also be related to a higher ecological stability of the predatory community in Riparian Forest. *Amblyseius tamatavensis* and *A. herbicolus* are globally distributed (Demite *et al*. 2019) and *A. floridanus* occurs in the American continent (Johann *et al*. 2013).

Arrenoseius gaucho, related to Grassland, has been described in a grassland environment, but there is no ecological information in literature regarding this species (Ferla *et al*. 2010). This species was indicator of the environment with higher temperature, luminosity, and exposure to wind. Similar to *Z. malvinae*, which is related to Grassland, it is distributed throughout Brazil and occurs in rubber trees, vineyards, citrus, and coffee crops (Matioli *et al.* 2002; Devis 2006; Mineiro *et al.* 2006; Diehl *et al.* 2012).

Only *T. aripo* was associated to Grazing Exclusions, thus indicating higher environmental instability. This species was also associated to unstable environments with extreme abiotic wind action (Rocha *et al.* 2015).

Predator richness found in the present study (28 species on 33 plant species) was similar to other studies. Ferla & Moraes (2002) found 23 species on 46 plant species; Rocha *et al*. (2015) reported 29 species on 62 plants. A total of 34 species have been found on 37 plants evaluated in the Brazilian Cerrado (Demite *et al*. 2017). The robustness of the networks was similar to that found by (Araujo and Kollár 2019). Many plants had more than one mite species and the interaction value was high; 98 interactions of 28 mite species. This can be explained by the fact that these mites are predators, as networks with phytophagous mites have lower connectivity (Araújo and Daud 2018a). *Typholodromalus aripo* had interactions in the three environments; this might be explained by the fact that it is a generalist species (McMurtry *et al.* 2013).

The list of predatory species with associated plants helps to identify new environmental resources preserved in this phytophysiognomy. Predatory mites are important biological control agents of other mite groups, and have been used in many biological control programs (Helle and Sabelis 1985; Lindquist *et al*. 1996; Sabelis and Van Rijn 1997; McMurtry *et al.* 2013; Gerson *et al*. 2003). The results obtained in this study indicate that natural vegetation remnants play an important role in the biological control of agroecosystems as some beneficial mites might migrate from native plants to plantations (Rocha *et al.* 2015; Demite *et al.* 2013; Tixier, 2018, Araújo & Daud, 2018b).

Plant species that maintain a higher number of predatory species could be the object of samplings for future studies on environmental indicators. They can also be considered predator reservoirs in applied biological control programs. These plants with higher predator richness are distributed in several countries and biomes and are widely distributed in South America (Ezcurra, 1989; Patrício & Cervi 2005; Borges & Forzza 2008; Carvalho 2008; Cruz *et al.* 2016). Tixier (2018) emphasizes the influence of plant species on predatory mite fauna, and the presence of trichomes and domatia is a positive trait. Plants and predatory mites have a common evolutionary history.

Even though *P. macropilis* (9) and *N. californicus* (1) were collected in small amounts in GE and G, they have economic importance in agro-ecosystems around the world. They are reported as applied biological control agents of mites and marketed in Brazil (Mapa 2019). Dameda *et al*. (2016) tested the potential of *P. macropilis* for controlling *Tetranychus urticae* (Koch) at different temperatures; *T. urticae* is found in a significant number of host plants, causing significant damages (Moraes & Flechtmann 2008). This predator also feeds on *Mononychellus planki* McGregor (Majolo & Ferla 2014). *Neoseiulus californicus* feeds on *T. urticae, Tetranychus ludeni* Zacher and *M. planki* (Toldi *et al.* 2016). Other predators found in this study have had their economic importance in biological control proved in other studies: *Agistemus floridanus* (Ferla & Moraes 2003), *A. limonicus* (Vangansbeke 2014), *A. tamatavensis*

(Cavalcante *et al.* 2017), *E. mesembrinus* (Landeros *et al.* 2004), *T. aripo* (Onzo *et al.* 2012).

Some authors have recently tried to identify the factors that affect mite fauna in different environments (Rocha *et al.* 2015; Rieff *et al.* 2016; Demite *et al.* 2017; Tixier, 2018). However, many variables affect the establishment of these mites, such as humidity, radiation, temperature, and morphology of plants where these organisms can be found. The results in our study provide sufficient evidence that these predators respond to changes in temperature, humidity, and composition of the associated plants. The hypothesis that there would be an increase in predatory mite richness and abundance in the forest environment was confirmed. Although the forest Environment had higher predatory mite richness and abundance, grasslands were the environments that had the two major predatory mite species using in biological control in Brazil (*N. californicus* and *P. macropiris*). The different environments of the Brazilian Pampa conserve a rich diversity of predators and might foster biological control programs. Further studies might be conducted to define strategies for the conservation of these predators.

5 FOUR NEW SPECIES OF ERIOPHYOID MITES (ACARI: TROMBIDIFORMES: ERIOPHYOIDEA) ON NATIVE VEGETATION FROM PAMPA BIOME, IN THE STATE OF RIO GRANDE DO SUL (BRAZIL)

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Abstract

Four new species of eriophyoid mites (Acari: Trombidiformes: Eriophyoidea) were collected from native plant species from Pampa biome, in the state of Rio Grande do Sul, Brazil, and are described and illustrated herein. They have been named: *Aculus pampae* **sp. nov.** on *Elephantopus mollis* Kunth (Asteraceae); *Costarectus lanceatus* **sp. nov.** on *Sebastiania brasiliensis* Spreng. (Euphorbiaceae); *Dichopelmus sigfeminae* **sp. nov.** and *Rhynacus symflori* **sp. nov.** both on *Symplocos uniflora* (Pohl) Benth. (Symplocaceae). All new species appeared to be vagrants and no apparent damage was associated to them.

Keywords: *Tegolophus, Aculus, Rhynacus, Neocolopodacus*, vagrant

5.1 Introduction

Pampa biome covers a huge area in the geographic region including Argentina, Brazil, Paraguay and Uruguay. This biome shows high fauna and flora biodiversity, and high rates of endemism (Behling *et al.* 2005; 2009; Bencke 2009; Carvalho *et al.* 2009; Roesch *et al.* 2009). The vegetation of this biome is mainly characterized by grasses and riparian forests (Behling *et al.* 2005; Carvalho *et al.* 2009; Roesch *et al.* 2009).

The economic importance of Eriophyoidea as plant feeders is overcome only by the Tetranychidae and many eriophyoid species are considered pests in several parts of the world (Moraes & Flechtmann 2008; de Lillo *et al*. 2018). This taxon is composed of the families Eriophyidae, Diptilomiopidae and Phytoptidae, and most of them are highly host specific (Skoracka *et al*. 2009). Diptilomiopids are vagrants on leaves, while eriophyids and phytoptids include the most of pests on broad-leaf plants. Many eriophyids are also involved in the transmission of plant pathogens (de Lillo *et al.* 2018).

Little is known about the Eriophyoidea fauna in southern Brazil. Recently, the occurrences of *Aculus schlechtendali* Keifer in apple orchards (Ferla *et al.* 2018) and the description of *Diptilostatus chimarricus* Silva & Ferla (Diptilomiopidae) on yerba mate (*Ilex paraguariensis* A. St.-Hill.; Aquifoliaceae) (Silva *et al.* 2017) have been added to the previous records (de Azambuja & da Silva 1982; Ferla *et al.* 2005; Moura *et al.* 2008; Johann *et al.* 2009; Ricalde *et al.* 2012 and few older papers).

The present contribution provides the description and illustration of four new eriophyoid mites collected on plant species typical of the Pampa biome, state of Rio Grande do Sul, Brazil.

5. 2 Materials and methods

During 2017-2018 samples of leaves were collected from Pantano Grande county (30°19'18.00"S, 52°29'00.00"W; 30°19'15.00"S, 52°28'50.00"W and 30°19'45.00"S, 52°29'50.00"W), state of Rio Grande do Sul, Brazil (Fig. 5.1). The samples were examined under a stereomicroscope Leica EZ4HD in the laboratory of Acarology of Univates University. The specimens were slide mounted in Hoyer’s medium (Jeppson *et al.* 1975) without posing fibers between slide and coverslip.

Slide mounted specimens were studied using a phase-contrast microscope Olympus

BX50 in the laboratory of University of Bari Aldo Moro, Italy. The terminology and setal notation used in the morphological description of the species follow mainly Lindquist (1996). The genera were compared with those reported into the generic key by Amrine *et al.* (2003) and with those described and redescribed after 2003 within the subfamilies Phyllocoptinae and Diptilomiopinae (Amrine & de Lillo, unpublished database). The number of measured specimens (*n*) is given within parentheses in the description of each stage. Measurements were made according to Amrine and Manson (1996) as modified by de Lillo *et al.* (2010) and are given in micrometers (µm). Slight clarifications should be added as follows: dorsal semiannuli were counted from the first semiannulus behind the rear margin of the prodorsal shield; ventral semiannuli were counted from the first complete annulus after coxae II; coxigenital semiannuli were counted medially from the coxal region to the anterior margin of the external genitalia and were not included in the ventral semiannuli count. Measurements and means are rounded off to the nearest integer when required, referring to the length of the morphological traits unless otherwise specified. The holotype measurements are followed by the range values of the studied population (holotype plus measured paratypes) which are given in parentheses. When the measurements of the holotype could not be taken, due to the slide mounting position of the specimens, the mean values of the paratypes are reported and are marked in the description by an asterisk (*). Line drawings were hand-made through a *camera lucida* according to de Lillo *et al.* (2010) and the figure abbreviations follow mainly Amrine *et al.* (2003). Drawings of the latero-anterior side of *Rhynacus symflori* **sp. nov.** was not possible to be taken. The host plant names are in accordance with "*The plant list on-line database*" (2013).

Type materials are deposited at reference collection of the Museum of Natural Science (ZAUMCN), Univates University, Lajeado, the State of Rio Grande do Sul, Brazil, and at the Department of Soil, Plant and Food Sciences (DiSSPA), University of Bari Aldo Moro, Italy

(UNIBA), formerly indicated as UBI by Zhang (2018).

5.3 Family Eriophydae

Subfamily Phyllocoptinae

Tribe Anthocoptini

Aculus pampae Toldi, de Lillo & Ferla sp. nov. (Fig. 5.2)

Description. FEMALE: (n = 6). **Body** fusiform, 140 (126–159, including gnathosoma), 56 (53–57) width, 55* (55–56) thickness. **Gnathosoma** 17 (17–20) projecting obliquely down, pedipalp coxal setae *ep* 2 (no range), dorsal pedipalp genual setae *d* 3 (2–3), unbranched, palp tarsus seta *v* very short. Cheliceral stylets 12 (12–13). **Prodorsal shield** 31 (30–33), including frontal lobe, 50* (48–52) width; semicircular in anterior shape with a broad, semicircular frontal lobe 3 (3–4) over gnathosomal base. Indistinct pattern composed of rounded small granules more concentrated on lateral and anterior shield margin; two faints granulated admedian lines, medially convergent on the middle half of the shield, extended from the frontal lobe up to the rear shield margin. Tubercles of scapular setae *sc* papilla-shaped, on rear shield margin, 23 (20–23) apart, scapular setae *sc* 10 (no range), projecting posteriorly. **Legs** with all usual segments and setae. **Leg I** 27 (25–28), femur 7 (7–10), genu 4 (no range), tibia 5 (5–6), tarsus 5 (no range), solenidion *ω* 5 (5–6) distally lightly knobbed, empodium simple, 4 (4–5), 6-rayed; femoral setae *bv* 7 (7–8), genual setae *l″* 15 (14–16), tibial setae *l′* 4 (3–4), tarsal setae *ft′* 13 (12–14), setae *ft″* 19 (16–19). **Leg II** 25 (24–26), femur 8 (7–9), genu 4 (no range), tibia 4 (4–5), tarsus 5 (5–6), solenidion *ω* 5 (5–6) distally lightly knobbed, empodium simple, 5 (4–5), 6-rayed; femoral setae *bv* 9 (8–9), genual setae *l″* 4* (3–4), tarsal setae *ft′* 4 (3–4), setae *ft″* 15 (15–16). **Coxae** with short lines; setae *1b* 5 (5–6), tubercles *1b* 7 (7–9) apart, setae *1a* 12 (no range), tubercles *1a* 7 (7–9) apart, setae *2a* 30 (27–30), tubercles *2a* 20 (19–20) apart. Prosternal apodeme 6 (5–6). **Opisthosoma** arched, with 25 (no range) dorsal semiannuli, 44 (41–49) ventral semiannuli and 9 (8–9) semiannuli between coxae and genital region. Small and slightly elliptical microtubercles on ventral and lateral sides of the ventral semiannuli; linear microtubercles on the last 4-5 annuli; sparse and elongated microtubercles on the dorsal semiannuli. Setae *c2* 6 (6–7), on ventral semiannulus 7 (6–7); setae *d* 29 (24–29), on ventral semiannulus 17 (15–18); setae *e* 7 (7–8), on ventral semiannulus 25 (25–30); setae *f* 13 (12–15), on ventral semiannulus 41 (37–45), 4 annuli after setae *f*. Setae *h1* 3 (2–3), setae *h2* 32 (32–35). **Female genitalia** 15 (no range), 20 (19–20) width, coverflap with 7 (4–9) longitudinal striae, setae *3a* 6 (no range), 12 (11–12) apart.

MALE. Measurements not taken.

Type host plant. *Elephantopus mollis* Kunth (Asteraceae), tobacco weed or soft elephantsfoot is a pantropical and perennial herb bush native from American continent and occurring in all regions of Brazil (Cruz *et al.* 2016).

Relation to the host plant. No apparent symptom was observed.

Type locality. Protected fields, Pantano Grande county, state of Rio Grande do Sul, Brazil (30°19'18.00"S, 52°29'00.00"W), 126 m above sea level, February 2018, coll. M. Toldi.

Type material. Holotype female and five paratype females on a microscope slide (35MT), in Italy. Four paratypes and 1 male on a microscope slide (36MT), in Brazil.

Etymology. The name *pampae* refers to the place (Pampa biome) where the mite was collected. The name was Latinized in the genitive case.

