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Ground-Dwelling and Vegetation Ant Fauna in Southern Brazilian Grasslands

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Abstract

Non-forest ecosystems, as natural grasslands from Southern Brazil, are still neglected in conservation policies. Measuring their biodiversity is one of the main steps to generate management strategies for these habitats. This study aims to (i) describe grassland ant richness and composition in Rio Grande do Sul state, and (ii) compare ant communities sampled on the ground and in grassland vegetation, adding to our knowledge of habitat use patterns and vegetation associated species. Six sites were sampled, three belonging to the Pampa biome and three in highland region from the Atlantic Forest biome. Ant fauna was collected once per year in summer during four years in each site with pitfalls traps and sweeping nets. Overall, 29,812 ant individuals were sampled belonging to eight subfamilies, 30 genera e 106 species. The grasslands of Pampa accumulated 91 species and 45 exclusive species, while highland grasslands summed up 61 species and only 15 exclusive species. Species composition differs between biomes as well as between sampling methods. Ant communities sampled from vegetation represented a clear subset of the fauna sampled with pitfall traps, and indication analysis showed only two species associated with this stratum: *Myrmelachista gallicola* and *Pseudomyrmex* nr. *flavidulus*. This study highlights the importance of Southern Brazilian grasslands and the need for specific conservation strategies for the natural grasslands from each biome.

Introduction

Non-forest ecosystems, as grasslands, savannas, shrublands and open woodlands, cover large extensions of land in four of the six Brazilian biomes (Overbeck et al., 2015). Such ecosystems host high levels of unique biodiversity that provisions ecosystem services but are severely neglected concerning conservation policies and protection when compared to forest ecosystems (Andrade et al., 2015). Suitable conservation strategies of non-forest ecosystems often demand different perceptions related to land management (e.g. the role of grazing and fire in grasslands) and offers great opportunities to conciliation with sustainable economic use (Overbeck et al., 2007). Considering the high habitat conversion rates of these ecosystems to other land uses, ecological restoration is a highly necessary component of their conservation (Overbeck et al., 2013). Nevertheless, to define suitable conservation and restoration strategies it is first necessary to better understand species diversity patterns and composition.

In South Brazil, grasslands are naturally widespread over the states of Rio Grande do Sul, Santa Catarina and Paraná, where they are known as Campos. The southern part of Campos ecosystems embraces the Pampa biome, which are among the most species-rich grasslands in the world, extending to Argentina and Uruguay (Rio de La Plata grasslands) (Bilenca & Miñarro, 2004; Overbeck et al., 2007). The northern part of Campos comprises highland grasslands (altitude at about 800 to 1,000 m, with highest peaks up to 1,800 m) that belong to the Atlantic Forest biome where it forms mosaics with *Araucaria* forests (Andrade et al., 2016). Grassland physiognomy and structure varies greatly depending

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on the region, the altitude, and mostly on the management it receives, ranging from very short vegetation in highly grazed systems, to very tall and complex vegetation with shrub and treelet species under low management (Overbeck et al., 2007). Pampa grasslands are more intensively grazed than the highland grasslands of Atlantic Forest, presenting typically a high dominance of prostate plant species. On the other hand, highland grasslands receive frequent burnings (i.e. every one or two years) in the end of the winter, and its physiognomy is dominated by highly fire-resilient grass tussock species (Boldrini, 2009).