Differential diagnosis. More than 300 *Aculus species* are known worldwide (Tajaddod *et al*. 2018). Among the five *Aculus* species associated to Asteraceae, *Aculus rigidus* (Nalepa 1894) and *Aculus osteospermi* Meyer (Smith) & Ueckermann, 1990, are provided with longitudinal lines on the prodorsal shield. *Aculus ambrosiae* (Keifer 1943) and *Aculus maximilianae* (Briones & McDaniel 1976) have granulated lines on the prodorsal shield. *Aculops helichrysi* Meyer (Smith) & Ueckermann, 1990, and *Vasates asteri* Petanović & Boczek, 2000, have morphological characters which fit with *Aculus* type and have to be reassigned to this genus. The first one is provided with a lined prodorsal shield design, the second one has a largely smooth prodorsal shield with a curved transverse lines close to the rear margin. Also, *Aculus asteri* Xue, Song & Hong, 2008, was found on an Asteraceae and its homonymy with the species described by Petanović and Boczek should be managed. This last species seems to be morphologically close to *A. pampae* **sp. nov.** from which it differs for: a transverse line on the posterior half of the prodorsal shield among the tubercles of setae *sc* (*A. pampae* **sp. nov.** does not show any transverse line and some granules are lined as in two admedian lines); a lower number of rays on the empodium (4 *versus* 6 of *A. pampae* **sp. nov.**); the distribution of microtubercles on the dorsal semiannuli (smooth in *A. asteri* versus sparse microtuberculated dorsal semiannuli on the region anterior to setae *f*. in *A. pampae* **sp. nov.**).

Remarks. A male of *A. pampae* **sp. nov.** was found but measurements were not possible to be taken because the specimen was deformed. However, the qualitative traits related to the prodorsal shield fitted perfectly with those of the female.

Also, a Diptilomiopid mite, *Diptilomiopus holmesi* (Keifer 1962), was previously collected on *E. mollis* in Philippine Islands and it was reported to be vagrant. Further eight species within the genus *Aculus* were reported from Brazil (Amrine & de Lillo unpublished database), but any of them was associated to the Asteraceae.

Subfamily Phyllocoptinae

Tribe Anthocoptini

Costarectus lanceatus Toldi, de Lillo & Ferla sp. nov. (Fig. 5.3)

Description. FEMALE: (n = 7). **Body** fusiform, 195 (180–225, including gnathosoma), 62 (55–62) width, 54* (50–62) thickness. **Gnathosoma** 25* (25–30) projecting obliquely down, pedipalp coxal setae *ep* 3* (no range), dorsal pedipalp genual seta *d* 9 (7-10), unbranched, palp tarsus setae *v* very short, cheliceral stylets 12 (12–13). **Prodorsal shield** 54 (50–55), including frontal lobe, 60 (60–72) width, frontal lobe 8 (8–9). An interrupted median line on the posterior half and tortuous admedian lines; two open cells close to the rear margin of the prodorsal shield composed of median and admedian lines; lanceolate-shaped figure on the anterior prodorsal shield composed of the tourtuous admedian lines alone; submedian lines with short outer branches. The anterior part of the lanceolate-shaped figure provided with outer short lines producing wax. Tubercles of scapular setae *sc* finger-like and on the rear margin, scapular setae *sc* 16 (16–19), 39 (36–39) apart. **Legs** with all usual segments and setae. **Leg I** 35 (33–37), femur 12 (11–13), genu 5 (4–6), tibia 10 (9–10), tarsus 5 (5–6), solenidion *ω* 5 (5–6), distally knobbed, empodium simple, 4 (no range), 4-rayed; femoral setae *bv* 10 (9–11), genual setae *l″* 33 (30–34), tibial setae *l′* 7 (5–7), tarsal setae *ft′* 18 (17–19), setae ft″ 18 (18–20). **Leg II** 32 (29–33), femur 12 (10–12), genu 4 (4–5), tibia 7 (6–7), tarsus 5 (no range), solenidion *ω* 5 (no range), distally knobbed, empodium simple, 4 (4–5), 4-rayed; femoral setae *bv* 8 (7–10), genual setae *l″* 6 (6–7), tarsal setae *ft′* 5 (no range), setae *ft″* 17* (16–19). **Coxae** with short lines on the first pair, second pair smooth; setae *1b* 6 (no range), tubercles *1b* 9 (7–9) apart, setae *1a* 8 (8–13), tubercles *1a* 6 (6–7) apart, setae *2a* 49 (30–49), tubercles *2a* 25 (20–25) apart. Prosternal apodeme 7 (7–9). **Opisthosoma** with 46 (42–46) dorsal semiannuli, 59 (57–63) ventral semiannuli, 8 (no range) smooth semiannuli between coxae and genital region. Five longitudinal ridges (one median, two lateral and two sublateral) on the dorsal side; median ridge ending at posterior third of opisthosoma. The ridges produce strips of wax. Linear microtubercles on the dorsal side, located only on the ridges. Rounded and small microtubercles on the ventral and lateral sides of the opisthosoma. Setae *c2* 19 (18–23), on ventral semiannulus 3 (3–4), setae *d* 66* (57–74), on ventral semiannulus 15 (13–16); setae *e* 16 (15–17), on ventral semiannulus 36 (33–37); setae *f* 20 (17–20), on ventral semiannulus 56 (52–59), 5 (4–6) annuli after. Setae *h1* 2 (no range), setae *h2* 71* (67–76). **Female genitalia** 19 (16–19), 30 (25–33) width, coverflap with 7 (6–7) longitudinal striae on the proximal half and on the distal half, setae *3a* 10 (9–11), 19 (15–18) apart.

MALE (n = 1). **Body** fusiform, 181 (including gnathosoma), 53 width, 48 thickness. **Gnathosoma** 20, projecting obliquely downwards, cheliceral stylets 15 palp genual setae *d* 10, unbranched, palp tarsus setae *v* very short. **Prodorsal shield** 50 including the frontal lobe, 55 width, frontal lobe 7. Shield pattern similar to that of the female. Setae *sc* 16, sc 35 apart. **Leg I** 34, femur 12, genu 5, tibia 10, tarsus 5, solenidion *ω* 5, distally knobbed, empodium simple, 4, 4-rayed; femoral setae *bv* 9, genual setae *l''* 30, tibial setae *l'* 10, tarsal setae *ft'* 16, setae *ft''* 18. **Leg II** 31, femur 10, genu 5, tibia 7, tarsus 5, solenidion *ω* 5, distally knobbed, empodium simple, 4, 4-rayed; femoral setae *bv* 10, genual setae *l''* 7, tarsal setae *ft'* 3, setae *ft''* 18. **Coxae** ornamented like in the female; setae *1b* 6, tubercles 1b 9 apart, setae *1a* 10, tubercles *1a* 6 apart, setae *2a* 37, tubercles *2a* 21 apart. Prosternal apodeme 9. **Opisthosoma** similar to that of the female, with 42 dorsal semiannuli, 60 ventral semiannuli, 8 microtuberculated semiannuli between coxae and genital region. Setae *c2* 18 on ventral semiannulus 3, setae *d* 60 on ventral semiannulus 13, setae *e* 17 on ventral semiannulus 32, setae *f* 18 on ventral semiannulus 54, 4 annuli after setae *f*. Setae *h1* very short, *h2* 60. **Male genitalia,** setae *3a* 7, 13 apart.

Type host plant. *Sebastiania brasiliensis* Spreng. (Euphorbiaceae). It is a tree with pantropical distribution, occuring in Argentina, Bolívia, Brazil, Paraguay and Uruguay (Sobral' *et al.* 2013).

Relation to the host plant. No apparent symptom was observed.

Type locality. Riparian protected forest, in Pantano Grande County, Pampa biome, state of Rio Grande do Sul, Brazil (30°19'45.00"S, 52°29'50.00"W), 112 m above sea level; February 2018, coll. M. Toldi.

Type material. Holotype female on a microscope slide (41MT) in Italy. Two paratype females and one male on a microscope slide (27MT) and four paratype females and six males on a microscope slide (28MT), in Brazil.

Etymology. The name species comes from the Latin adjective *lanceatus, -i,* which means arrowead and refers to the particular shape of the prodorsal shield pattern; it is in the nominative singular case.

Differential diagnosis. This is the third species known within the genus *Costarectus*, after *C. zeyheri* Meyer (Smith) & Ueckermann, 1995, and *C. akoensus* Huang, 2001. The new species has morphological characters consistent with those reported for the type genus, *C. zeyheri*. The morphology of the prodorsal shield pattern of *C. lanceatus* **sp. nov.** is largely different from that of *C. akoensus*, characterized by two admedian lines and a short submedian line on the anterior half, and from that of *C. zeyheri*, characterized by a median line on the posterior third, complete admedian lines, and a curved submedian line rising from the third anterior of the admedian line.

Subfamily Phyllocoptinae

Tribe Acaricalini

Dichopelmus sigfeminae Toldi, de Lillo & Ferla sp. nov. (Fig. 5.4)

Description. FEMALE: (n = 10). **Body** fusiform, 194 (192–230, including gnathosoma) long, 68 (68–75) width, 70* (67–75) thickness. **Gnathosoma** 23* (18–27) projecting obliquely down, pedipalp coxal setae *ep* 3 (2–3), dorsal pedipalp genual seta *d* 4 (3–4), unbranched, palp tarsus setae *v* very short, cheliceral stylets 13 (12–17). **Prodorsal shield** 46 (46–57), including frontal lobe, 64 (63–70) width, frontal lobe length 5 (4–6). Median line combined with admedian lines on the posterior half of the prodorsal shield forming two pairs of cells; median line on the anterior quarter of the prodorsal shield, interrupted by a short transversal line on the frontal lobe forming a sort of cross; admedian lines on the anterior half forming a median inpair cell and joining with a pair of arch-shaped transversal (submedian) lines coming from the lateral sides of the prodorsal shield; three pairs of short lines coming out from this trasversal line; a further transversal line is between and ahead the tubercles of scapular setae *sc*. Tubercles of scapular setae sc papilla-shaped, on rear shield margin, scapular setae *sc* 48 (37–52), 34 (34–38) apart. **Legs** with all usual segments and setae. **Leg I** 32 (31–33), femur 10 (10–11), genu 4 (4–5), tibia 9 (7–9), tarsus 6 (5–6), solenidion *ω* 6* (5–6), distally knobbed, empodium divided in two arms 4 (4–5), 5-rayed; femoral setae bv 8 (8–13), genual setae *l″* 24 (19–25), tibial setae *l′* 3 (no range), tarsal setae *ft′* 14 (12–16), setae ft″ 18 (14–20). **Leg II** 30 (29–32), femur 10 (9–10), genu 4 (4–5), tibia 7 (7–10), tarsus 6 (5–6), solenidion *ω* 5 (5–6), distally knobbed, empodium divided in two arms, 4 (no range), 5-rayed; femoral setae *bv* 8 (8–12), genual setae *l″* 7 (7–10), tarsal setae *ft′* 5* (4–6), setae *ft″* 18* (16–19). **Coxae**, setae *1b* 6 (no range), tubercles *1b* 9 (8–10) apart, setae *1a* 17 (10–22), tubercles *1a* 8 (7–9) apart, setae *2a* 46 (43–50), tubercles *2a* 22 (22–24) apart. Prosternal apodeme 8 (7–8). **Opisthosoma** with 35 (32–36) dorsal semiannuli, 56 (51–57) ventral semiannuli, longitudinal middle depression and 5 (4–6) semiannuli between coxae and genital region. Setae *c2* 35 (28–36), on ventral semiannulus 6 (5–9), setae *d* 59 (59–66), on ventral semiannulus 21 (20–25); setae *e* 20 (19–24), on ventral semiannulus 35 (31–39); setae *f* 23 (18–

24), on ventral semiannulus 50* (46–54), 5 (no range) annuli after. Setae *h1* 1 (no range), setae *h2* 64 (57–68). **Female genitalia** 14 (12–18), 22 (19–30) width, coverflap with 5 (4–6) longitudinal striae, setae *3a* 14 (12–19), 14 (14–15) apart.

MALE (n = 1). **Body** fusiform, 171 (including gnathosoma), 40 width, 60 thickness. **Gnathosoma 18,** projecting obliquely downwards, cheliceral stylets 12, palp coxal setae *ep 2,* palp genual setae *d* 4, unbranched, palp tarsus setae *v* very short. **Prodorsal shield** 47 including the frontal lobe, 56 width, frontal lobe 6. Shield pattern similar to that of the female. Setae *sc* 42, sc 30 apart. **Leg I** 28, femur 9, genu 4, tibia 8, tarsus 6, solenidion *ω* 5, distally knobbed, empodium divided in two arms, 4, 5-rayed; femoral setae *bv* 6, genual setae *l''* 20, tibial setae *l'* 3, tarsal setae *ft'* 15, setae *ft''* 16. **Leg II** 28, femur 8, genu 4, tibia 7, tarsus 6, solenidion *ω* 5, distally knobbed, empodium divided in two arms, 4, 5-rayed; femoral setae *bv* 8, genual setae *l''* 4, tarsal setae *ft'* 4, setae *ft''* 14. **Coxae** ornamented like in the female; setae *1b* 5, tubercles *1b* 9 apart, setae *1a* 12, tubercles *1a* 6 apart, setae *2a* 36, tubercles *2a* 19 apart. Prosternal apodeme 8. **Opisthosoma** similar to that of the female, with 34 dorsal semiannuli, 48 ventral semiannuli. Setae *c2* 28 on ventral semiannulus 6, setae *d* 50 on ventral semiannulus 16, setae *e* 15 on ventral semiannulus 28, setae *f* 19 on ventral semiannulus 43, 5 annuli after setae *f*. Setae *h1* very short, *h2* 57. **Male genitalia**, setae *3a* 3, 12 apart.

Type host plant. *Symplocos uniflora* (Pohl) Benth. (Symplocaceae) is a native tree species in Argentina, Brazil, Paraguay and Uruguay (Sobral *et al.* 2013).

Relation to the host plant. No apparent symptom was observed.

Type locality. Riparian protected forest, Pantano Grande county, Pampa biome, Brazil (30°19'15.00"S, 52°28'50.00"W), 124 m above sea level; February 2018, coll. M. Toldi.

Type material. Holotype female, five paratype females and two males on a microscope slide (46MT), in Italy. Four paratype females and twelve males on microscope slide (31MT), in Brazil.

Etymology. The epithet comes from the combination of the initial part of the Latin words *signum, -i*, meaning mark, symbol, and *femina, -ae*, meaning female, in the genitive singular case. This name was chosen because of a mark on the anterior half of the prodorsal shield which looks like the overturned graphic female symbol.

Differential diagnosis. This is the tenth species within the genus *Dichopelmus*. *Dichopelmus sigfeminae* **sp. nov.** is distinctive in the prodorsal shield, composed of fours cells in the middle field on the posterior half and on a typical design resembling the upturned female symbol. This pattern is unique among the species previously assigned to the genus *Dichopelmus* among which no cells were observed on the prodorsal shield.

5.4 Family Dipitilomiopidae

Subfamily Diptilomiopinae

Rhynacus symflori Toldi, de Lillo & Ferla sp. nov. (Fig. 5.5)

Description. FEMALE: (n = 7). **Body** fusiform, 202 (185–228, including gnathosoma) long, 89 (80–92) width, 90* (85–95) thickness. **Gnathosoma** 50* (49–50) projecting obliquely down, pedipalp coxal setae *ep* 2 (2–3), dorsal pedipalp genual seta *d* not detected, palp tarsus seta *v* 5 (4–6), cheliceral stylets 17 (17–19). **Prodorsal shield** 37 (33–37), without frontal lobe, 73 (70–74) width. Shield pattern composed of 9 cells on the latero-anterior side, 6 cells on the latero-posterior row and 3 cells in the middle row. Tubercles of scapular setae *sc* present and located ahead the shield margin each within a cell; setae *sc* absent. **Legs** with all usual segments; femoral setae absent on both legs, genual setae l'' absent on leg II. **Leg I** 38 (38–42), femur 14 (14–15), genu 5 (4–5), tibia 5 (4–6), tarsus 12 (12–14), solenidion *ω* 7 (7–8), knobbed, empodium divided in two arms, 9 (9–10), each 7-rayed; genual setae *l''* 48 (45–49), tibial setae *l'* 3 (3–4), tarsal setae *ft'* 36 (35–38), setae *ft''* 36 (35–38). **Leg II** 36 (35–39), femur 13 (12–14), genu 3 (3–4), tibia 5 (4–5), tarsus 11 (10–12), solenidion *ω* 7 (7–8), knobbed, empodium divided, 9 (8–10), each 7-rayed; tarsal setae *ft'* 8 (7–9), setae *ft''* 33 (30–35). **Coxae**, setae *1b* absent, setae *1a* 31 (31–34), tubercles *1a* 5 (4–8) apart, setae *2a* 62 (62–63), tubercles *2a* 23 (23–25) apart. Coxae I well separated each from the other by a furrow at the level of the prosternal apodeme, 13 (13–15). Coxae I with several short lines; coxae II with very few lines. **Opisthosoma** with smooth median ridge, with 54 (52–56) dorsal semiannuli, and 73 (66–78) ventral semiannuli, 7 microtuberculated semiannuli between coxae and genital region. Small and rounded microtubercles on the ventral semiannuli and on the lateral sides of dorsal semiannuli, linear microtubercles on the rings from the setae *f* to the anal lobes. Setae *c2* absent, setae *d* 65 (65–74), on ventral semiannulus 21 (18–22); setae *e* 22 (18–22), on ventral semiannulus 41 (35–46); setae *f* 45 (42–47), on ventral semiannulus 65 (56–71), 11 (10–12) annuli after setae *f*. Setae *h1* 1 (no range), setae *h2* 100 (87–114). **Female genitalia** 24 (19–24), 29 (28–30) width, coverflap with 8 (7–8) longitudinal striae on the distal part, setae *3a* 10 (7–10), 21 (20–23) apart.

MALE (n = 1). **Body** fusiform, 189 (including gnathosoma), 75 width. **Gnathosoma** projecting obliquely downwards, cheliceral stylets 17 palp coxal setae *ep* 2, dorsal pedipalp genual seta *d* not detected, palp tarsus seta *v* 6. **Prodorsal shield** 31, 60 width, frontal lobe absent. Shield pattern similar to that of the female. Setae *sc* absent, tubercles present and placed ahead posterior shield margin. **Legs** with all usual segments; femoral setae absent, genual setae *l"* on leg II absent. **Leg I** 35, femur 13, genu 4, tibia 4, tarsus 12, solenidion *ω* 6, knobbed, empodium divided, 9, 7-rayed; genual setae *l''* 37, tibial setae *l'* 4, tarsal setae *ft'* 38, setae *ft''* 39. **Leg II** 29, femur 10, genu 4, tibia 4, tarsus 8, solenidion *ω* 6, knobbed, empodium divided in two arms, 9, 7-rayed; tarsal setae *ft′* 5, setae *ft″* 26. **Coxae** with ornamentation similar to those of the female; setae *1b* absent, setae *1a* 25, tubercles *1a* 5 aparts, setae *2a* 52, tubercles *2a* 20 apart. Coxae I well separated each from the other by a furrow at the level of the prosternal apodeme 12. **Opisthosoma** with 49 dorsal semiannuli, 60 ventral semiannuli, 7 microtuberculated semiannuli between coxae and genital region. Setae *c2* absent, setae *d* 50 on ventral semiannulus 14; setae *e* 15 on ventral semiannulus 30; setae *f* 33 on ventral semiannulus 50, 10 annuli after setae *f*. Setae *h2* 93, *h1* 1 (no range). **Male genitalia,** setae *3a* 7, 18 apart.

Type host plant. *Symplocos uniflora*.

Relation to the host plant. No apparent symptom was observed.

Type locality. Riparian protected forest, Pantano Grande County, Pampa biome, state of Rio Grande do Sul, Brazil (30°19'15.00"S, 52°28'50.00"W), 124 m above sea level; November 2017 and February 2018, coll. M. Toldi.

Type material. Holotype female, four paratype females and two males on a microscope slide (25MT), in Italy. Two paratype females on microscope slide (26MT), in Brazil.

Etymology. The epithet comes from the combination of the initial part of the plant genus name, *Sym-*, with the end of the plant species name, *-flori*, in the genitive singular case.

Differential diagnosis. Five *Rhynacus* species are currently known. *Rhynacus acerioides* Flechtmann, 2014, does not show any tubercles of setae *sc* on the contrary of all the other *Rhynacus* species as well as *R. symflori* **sp. nov.** All known *Rhynacus* species are provided with a paired number of cells on the median field of the prodorsal shield on the contrary of *R. sympflori* **sp. nov.** which is provided with two rows of impared cells (three and seven in first and second row, recpectivelly) in the middle field.

CAPTIONS

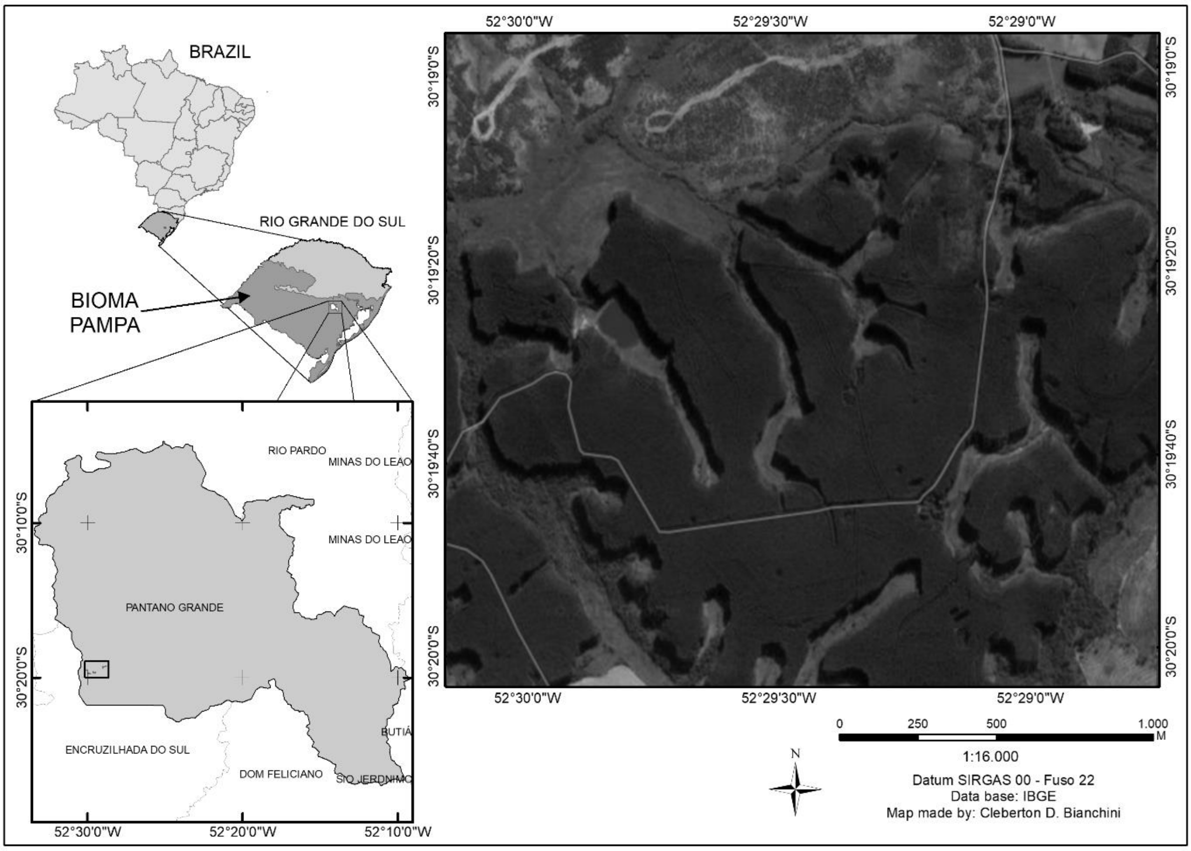

FIGURE 5.1. Collection sites of samples infested by the eriophyoid mites in Pantano Grande County, Pampa biome, state of Rio Grande do Sul, Brazil.

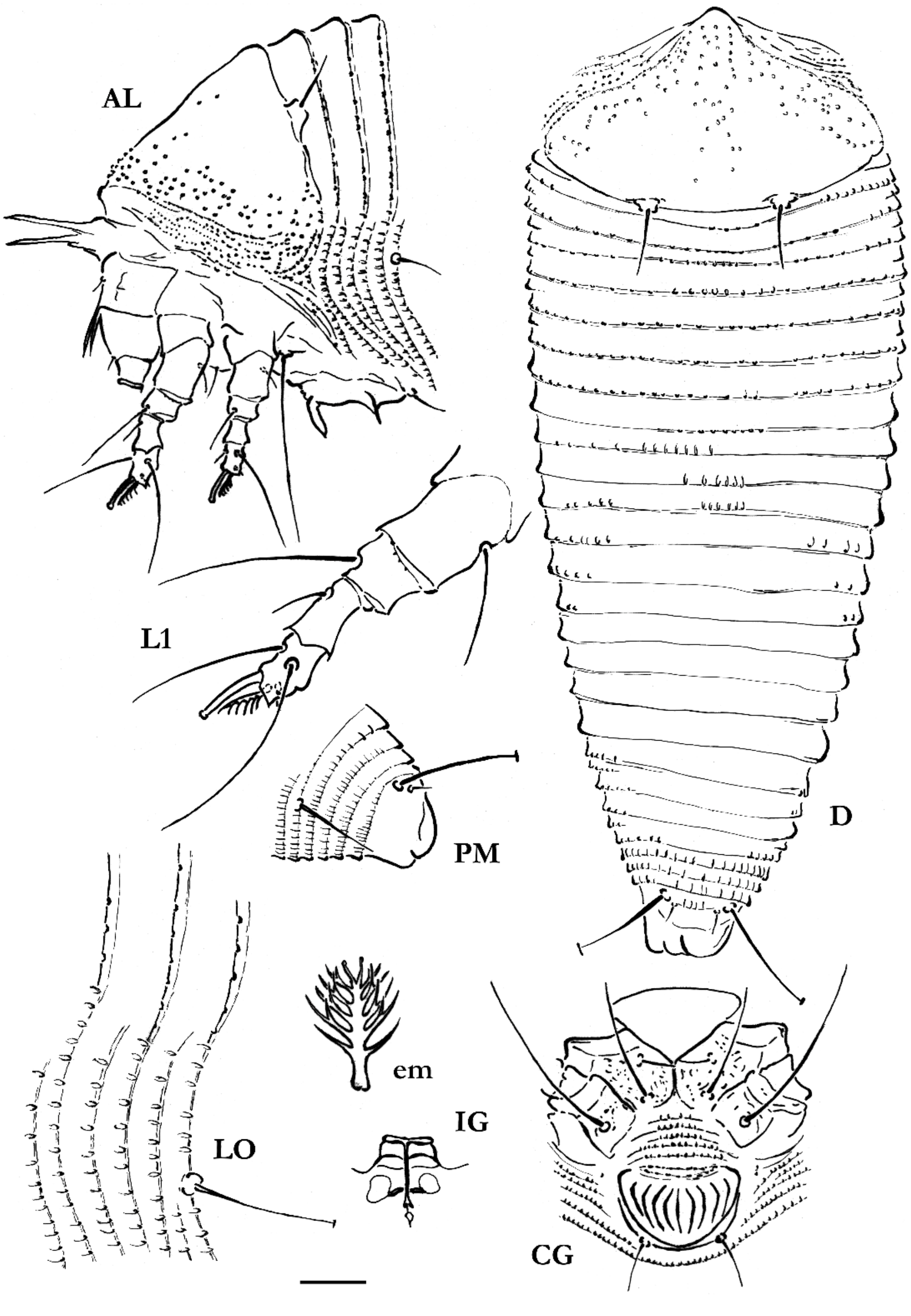

FIGURE 5.2. Line drawings of *Aculus pampae* **sp. nov.**: **D**. Dorsal view of an adult female; **AL**. Lateral view of anterior body region; **CG**. Female coxigenital region; **em**. Empodium; **IG**. Internal female genitalia; **LO**. Lateral view of annuli; **L1**. Leg I; **PM**. Lateral view of posterior opisthosoma. Scale bar: 10 µm for **D**, **AL**, **CG**, **GM**, **IG**, **PM**; 5 µm for **LO**, **L1**; 2.5 µm for **em**.