Although grassland plant diversity is relatively well known in South Brazilian grasslands, diversity patterns of invertebrate groups are barely studied at all. For example, there is a huge information gap regarding ant fauna when compared to other Brazilian biomes. Ants are abundant and diverse organisms that present very special roles as ecosystem engineering and in provision of ecosystem services (Hölldobler & Wilson, 1990). Worldwide, ant communities have been largely used as a bioindicator group for land use changes and disturbance analysis (e.g. Underwood & Fisher, 2006; Nemec, 2014), showing very positive contributions to rangeland systems monitoring (e.g. Hoffmann, 2010), and evaluation of habitat restoration success (e.g. Andersen & Sparling, 1997). Until now, only a few ant studies were conducted and published specifically in Campos ecosystems. Among them, Albuquerque and Diehl (2009) present an ant survey on highland grasslands, Pinheiro et al. (2010) analyze edge effects in grassland-forest transitions also at this region, and Rosado et al. (2012) compare ant fauna from vineyards and adjacent grassland ecosystems in the Pampa biome. In northeastern Argentina, Calcaterra et al. (2010) and Calcaterra et al. (2014) studied ground-foraging ant responses to grazing and fire in grasslands and savannas in the Iberá Nature Reserve.

Ants explore different resources in a variety of microhabitats (Hölldobler & Wilson, 1990), occupying from forest canopies to subterranean layers. The differential use of a specific strata or microhabitat is commonly found in ant communities in several systems (Vasconcelos & Vilhena, 2006; Schmidt & Solar, 2010; Wilkie et al., 2010), including non-forest ecosystems as savannas (e.g. Cerrado: Campos et al., 2008). Similarly to the habitat heterogeneity hypothesis (Sarty et al., 2006), habitat vertical partition commonly increases species diversity in the ecosystems by reducing competition by resources and allowing coexistence of more species. It is still unknown how ant communities are structured between ground and vegetation layers in grassland ecosystems of South Brazil (e.g. species foraging patterns and microhabitat use), and whether habitat partition can actually occur.

This study describes the ant fauna of sites in a Long-Term Ecological Research program in South Brazilian Grasslands (LTER/PELD Campos Sulinos - CNPq), including six natural grassland ecosystems both in Pampa and Atlantic Forest biomes, sampled with two different methods. The aims of this study were to describe grassland ant community richness and composition (i) from different sites and regions of the Campos ecosystems, and (ii) from ground and vegetation strata, pointing out habitat use patterns and vegetation associated species.

Material and methods

Study area

The study was undertaken in six natural grasslands under traditional cattle grazing in the state of Rio Grande do Sul, Brazil. Three sites were located within private properties in the Pampa biome: Aceguá (31º38'55"S, 54º09'26"W), Alegrete (30º04'11"S, 55º59'34"W) and Lavras do Sul municipalities (30º42'02"S, 53º58'53"W). The other three sites were located within conservation units in the Atlantic Forest biome, in the highland region: Cambará do Sul (29º08'19"S, 50º09'27"W; Aparados da Serra National Park), Jaquirana (29º05'43"S, 50º22'02"W; Tainhas State Park) and São Francisco de Paula municipalities (29º23'35"S, 50º14'26"W; Aratinga Ecological Station) (Fig 1).

Climate in RS is temperate, wet, with hot summers and no dry season (Nimer, 1979). According to the Köppen climate classification, the largest area of RS is classified as Cfb climate, with Cfa restricted to regions with high altitudes in the Pampa and in the highland region of the Atlantic Forest (Kuinchtner & Buriol, 2001). Sites sampled in the Pampa biome have annual mean temperatures of 18ºC, annual mean precipitation of 1423 mm and mean altitude of 224 m. However, sites in the Atlantic Forest have annual mean temperature of 15.3ºC, annual mean precipitation of 1935 mm and mean altitude of 931 m (Climate Data, 2016).