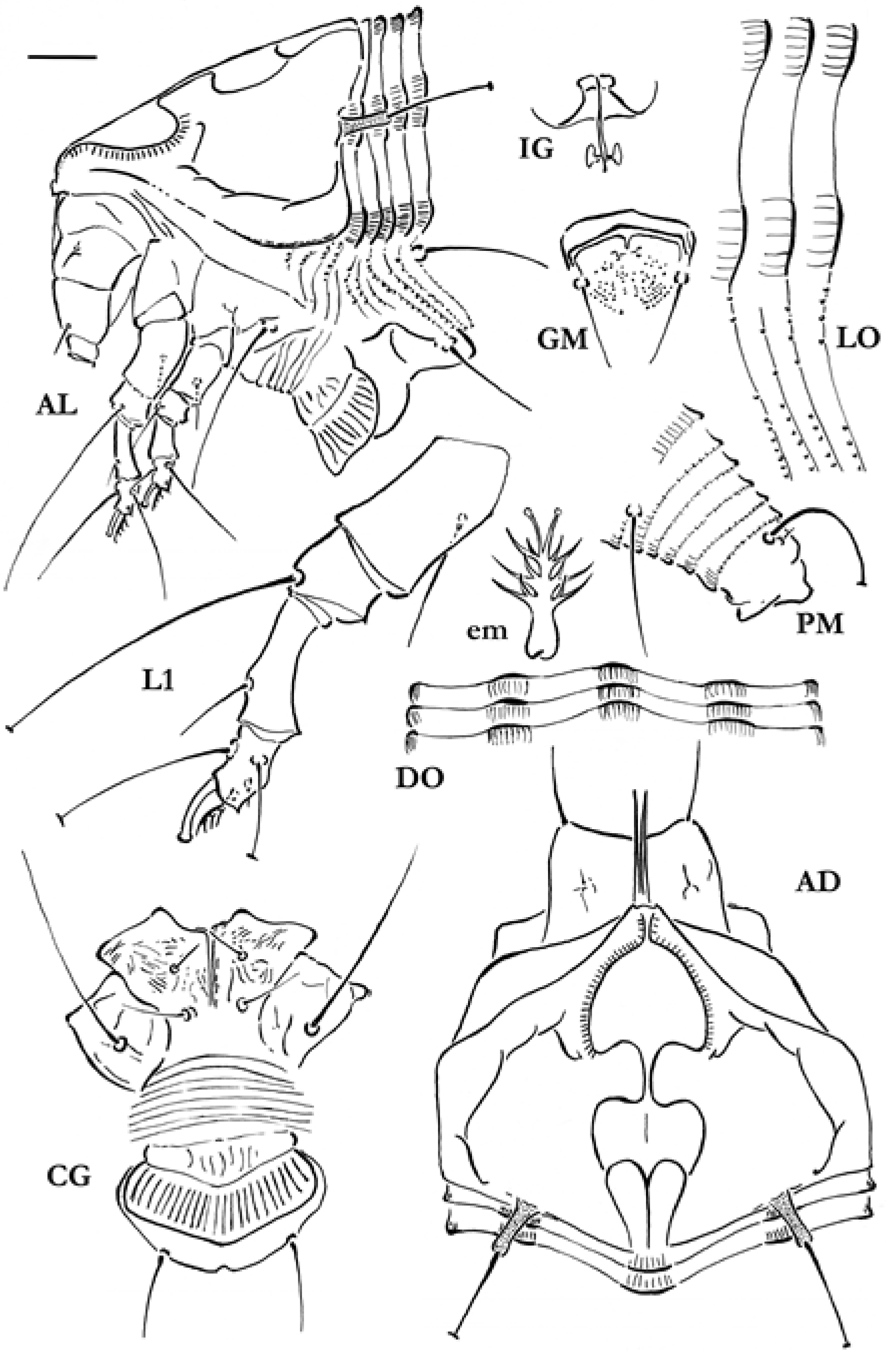

FIGURE 5.3. Line drawings of *Costarectus lanceatus* **p. nov.**: **AD**. Prodorsal shield; **AL**. Lateral view of anterior body region; **CG**. Female coxigenital region; **em**. Empodium; **GM**. male genitalia; **IG**. Internal female genitalia; **LO**. Lateral view of annuli; **L1**. Leg I; **PM**. Lateral view of posterior opisthosoma. Scale bar: 10 μm for **AD**, **AL**, **CG**, **GM**, **IG**, **PM**; 5 μm for **LO**, **L1**;

2.5 µm for **em**.

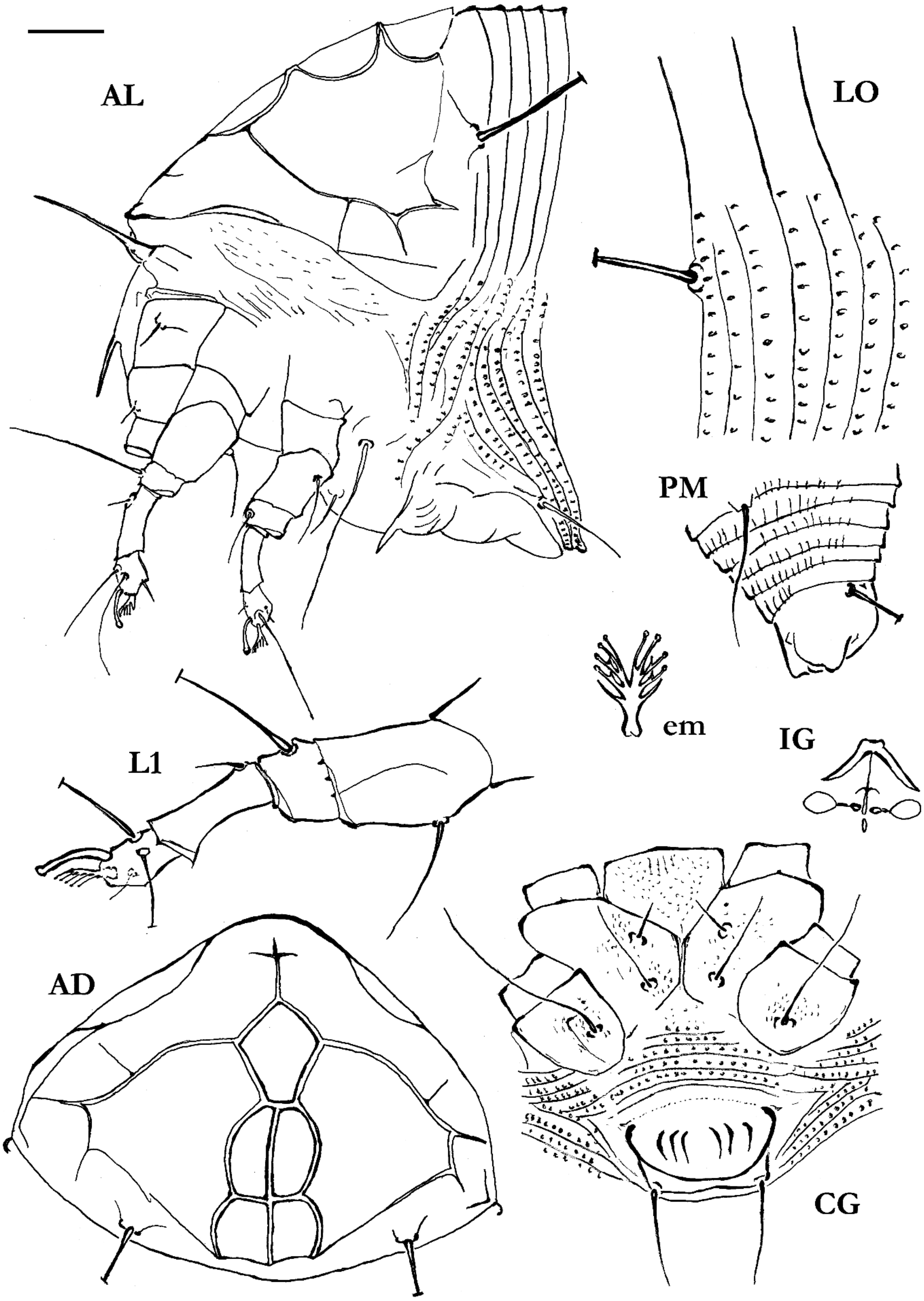

FIGURE 5.4. Line drawings of *Dichopelmus sigfeminae* **sp. nov.**; **AD**. Prodorsal shield; **AL**. Lateral view of anterior body region; **CG**. Female coxigenital region; **em**. Empodium; **IG**. Internal female genitalia; **LO**. Lateral view of annuli; **L1**. Leg I; **PM**. Lateral view of posterior opisthosoma. Scale bar; 10 µm for **AD**, **AL**, **CG**, **IG**, **PM**; 5 µm for **LO**, **L1**; 2.5 µm for **em**.

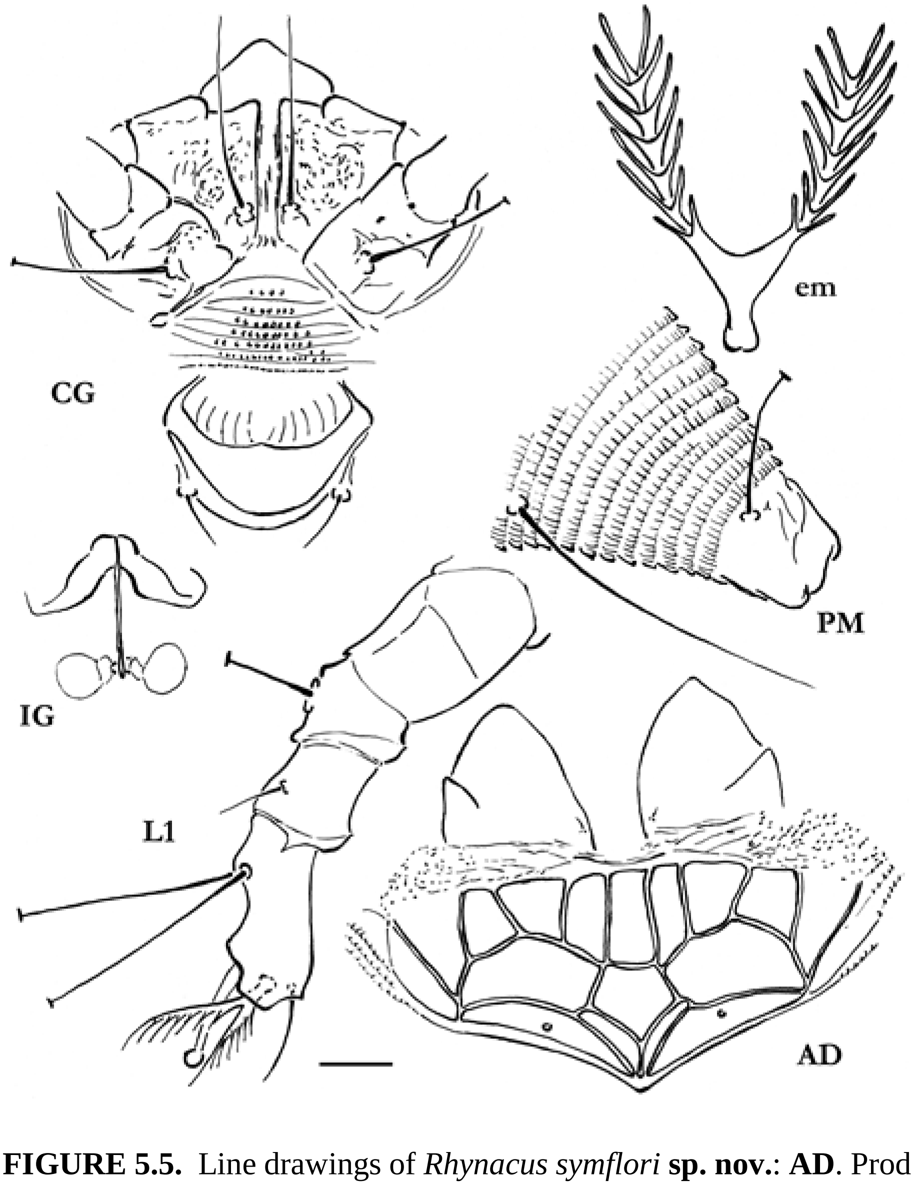

FIGURE 5.5. Line drawings of *Rhynacus symflori* **sp. nov.**: **AD**. Prodorsal shield; **CG**. Female coxigenital region; **em**. Empodium; **IG**. Internal female genitalia; **L1**. Leg I; **PM**. Lateral view of posterior opisthosoma. Scale bar: 10 µm for **AD**, **CG**, **IG**, **PM**; 5 µm for **L1**; 2.5 µm for **em**.

6 REDESCRIPRION OF *TETRANYCHUS ARMIPENIS* (TETRANYCHIDAE)

Abstract

Spider mite surveys conducted in Rio Grande do Sul state, Brazil, allowed us to provide the redescription of *Tetranychus armipenis* Flechtmann & Baker (Tetranychidae) from specimens collected on native plant species from Pampa biome. Also were reported for the first time *Atrichoproctus uncinatu*s Flechtmann and *Schizotetranychus oryzae* Rossi de Simons on a new host.

Keywords: *Atrichoproctus uncinatu*s; Pampa biome; spider mites; *Schizotetranychus oryzae*; Solanaceae.

6. 1 Introduction

In the south of Rio Grande do Sul State the Pampa biome stands out for presenting high levels of fauna and flora biodiversity and for having a unique richness, also presenting high rates of diversity and endemism (Behling *et al*. 2009; Bencke 2009). In this biome, the vegetation is characterized mainly by grasses in fields and some riparian forest (Carvalho *et al*. 2009; Behling *et al*. 2005; Roesch *et al*. 2009). Nowadays, a little part of the pampa biodiversity is known (Nabinger *et al*. 2006).

There are 112 species of Tetranychidae in Brazil, being *Tetranychus* the second more abundant genus, with 24 species (Migeon, Nouguier & Dorkeld 2011). The *Tetranychus* has some specific characteristics, like opisthonotum with 11 pairs of setae (c1-3, d1-2, e1-2, f1-2, h2-3; setae h3 ventral); setae f1 in dorsocentral position, dorsal opisthosoma with striae, two pairs of genital setae (g1-2), two pairs of pseudo anal setae (ps1-2); tarsus I with two pairs well separated duplex setae, tarsus II with one pair of duplex setae and empodium split distally with four tenent hairs (Baker & Tuttle 1994).

The original description of T. armipenis was in 1970 with only the design of the edego. So the objective of this paper is to report tetranychid species new host pllants records, to provide and redescription of *Tetranychus armipenis* Flechtmann & Baker.

6.2 Materials and methods

It was made a collect, with 324 plants samples, and leaves of *Sida rhombifolia* L., was collected to made the redescription. All leaves were placed in plastic bags, kept in Styrofoam box with Gelox® at low temperature until they arrived at the laboratory. All mites were collected under stereomicroscope using fine hair brush, mounted on glass slides in Hoyer's medium and kept in a kiln at a temperature between 50–60°C for approximately ten days before observations.

The identification of tetranychid mites was done with dichotomous keys and phase contrast microscope. Is was only consider new record when it was found more than one specimen on the plant species.

To the redescription and description, the morphological details were made with the help of an optical microscope with phase contrast. Drawings were made using a camera Lucida and Nankin. All measurements are in micrometers (µm).

Specimens collected of the new species were deposited at the Mite Reference Collection in the Museum of Sciences of Universidade do Vale do Taquari – Univates, Lajeado, Rio Grande do Sul, Brazil and Acarology and Entomology Department, Escola Superior de Agricultura "Luiz de Queiroz", Universidade de São Paulo (ESALQ/USP), Piracicaba, São Paulo state, Brazil.

6.3 Results

In the data collected, three new records were made to the world of Tetranychidae in native plant species: *Atrichoproctus uncinatu*s Flechtmann on *Desmodium incanum* DC. and *Desmodium adscendens* (Sw.) DC. (Fabaceae) and *Schizotetranychus oryzae* Rossi de Simons on *Piptochaetium montevidense* (Spreng.) (Poaceae). These records on native plants can support future work with these species of mites.

Systematics

Family Tetranychidae Donnadieu

Subfamily Tetranychinae Berlese

Tribe Tetranychini Reck

Genus *Tetranychus* Dufour, 1832:276; Pritchard & Baker, 1955:373; Wainstein, 1960: 149; Tuttle & Baker, 1968: 1244; Meyer 1974:216.

Tetranychus armipenis Flechtmann & Baker

Figures 6.1 – 6.16

General diagnosis – Peritreme hooked distally; female dorsal striae with a unique pattern and with hourglass pattern between setae *e1* and *f1*; female empodia split distally without mediodorsal spur; pre-genital striae solid; dorsal and ventral lobes on striation in varied shapes, slightly separated. Male aedeagus forked, curved upwards in a weakly sigmoid curve and the posterior margin has a prong-like extension; empodia with mediodorsal spur.

Redescription. Adult female.

Dorsum. (Figs 6.1 – 6.7) Idiosomal length 436 (390 - 488), width 364 (308 - 403). Dorsal striae with lacking lobes. Palpus spinneret 6 (6.5 – 6.7) long and 5 (6.4 – 6.6) wide. Peritreme hooked distally. Dorsal setae slightly barbed, longer than the distance between their longitudinal bases. Measurements of dorsal setae: *v2–h1* 376 (320 - 364), *sc2–sc2* 243 (225 - 262); Setal lengths: *v2* 69 (62 - 73), *sc1* 175 (168 - 179), *sc2* 113 (108 - 120), *c1* 161 (149 - 175), *c2* 155 (145 - 161), *c3* 139 (133 - 150), *d1* 149 (143 - 155), *d2* 149 (142 - 157), *e1* 144 (138 - 149), *e2* 143 (137 - 150), *f1* 116 (107 - 125), *f2* 96 (92 - 100), *h2* 60 (58 - 64), *h3* 55 (50 - 59). Distance between dorsal setae: *v2–v2* 66 (65 - 67), *sc1–sc1* 94 (90 - 98), *c1–c1* 63 (48 - 70), *c3–c3* 286 (253 - 356), *d1–d1* 93 (89 - 97), *d1–d2* 69 (63 - 73), *e1–e1* 46 (43 -50), *e1–e2* 78 (74 - 84), *f1–f1* 46 (40 - 54), *h2–h2* 26 (17 - 30); *c1–d1* 67 (49 - 84), *d1–e1* 56 (45 - 75); *e1–f1* 77 (67 - 84); *f1–h2* 104 (80 - 150). Hourglass striae pattern for the region between *e1–e1* and *f1– f1*.

Venter. (Fig 6.8 – 6.9) All venter setae thin, smooth. Anterogenital striae solid, entire and longitudinal. Measurements: *ag* 75 (70 - 81), *g1* 53 (50 - 60), *g2* 52 (45 - 58), *ps1* 24 (23 - 27), *ps2* 25 (24 - 26).

Legs. (Figs 6.10 – 6.11) Empodia split distally into 3 pairs of hairs, with four tenent hairs. All the segments of the legs with striae. Number of setae on legs (from coxae to tarsi, solenidia in parentheses) as follows:

Leg I - 2 - 1 - 10 - 5 - 9 (1) - 13 (1) + 2 duplex setae
Leg II - 2 - 1 - 6 - 5 - 7 - 13 (1) + 1 duplex setae
Leg III - 1 - 1 - 4 - 4 - 6 - 9 (1)
Leg IV - 1 - 1 - 4 - 4 - 7 - 10 (1).

Redescription. Adult male.

Dorsum and venter. Length of idiosoma 333 (300 - 375), width 237 (218 - 265). Setae similar to female.

Palpus. (Fig 6.12) Palpus with spinneret 5 long and 3 wide.

Legs. (Figs 6.13 – 6.14). Empodium split distally into with mediodorsal spur. All the segments of the legs with transverse striae. Number of setae on legs (from coxae to tarsi, solenidia in parentheses) as follows:

Leg I - 2 - 1 - 10 - 5 - 9 (4) - 12 (3) + 2 duplex setae
Leg II - 2 - 1 - 6 - 5 - 6 (1) - 13 (1) + 1 duplex setae
Leg III - 1 - 1 - 4 - 4 - 6 - 9 (1)
Leg IV - 1 - 1 - 4 - 4 - 7 - 10 (1).

Aedeagus. (Figs 15 - 16). Unique, distinctive, curved upwards at a right angle and forked. Distal knob of upturned part with anterior projection minute, acute, posterior one well developed, acute and slightly bent downward distally, forming an angle about 45° with shaft axis.

Family Tetranychidae Donnadieu
Subfamily Tetranychinae Berlese
Tribe Eurytetranychini Reck
Genre *Aponychus* Rimando, 1966: 105, Tuttle & Baker, 1968:82; Meyer, 1974:156.

Figure captions

Figure 6.1: *Tetranychus armipenis* female. Idiosoma in dorsal view.

Figure 6.2: *Tetranychus armipenis* female. Phase contrast microscope. Idiosoma striae pattern, from setae *v2* to *d2*.

Figure 6.3: *Tetranychus armipenis* female. Phase contrast microscope. Idiosoma striae pattern, from setae *c1* to *f2*.

Figure 6.4: *Tetranychus armipenis* female. Phase contrast microscope. Dorsal lobes.

Figure 6.5: *Tetranychus armipenis* female. Palpus.

Figure 6.6: *Tetranychus armipenis* female. Peritreme.

Figure 6.7: *Tetranychus armipenis* female. Phase contrast microscope. Hourglass patterns, between setae *e1* and *f1*.

Figure 6.8: *Tetranychus armipenis* female. Phase contrast microscope. Anogenital region, in ventral view.

Figure 6.9: *Tetranychus armipenis* female. Phase contrast microscope. Pre-genital striae.

Figure 6.10: *Tetranychus armipenis* female. A - tibia and tarsus of leg I; B - tibia and tarsus of leg II

Figure 6.11: *Tetranychus armipenis* female. A - empodium I; B - empodium II; C - empodium III; D - empodium IV.

Figure 6.12: *Tetranychus armipenis* male. Palpus.

Figure 6.13: *Tetranychus armipenis* male. A - tibia and tarsus of leg I; B - tibia and tarsus of leg II.

Figure 6.14: *Tetranychus armipenis* male. A - empodium I; B - empodium II; C - empodium III; D - empodium IV.

Figure 6.15: *Tetranychus armipenis* male. Aedeagus.

Figure 6.16: *Tetranychus armipenis* male. Phase contrast microscope. Comparison between two aedeagus.