Sampling design

At each grassland site, a homogeneous area of approximately 14.700 m² with traditional grazing was chosen

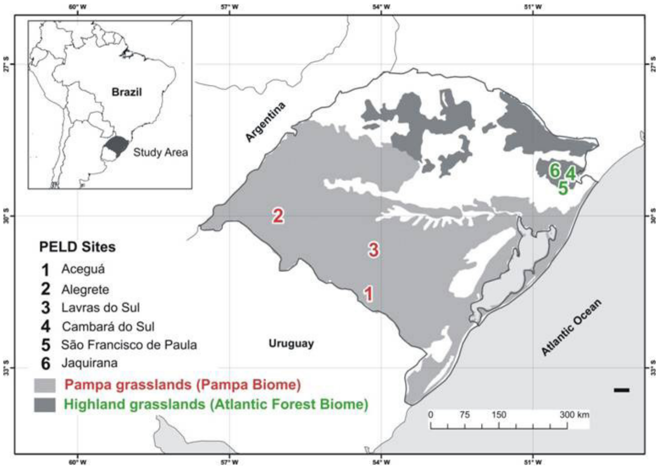

Fig 1. Study sites of Long-Term Ecological Research program (LTER/PELD Campos Sulinos) in Rio Grande do Sul state, Brazil (numbers 1-6). Light gray area represents Pampa grasslands original area, and dark gray area the Atlantic Forest biome.

where 3 plots of 70 x 70 m were settled. By chance, one experimental plot was completely excluded from grazing, another received a conservative grazing management, and the third remained with the local traditional grazing regime. For the present study, treatment information from these three plots was not considered; the differences in ant fauna regarding the grazing treatments will be addressed in a further study.

Ants were sampled once per year in November/December during four years at each site. All samples were carried out from 2011 to 2014; except for Cambará do Sul where they occurred from 2012 to 2015. Two sampling methods were employed: pitfall traps for ground-dwelling ants and sweeping net for ants from grassland vegetation. At each plot eight pitfall traps were installed (24 per site) at least 15 m far from each other. The trap consisted in a 500 ml transparent plastic jar (10 cm diameter, 12 cm deep) filled with 150 ml of formalin (3% formaldehyde), which remained open during seven days. To reduce the evaporation rate of formalin and to protect the traps from direct rainfall, green plastic dishes sustained by wooden sticks were used as rain guards. The ants from vegetation were sampled with sweep net (50 cm wide; sampling area of 0.1 m^2) along four parallel transects in each plot, all pooled together in a unique sample. Vegetation was swept during two different occasions (before pitfall installation and just before their removal) per year, totaling six samples per site (3 plots x 2 occasions). The ant specimens were previously stored in a plastic bag with ethyl acetate. All ant individuals were preserved in ethanol 80% and stored in the Laboratório de Ecologia de Interações (LEIN) in Universidade Federal do Rio Grande do Sul (UFRGS).

Ants were assigned to genera based on dichotomic keys (Baccaro et al., 2015). For species classification, specific literature was used and comparisons were done with material in scientific ant collections in LEIN and the Entomological Collection Padre Jesus Santiago Moure of the Universidade Federal do Paraná (DZUP). All species/morphospecies names follow a standard number from the ant collection of LEIN to standardize different studies and further publications. Vouchers are deposited in ant collections of LEIN and DZUP.

Data analysis

To compare species richness among the different study sites, sample-based species rarefaction curves were calculated for each site with 9999 bootstraps with the iNEXT online tool (Hsieh et al., 2016). Separated curves were built for the different sampling methodologies as well. For that, a matrix with ant incidence data considering all the records of the species in all plots and years pooled together was used; for vegetation ants, incidence was the number of times the species was sampled by sweeping the vegetation (e.g. maximum 24 times per site), and for the ground it was the number of pitfall traps that the species was found in (e.g. maximum 96 pitfalls per site).

Non-metric multidimensional scaling (NMDS) was used to represent the ordination of species composition in the sites within the biomes, considering both methods of sampling. A species absence/presence matrix was built containing the ant species from vegetation and ground in the columns and the plots per site in the rows (18 sampling units), compared with Jaccard similarity index. To test whether there were significant differences in species composition between biomes and between sampling methods, an analysis of similarity (Two-way ANOSIM) was employed, with 9999 permutations. NMDS and ANOSIM analysis were performed with PAST software (Hammer et al., 2001).