FIGURE 6.1

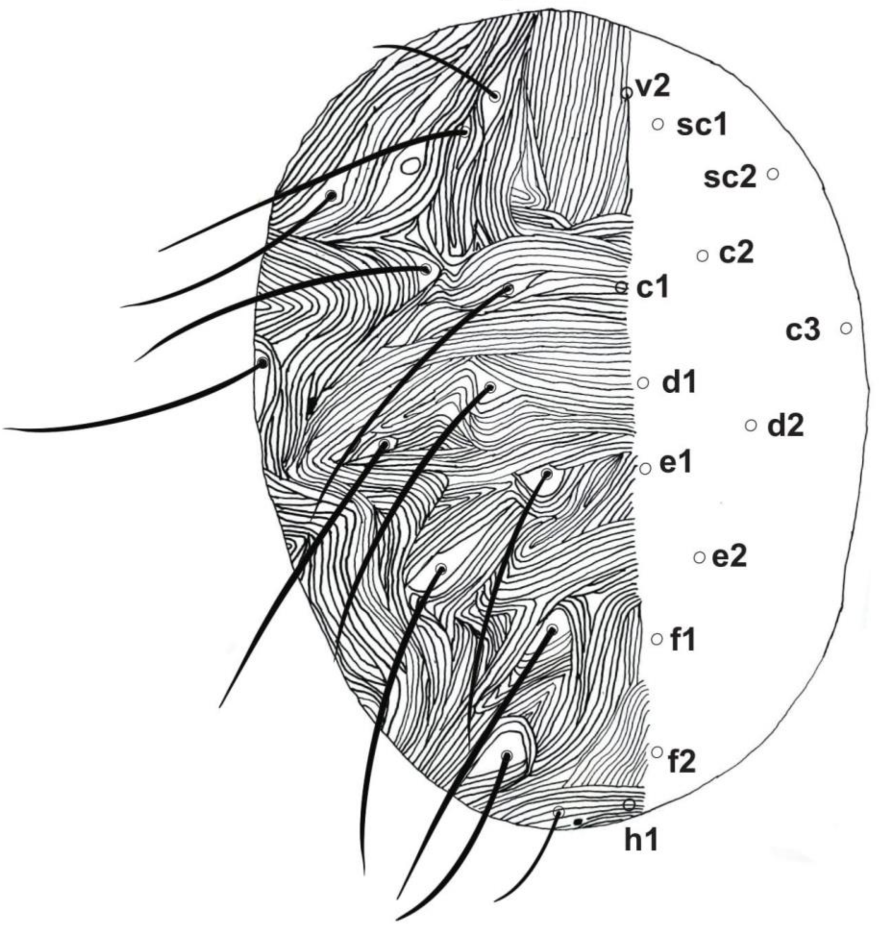

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FIGURE 6.2

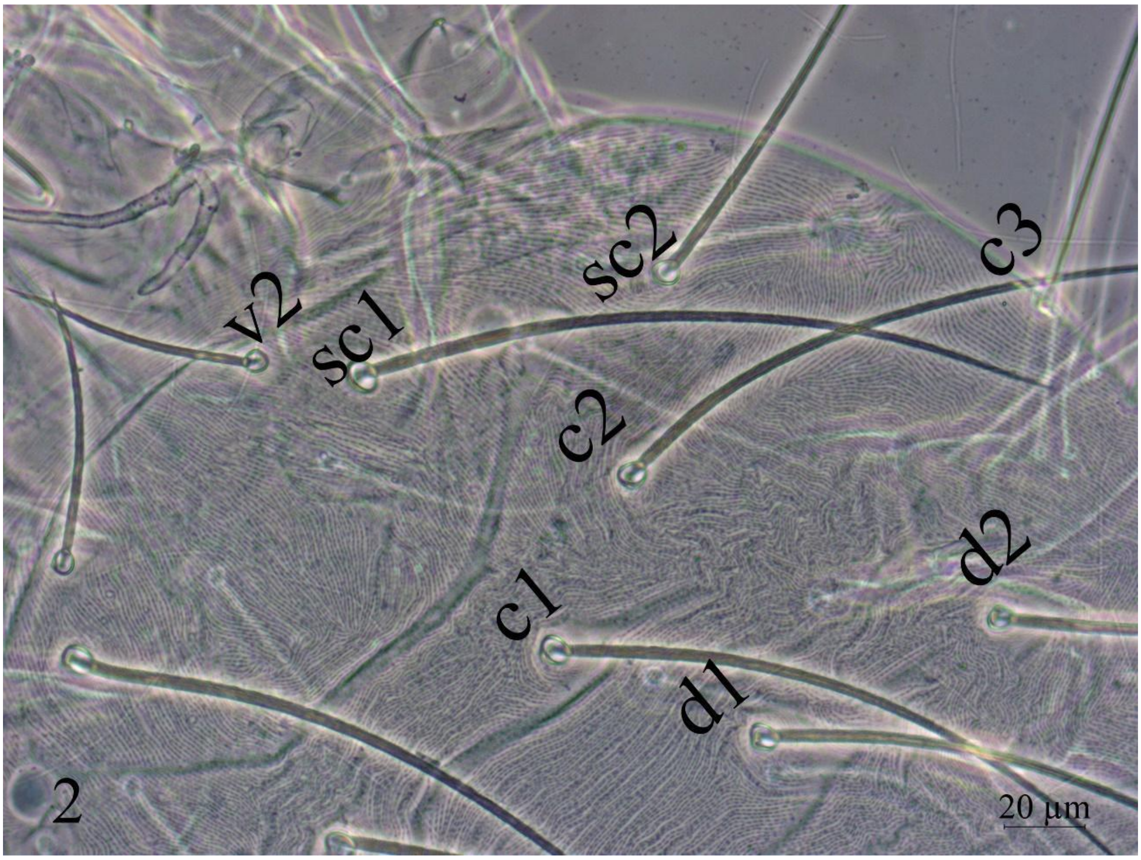

FIGURE 6.3

FIGURE 6.4

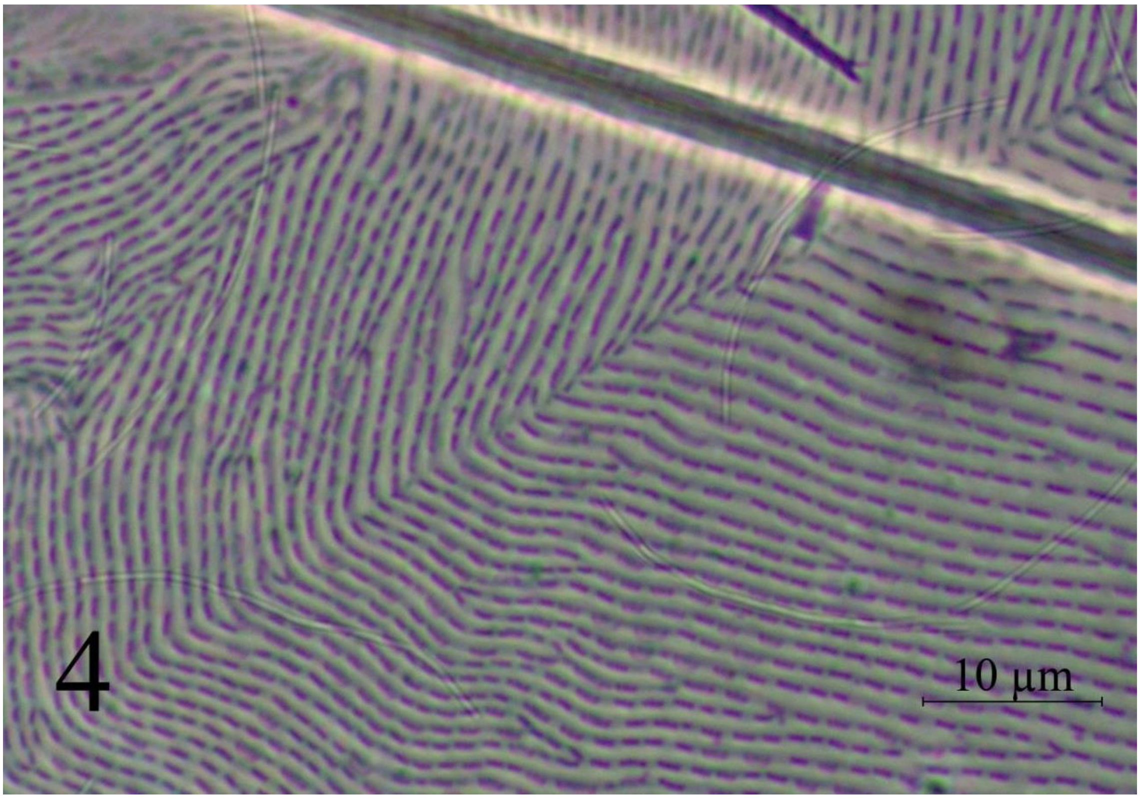

FIGURE 6.5

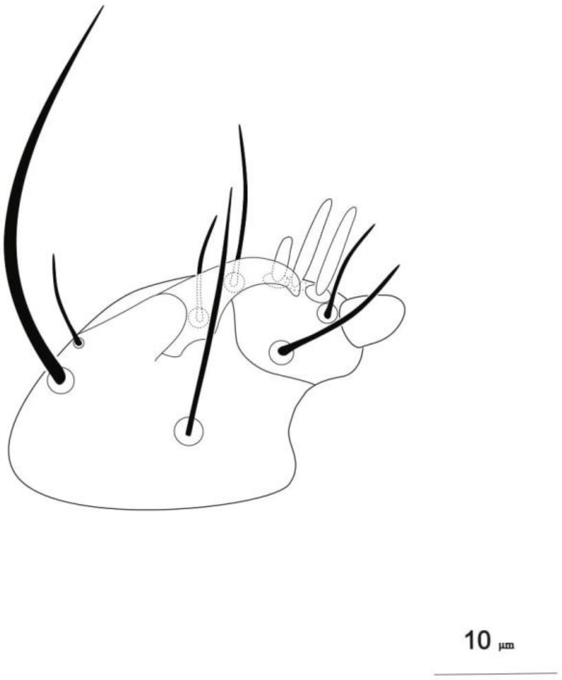

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FIGURE 6.6

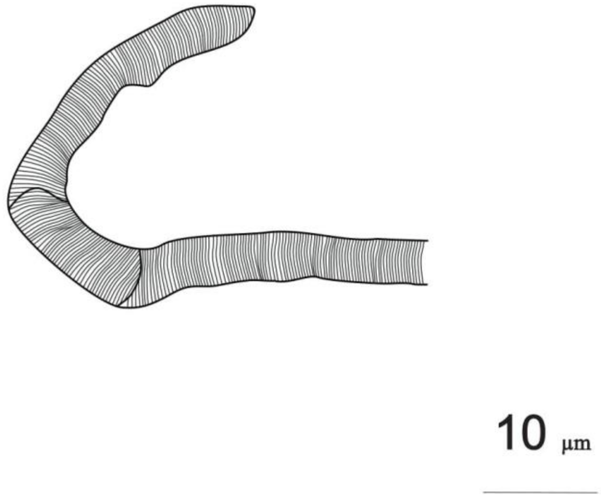

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FIGURE 6.7

FIGURE 6.8

FIGURE 6.9

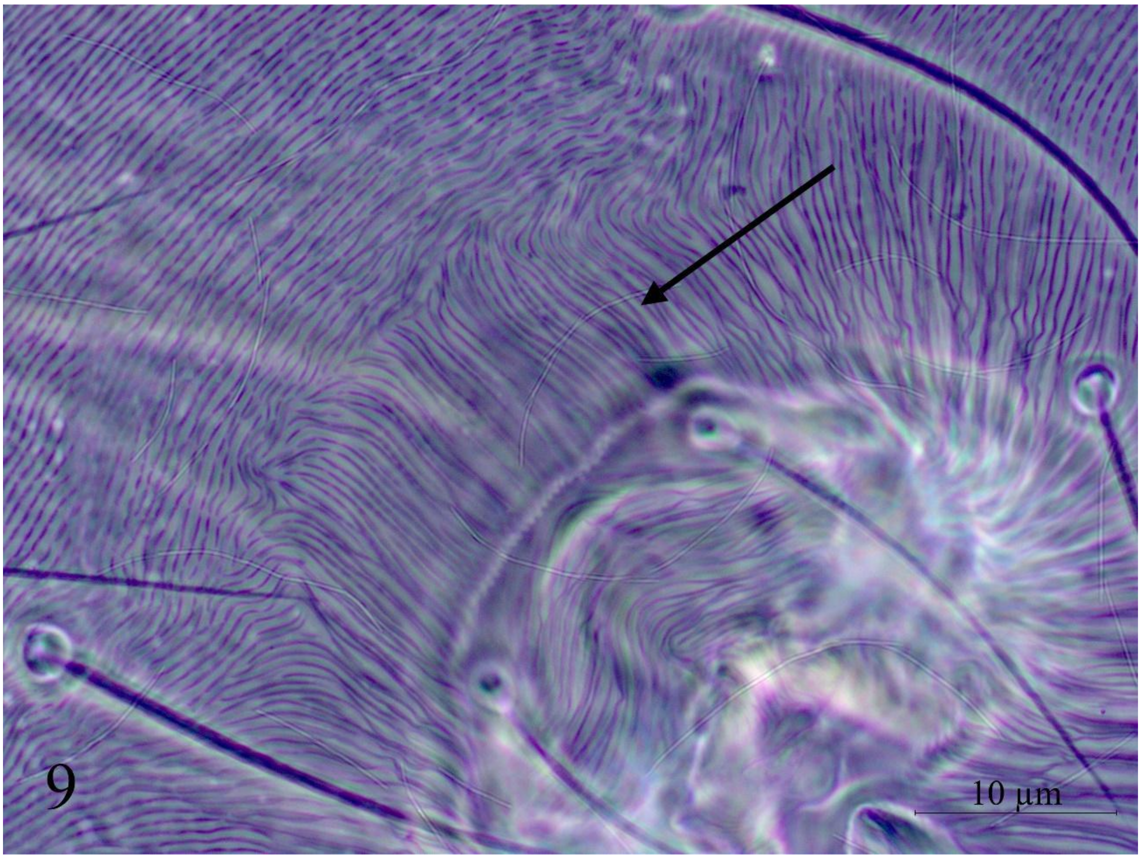

FIGURE 6.10

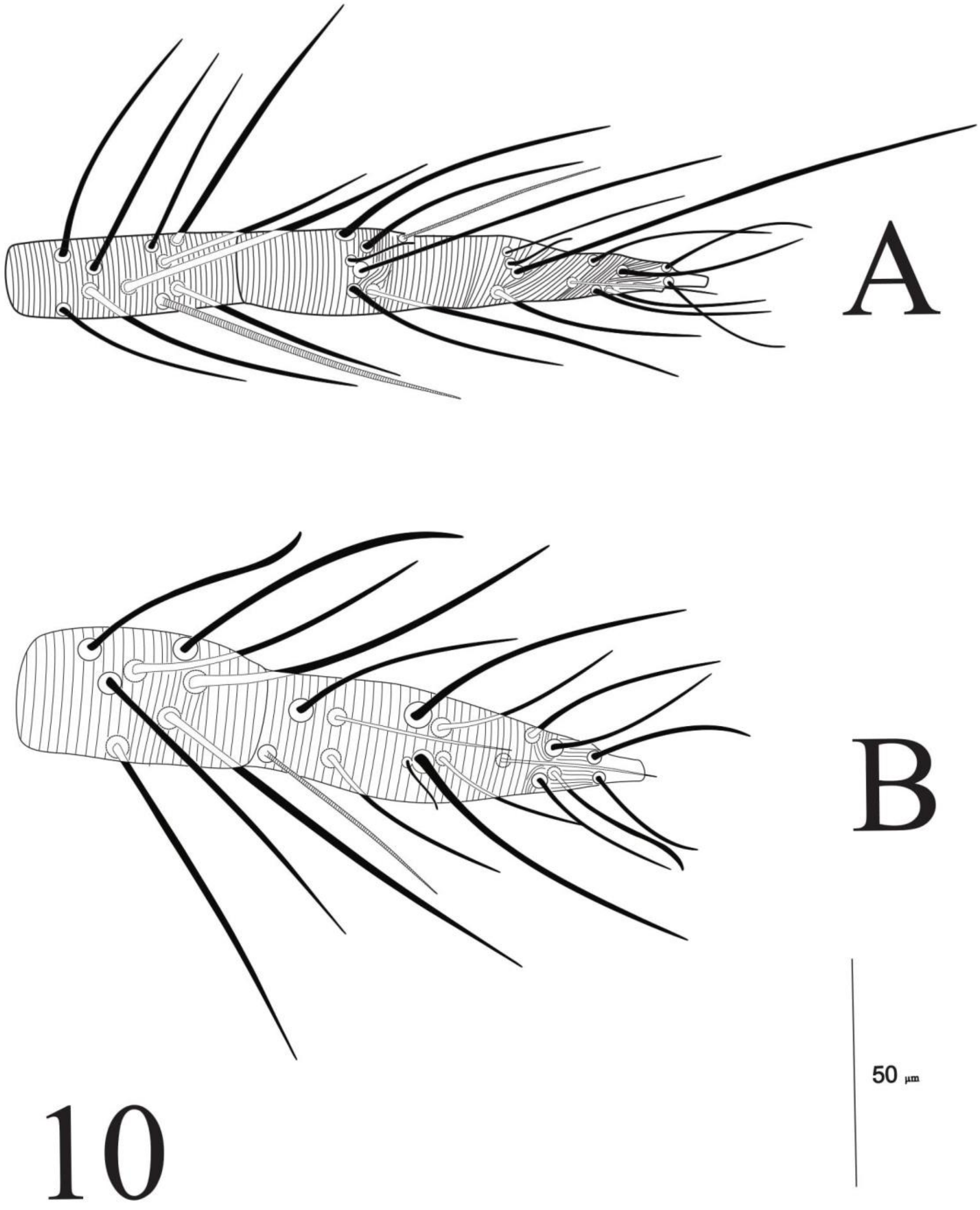

FIGURE 6.11

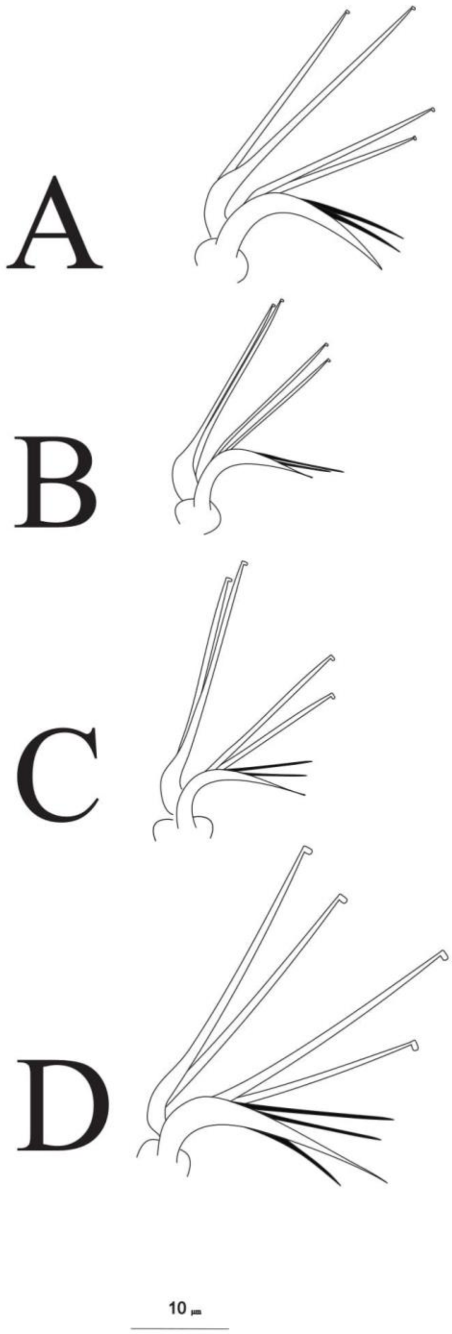

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FIGURE 6.12

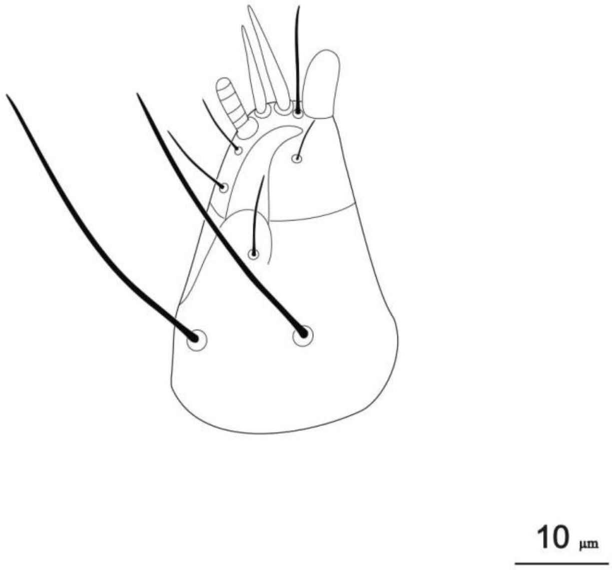

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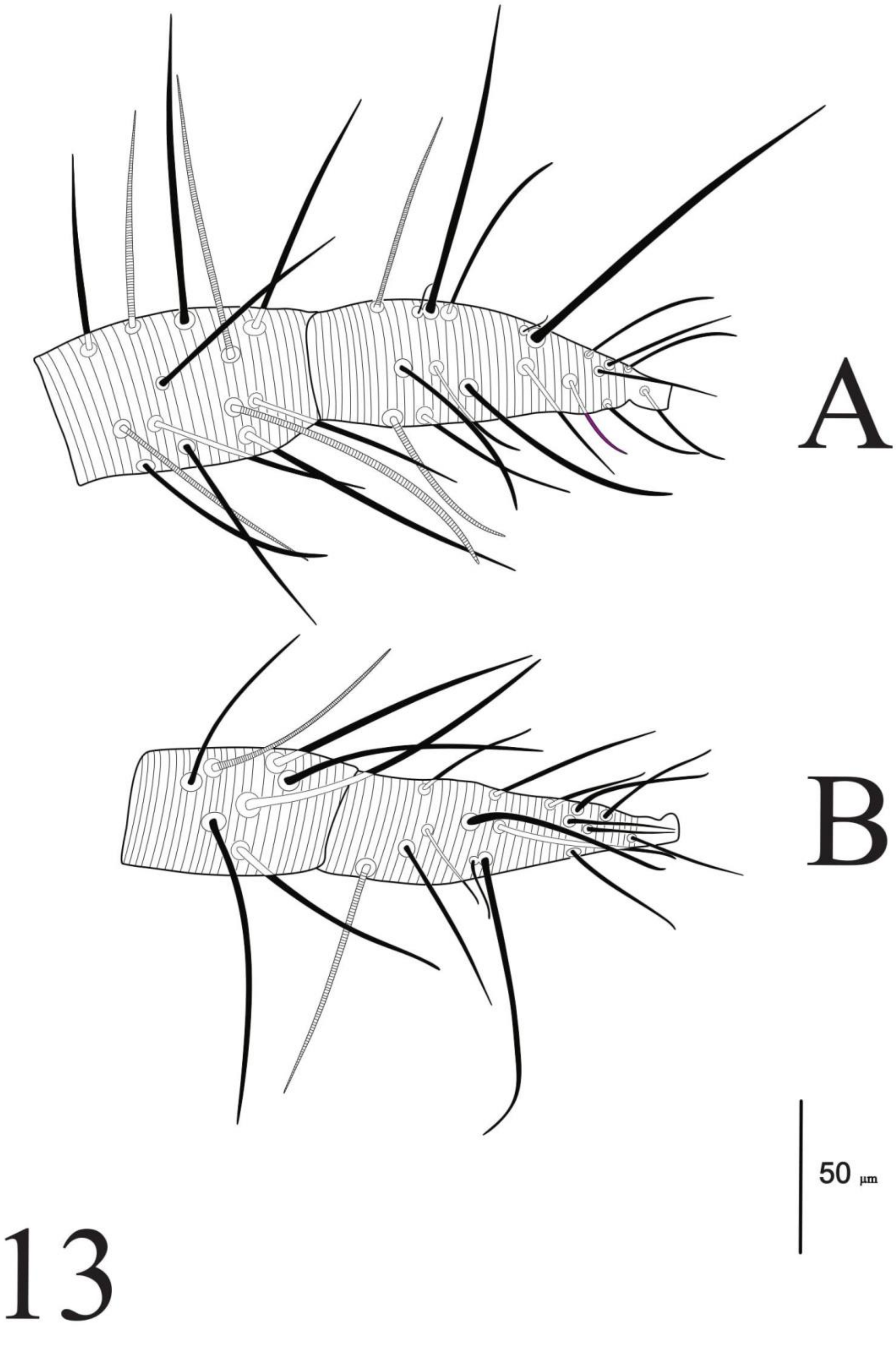
FIGURE 6.13



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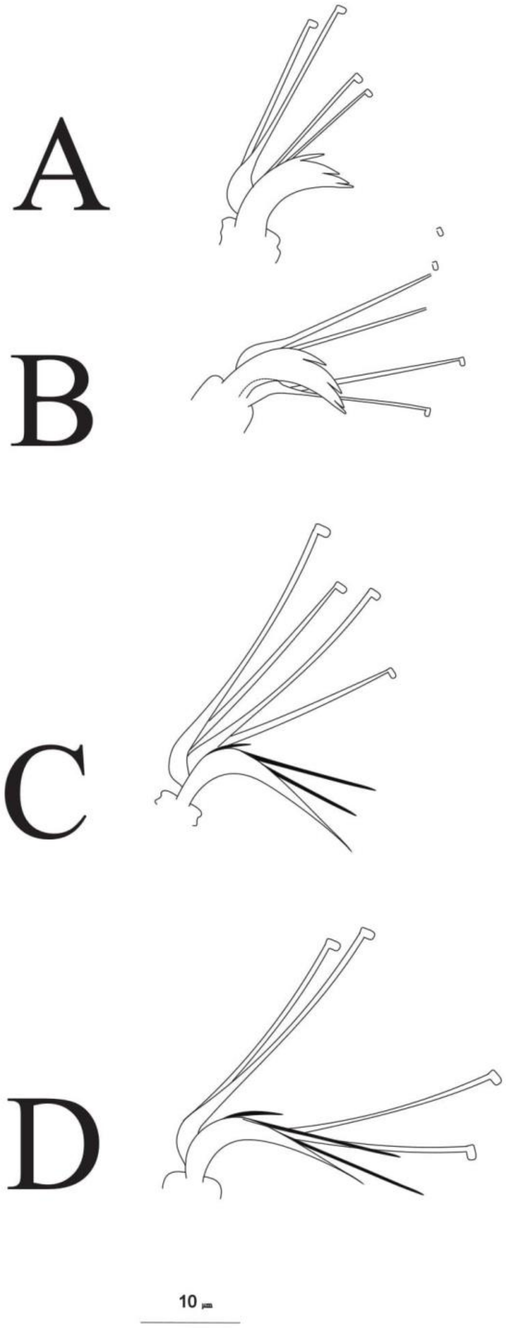

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FIGURE 6.15

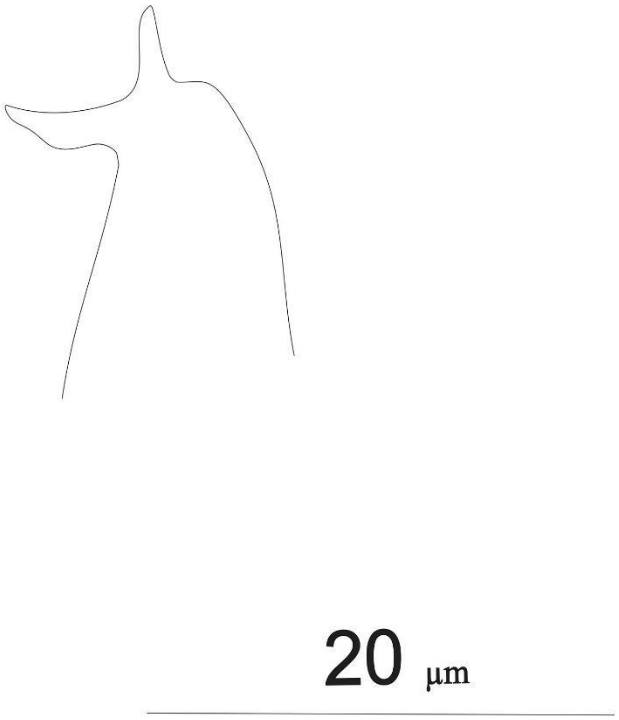

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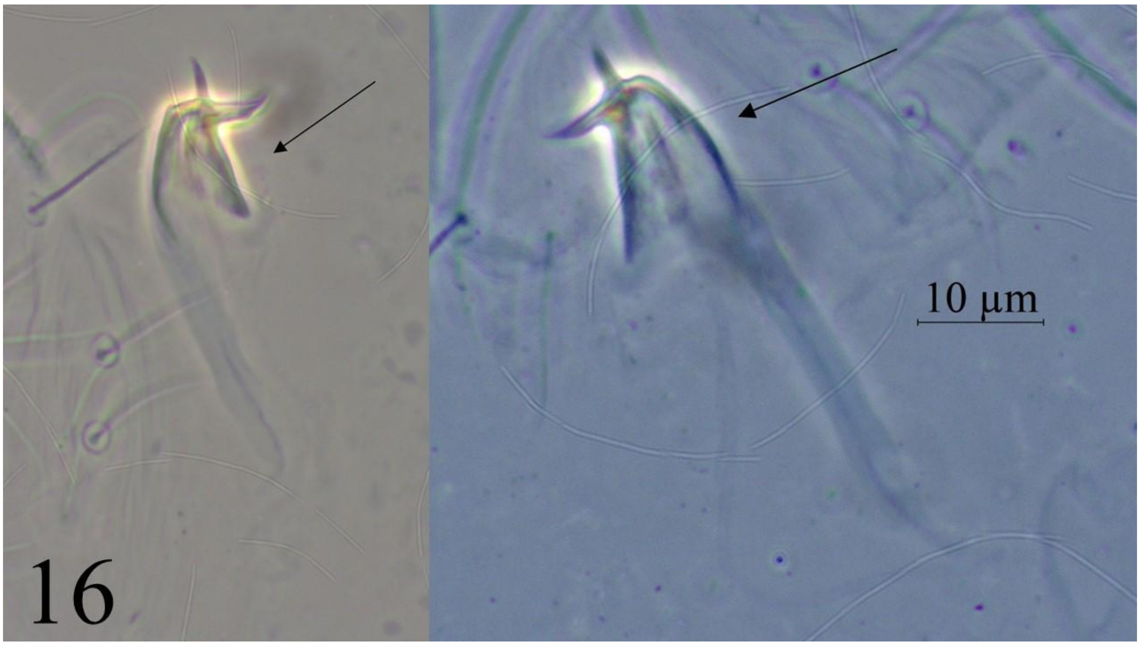
FIGURE 6.16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10 µm

7 COULD PHYTOSEIIDS MITES IMPAIR BIOLOGICAL CONTROL OF INVASIVE PLANT *Ailanthus altissima?*

Abstract: *Aculus mosoniensis* Ripka (Eriophyidae) is a potencial biologic control agent of *Ailanthus altissima* (Mill.) Swingle (Simaroubaceae). However, predatory mites present in Europa may hind the action of this eriophyids agent. This study had the aim of evaluate the biology of two predatory mites, *Euseius stipulatus* Denmark & Muma and *Typhlodromus* (*Typhlodromus*) *exhilaratus* Ragusa (Phytoseiidae) feeding on *A. mosoniensis*. The rearing stock were kept in germinating chambers at 25±1°C, photophase 12 hours and relative humidity of 70±5%. The data collected were compared through ANOVA test, at 5% of significance level through *R* program. From 40 eggs individualized of *T.* (*T.*) *exhilaratus*, only 42.5% achieved the adulthood, while *E. stipulatus* had viability of 80.5%. *Euseius stipulatus* showed a quick development on the immature stages, the average fecundity was higher to *E. stipulatus* (18 eggs/female) than to *T.* (*T.*) *exhilaratus* (3.5 eggs/ female). Both predatory mites evaluated in this study feed on *A. mosoniensis*. However, *E. stipulatus* seems to be an efficient predator capable to control *A. mosoniensis*. Thus, *E. stipulatus* may hinder the biological control of *A. altissima* in the field.

Key-words: *Aculus mosoniensis;* Biological control; *Euseius stipulatus; Typhlodromus* (*Typhlodromus*) *exhilaratus*.

7.1 Introduction

Ailanthus altissima (Mill.) Swingle (Simaroubaceae), native from China and North Vietnam, has world occurrence being invasive and dangerous to native species (Kowarik & Säumel 2007). With quick growing, invades natural areas establishing monospecific population of high density shading native species. It presents root allelopathy precluding growing and the developing of other plants species that occupy the same environment (Heisey 1996), becoming invasive in Europe and in all continents, excepting Antarctica (Kowarik & Säumel 2007). With the aim to decrease the environmental impact, in the last years, the practice of introduction herbivorous and pathogens to ecosystem management has been intensified controlling the density of the invasive plants. So, the biological control aims to reduce the population of some species with ecological or economic importance (Seastedt, 2015).

Aculus mosoniensis Ripka (Eriophyidae), described recently from specimens collected on plants in Hungary (Ripka & Érsek 2014), reported in Italy on *A. altissima*, could be used like a natural enemy of this exotic plant (de Lillo *et al.* 2017). The Eriophyoidea, in general, cause damage to countless cultures of agriculture, foresting and ornamental interesting (Lindquist *et al.* 1996). However, the severity of the symptoms depends on mite population density and the morphology of the plant attached (Oldfield 1996; Smith *et al.* 2010), reducing the biomass production and also mitigating the reproductive performance of the host plant (de Lillo & Skoracka 2010). About 80% of Eriophyoidea are found exclusively associated to unique host plant species (Skoracka *et al.* 2009), being thus excellent candidate to controlling invasive plants (SMITH *et al.* 2010). The monophagy is a strong point to Eriophyoidea in the controlling of invasive plants, guaranteeing the protection of the native plants in places of introduction.

Associated to eriophyids mites may be predatory mites, being the phytoseiids the most commons and abundant on plants (Tixier 2018). Various species are important control agents in the applied biological control in green houses (Zhang, 2003) and orchards (Parra *et al.* 2002). They feed on mites, insects, nematodes, fungi, pollen and plan exudates (McMurtry *et al.* 2013). *Amblyseius swirskii* (Phytoseiidae) Athias-Henriot, for example, controls *Aculops lycopersici* (Tryon) (Eriophyidae) on tomato plants (Park *et al.* 2010). *Typhlodromus* (*T.*) *exhilaratus* Ragusa (Phytoseiidae) is reported like generalist predator of type III (McMurtry *et al.* 2013), feeding on tetranychids, eriophyids and pollen (Ragusa 1981). *Typhlodromus* (*T.*) *exhilaratus* was already seen associated to *Aceria caulobius* (Eriophyidae) (Nalepa) inner the galls (de Lillo 1987; de Lillo & Monfreda 2004).

Mites of the genre *Euseius* are found commonly associated to plants with smooth leaves surface or little pubescent (Seelmann *et al.* 2007). They are reported on plants of eucalyptus, coffee plants and forest fragments (de Queiroz & Flechtmann 2011) and also associated to grapevines (Tixier *et al.* 2013). *Euseius stipulatus* (Athias-Henriot) (Phytoseiidae), is a generalist species, capable to develop using pollen as alternatives food (McMurtry *et al.* 2013). This species was already reported to Italy (Ragusa 1976), Spain (Ferragut *et al.* 1988) and Portugal (Silva *et* *al.* 2019) in environments with temperate climate, where develops fundamental level control of *Panonychus ulmi* (Koch) (Rodrigues 2005) (Tetranychidae). However, until nowadays nothing was found about predatory mites controlling *A. mosoniensis*.

However, there is a growing interest in the use of mites for biological weed control, especially those belonging to Eriophyoidea because they are host specific, weakening the host plant affecting the growing and the reproduction (Vásquez *et al.* 2015). Like this, the use of *A. mosoniensis* may help to control the infestation of this invasive plant (*A. altissima*) and to reduce their propagation. In this study we tested the hypothesis that the predatory mites found on European natural environment could reduce the biological control level of this plant. Thus, this assay had the aim to evaluate the predation capacity of *E. stipulatus* and *T.* (*T.*) *exhilaratus* feeding on *A. mosoniensis*.

7.2 Material and methods

The experiments were carried out in the acarology lab of Universitá degli Study di Bari Aldo Moro, Bari, Italy. *Euseius stipulatus* and *A. mosoniensis* were collected on *A. altissima* in the city of Bari, Italy (41°06'33.3"N 16°53'04.3"E), while *Typhlodromus exhilaratus* was collected on *Suaeda fruticosa* Forsk. (Amaranthaceae) in the city of Margherita di Savóia (41°22'23.5"N 16°07'38.6"E) (de Lillo 2004) were it was associated to *Aceria caulobia* Nalepa (Eriophyidae) population in galls. The rearing stock of the predatory mites were kept in the plants of *A. altissima* contaminated with populations of *A. mosoniensis* and *A. caulobia*. The rearing stock were kept at lab throughout the period in climatic chamber at 25±1°C, photophase 12 hours and 70±5% relative humidity.

The predatory biology was started from 40 eggs of each species, isolated in arenas made on Petri dish of 6cm of diameter with leaf circles contaminated with *A. mosoniensis* and attached on a pin and surrounded by distilled water. Three diary observations were made, at 8, 13 and 18 hours, during the immature phases. In the adulthood, females were mated with males obtained in the rearing stock and the evaluation were made once a day at 13 horas verifying the number of eggs laid and the survival. The eggs were collected and transferred to other arenas to determine the sexual ratio.

The data were compared through ANOVA test, Tukey *post-hoc* at level of 5% significance using the program *R* Studio (R Development Core Team 2010). The life table calculations were made according to Silveira *et al.* (1976). The values for the net reproductive rate (*Ro*=Σmx.lx-mx: total number of eggs/females; *lx:* specimens alive/total specimens), average generation time (*T*=mx.lx.x/mx.lx Σ), innate ability to increase (*rm*=log Ro/T.0.4343) and finite growth rate (*λ*= Antilog de *rm*) were calculated.

7.3 Results

Only 42.5% of *T.* (*T.*) *exhilaratus* achieved the adulthood phase, while 80.5% to *E. stipulatus* (Table 7.1). *Euseius stipulatus* showed, in most phases, developing shorter than *T. exhilaratus*, excepting larval phase. The period of egg-adult of *E. stipulatus* lasted in average 5.64 days, while the deutonymphal phase was faster. For *T. exhilaratus* the period of egg-adult was 7.66 days and the faster phase was larvae.

Table 7.1. Mean duration, in days (±EP), of the immature phases of the *Euseius stipulatus* and *Typhlodromus exhilaratus* feeding on *Aculus mosoniensis*, at 25±1°C, photophase of 12 hours and relative humidity of 70±5%.

	Immature stages				
	Egg	Larval	Protonymph	Deutonymph	Egg-to-adult
Euseius stipulatus	1.69±0.32Ba*	1.84±0.33Aa	1.4±0.67Bab	1.17±0.57Bb	5.64±1.05B
Viability (%)	**100**	**97.50**	**97.44**	**89.47**	**80.49**
Typhlodromus (*T.*) *exhilaratus*	2.75±0.90Aa	1.26±0.44Bc	2.75±1.24Aab	1.71±1.01Abc	7.66±1.22A
Viability (%)	**77.50**	**100**	**77.42**	**70.83**	**42.50**

*Same lowercase letters do not differ in row; same uppercase letters do not differ significantly in column in Tuckey test at 5% level.

The average fecundity was higher to *E. stipulatus* than to *T.* (*T.*) *exhilaratus* (Table 7.2). The results demonstrated that *E. stipulatus* is the predatory mite most efficient on *A. mosoniensis,* because achieves the adulthood and lay eggs. Differently, to *T.* (*T.*) *exhilaratus,* the number of females laying eggs and the amount of eggs were lower suggesting *A. mosoniensis* could be alternative food.

Table 7.2: Mean duration, in days (± SE), of immature stage, pre-oviposition, oviposition, longevity and average fecundity and egg per female (± EP) of *Typhlodromus* (*Typhlodromus*) *exhilaratus* and *Euseius stipulatus* feeding on *Aculus mosoniensis* at 25±1°C, 12 hours of photophase and 70 ± 5% relative humidity.

	N	*Typhlodromus exhilaratus*	N	*Euseius stipulatus*
Pre-oviposition	4	4.25±0.63a	18	2.17±0.20b
Oviposition	4	5.25±2.95b	18	10.56±1.75a
Post-oviposition	1	1	5	2.8 ±1.1
Longevity	15	6.93±1.36b	35	14.23±1.33a
Fecundity	4	3.50±1.19b	18	18±2.77a

N = number of mites evaluated; Mean (± SD) followed by the same letter in the line does not differ statistically from each other by the Tukey test, at a significance level of 5%.

The average durations of each generation (T) to *T.* (*T.*) *exhilaratus* was 18.68 days and 14.97 to *E. stipulatus.* The net reproduction rate (R_o) was higher to *Euseius stipulatus* (7.10) than with *T. exhilaratus* (0.18). *Euseius stipulatus* also had greater innate ability to increase in number (0.13) than *T. exhilaratus* (-0.09). This negative index indicates this predator's inefficiency in feeding only on its prey. The finite growth rate (λ) was 1.35 to *E. stipulatus* and 0.81 to *T.* (*T.*) *exhilaratus.* The specific fertility was higher at 17º day after the beginning of the biology to *T. exhilaratus* and between 10º and 22º days to *E. stipulatus* (Figure 7.1).

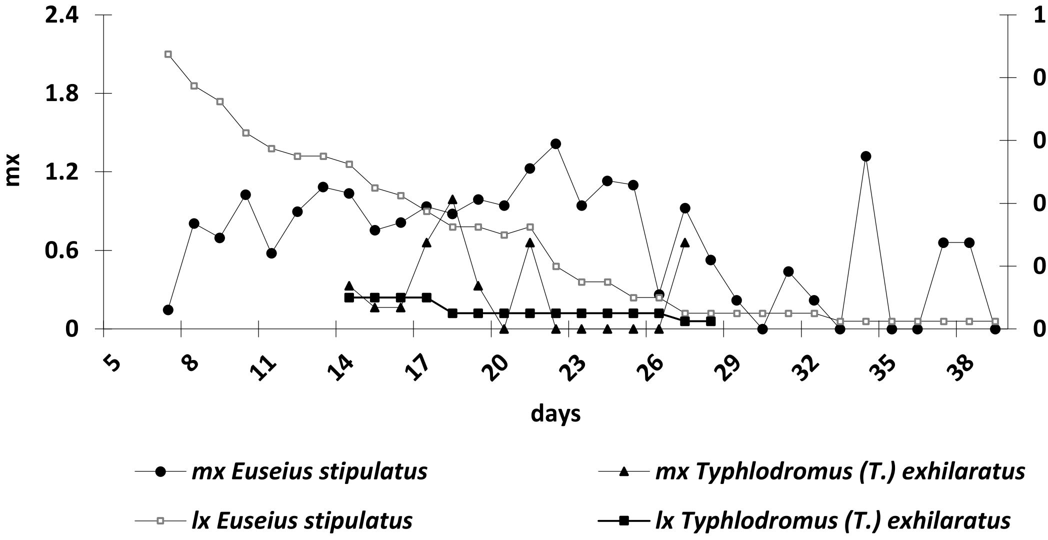

Figure 7.1: Specific fertility (mx) and survival of *T.* (*T.*) *exhilaratus* and *Euseius stipulatus* feeding on *Aculus mosoniensis* at 25 ±1°C, photophase 12 hours and relative humidity 70±5%.

7.4 Discussion

In a possible proposition of biological control of *A. altissima* in Europe with the use of *A. mosoniensis* it will be necessary to consider native predators when executing the program. Our study showed that both *E. stipulatus* and *T.* (*T.*) *exhilaratus* control *A. mosoniensis* suggesting therefore may interfere with a possible control program at this plant in southern Italy. The both species fed on *A. mosoniensis*, however *E. stipulatus* seems to be most efficiently predator of this prey. Populations of *A. mosoniensis* are insufficient to keep the population increasing of *T.* (*T.*) *exhilaratus*

The development time of *E. stipulatus* feeding on *A. mosoniensis* was similar to of the predator fed with *Carpobrotus edulis* (L.) N.E. Br. (Aizoaceae) pollen (Ferragut *et al*. 1987) when the egg-adult period was 5.5 days. However, when feeding on apple, almond, pear, apricot, plum, walnut and cherry pollen it was bigger (Bouras & Papadoulis, 2005). The same happens when fed with *Aleurothrixus floccosus* Maskell (Aleyrodidae), *Panonychus citri* McGregor, *Tetranychus urticae* (Koch) (Tetranychidae), *Lorryia formosa* Cooreman (Tydeidae), *Planococcus citri* Risso (Pseudococcidae) (Ferragut *et al*. 1987).

Until this moment exists just one study about the biology of *T. exhilaratus*, (Ragusa, 1981) where the egg-adult period was similar to our study even feeding on the pollen of *Borago officinalis* (L.) (Boraginaceae), *Rosmarinus officinalis* (L.) (Lamiaceae), *Bougainvillea* sp. (Nyctaginaceae), *Jasminum* sp. (Oleaceae), *Oxalis sp* (Oxalidaceae), *Duranta ellisia* Jacq. (Verbenaceae), the preys *P. citri* (Tetranychidae) and *T. urticae* (Tetranychidae).

The intrinsic growth rate of *E. stipulatus* was higher, 0.19, feeding of *Carpobrotus edulis* pollen (Ferragut 1987) indicating to be more efficient if compared to *A. mosoniensis* like prey. Also, the oviposition period and the fecundity were higher.

It was expected that the predator *T.* (*T.) exhilaratus* was efficient in the biological control of *A. mosoniensis*. However, the results obtained don't corroborated with our hypothesis. For Ragusa (1981) the diet of this predator includes tetranychids, eriophyids and pollen.

Euseius stipulatus has the biological control potential increased in environment wetter and temperatures milder (Thurman *et al*. 2017). It is considered tolerant to pesticides use if compared to other species normally found in the same environment, such as *Neoseiulus californicus* (McGregor) and *Phytoseiulus persimilis* (Athias-Henriot) (Argolo *et al.* 2014). Nonetheless, *E. stipulatus* was found only in the environment conserved (Silva *et al*. 2019).

Several studies provide information on the occurrence of predatory mites in plants. However, little is known about the parameters that explain this occurrence (Tixer 2018). The biological control of *A. altissima* with *A. mosoniensis* will be more efficient only under conditions unfavorable to *E. stipulatus*.

In conclusion, at lab level, *T.* (*T.*) *exhilaratus* did not perform well with *A. mosoniensis* as diet, therefore, at field level it doesn’t have potential for population control. Differently, *E. stipulatus* had satisfactory performance with *A. mosoniensis* as diet. Therefore, the results suggested that these predatory mites, especially *E. stipulatus*, may, at field level, hinder the biological control of *A. altissima* with *A. mosoniensis* like an agent of control.

8 CONCLUSIONS

The resulths obtained in this thesis allow some conclusions:

✓ The mite fauna in plants can be considered a meaningful indicator of environmental characteristics in the brazilian Pampa. The six year enclosure recovery action in Grazing Exclusion do not promote changes in the acarine community, suggesting similarity between the Grazing Exclusion and the Grassland. The practice of enclosure as a recovery action in the Pampa should be reevaluated, reagrding the subsistence of native fields;

✓ Plant interactions of the mite species are high in the three environments. The consolidation of the technique of identification of mites in plants as an index to environmental characteristics depends on new studies focused on attesting the efficiency of the sampling methodologies and on the recognition of mites species common to different environments;

✓ The difference in climatic conditions and in resources in Riparian Forest provided a higher habitat variability, with reduced competition between species, which led to higher ecological stability of the associated predatory mite community. Predatory mite community must be considered a valuable ecological indicator of the extent to which ecosystems have been disrupted in the Neotropical region. The list of predatory species with associated plants helps to identify new environmental resources preserved in this phytophysiognomy. The results obtained in this study indicate that natural vegetation remnants play an important role in the biological control of agroecosystems as some beneficial mites might migrate from native plants to plantations;

✓ Four new species of eriophyoid mites collected from native plant species from Pampa were described and illustrated herein. The new species described highlight the need for further bioecological research in these environments.

✓ Spider mite surveys conducted in Rio Grande do Sul state, Brazil, allowed us to provide the redescription of *T. armipenis* from specimens collected on native plant species from Pampa biome. Also, were reported for the first time *A. suncinatus* and *S. oryzae* on a new host.

✓ *Typhlodromus* (*Typhlodromus*) *exhilaratus* did not perform well with *A. mosoniensis* as diet, therefore, at field level it doesn’t have potential for population control. Differently, *E. stipulatus* had satisfactory performance with *A. mosoniensis* as diet. In a possible proposition of biological control of *A. altissima* in Europe with the use of *A. mosoniensis* it will be necessary to consider native predators when executing the program. The biological control of *A. altissima* with *A. mosoniensis* will be more efficient only under conditions unfavorable to *E. stipulatus.*

9 OTHER ARTICLES PUBLISHED DURING THE DOCTORATE PERIOD

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