To determine ant species association to a specific stratum (vegetation or ground), the Indicator Value (IndVal) method of Dufrêne and Legendre (1997) was used. This method combines measures of specificity of a species to a habitat type and its fidelity within that group. Species with values of 100 would mean perfect indication. Here a value of 70 or higher was considered sufficient for indication of a special relationship between a species and a habitat (Nakamura et al., 2007; Chen et al., 2011; Verdu et al., 2011). A matrix was arranged containing ant species composition in columns and the 18 sampling units (3 plots per site) in rows, in two different groups (vegetation and ground). As the different strata were sampled with different methods and sampling efforts, the matrix was standardized by only considering the incidence data of the species per year in each strata, varying from 0 (no incidence) to 4 (incidence in all years) in each plot. The Indicator Value was calculated for each species using the "multipatt" function of the R package "indicspecies" (R Development Core Team, 2016), based on 9999 permutations.

Results

Overall, 29,812 ant individuals from eight subfamilies, 30 genera and 106 species were sampled (Appendix 1). Myrmicinae was the richest subfamily, with 58 species, followed by Formicinae (16), Ponerinae (13), Dolichoderinae (10), Dorylinae (4), Ectatomminae (3) and Heteroponerinae

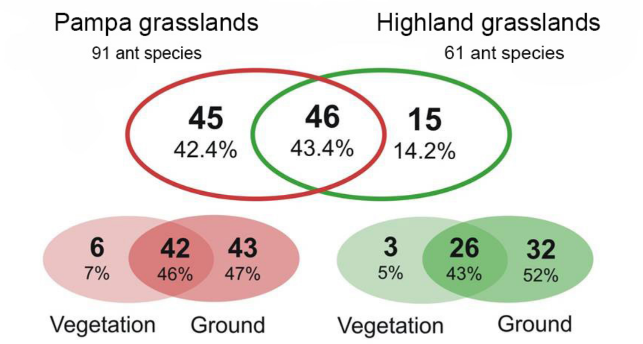

Fig 2. Venn diagrams showing the number and percentage of exclusive and shared ant species for Pampa (red) and Highland (green) grasslands (Atlantic Forest biome), and their respective contribution to differences between vegetation and ground samples.

and Pseudomyrmecinae (one species each). The richest genera were *Pheidole* (18 species), *Solenopsis* (13), *Hypoponera* (10) and *Camponotus* (8). The most frequent genera were *Pheidole* (1,694 occurrences), *Solenopsis* (864), *Camponotus* (514) and *Brachymyrmex* (457).

Overall, the grasslands from Pampa biome accumulated 91 species, while the highland grassland summed up 61 species. In addition, Pampa grasslands had more exclusive species (45 species) than highland ones (15 species), with 46 shared species (Fig 2). Rarefaction curves showed Lavras do Sul and Alegrete municipalities to present significantly higher ant species richness for both sampling methods (Fig 3). Yet, Aceguá and Jaquirana municipalities presented an intermediate richness when considering the ground ant fauna, accumulating more species than Cambará do Sul and São Francisco de Paula sites.

The ant species composition showed clear differences between biomes (ANOSIM: R=0.55, p<0.001), as well as between ground and vegetation sampling (ANOSIM: R=0.68, p<0.001) (Fig 4).

Pitfall traps sampled a total of 100 ant species, and sweeping net 55 species, with 49 species (46%) shared between sampling methods. The proportion of shared species between methods was similar between biomes (Fig 2). Overall, six species occurred exclusively in samples from the vegetation (5% of the total species richness), but the indicator analysis (IndVal) showed only two species particularly associated to this stratum: *Myrmelachista gallicola* Mayr, 1887 and *Pseudomyrmex* nr. *flavidulus* (Smith, 1858) (Appendix 2). Fifty-one species (48% of the total species richness) were only sampled from the ground, and the indicator analysis revealed 17 species strictly associated to the ground stratum (Appendix 2).

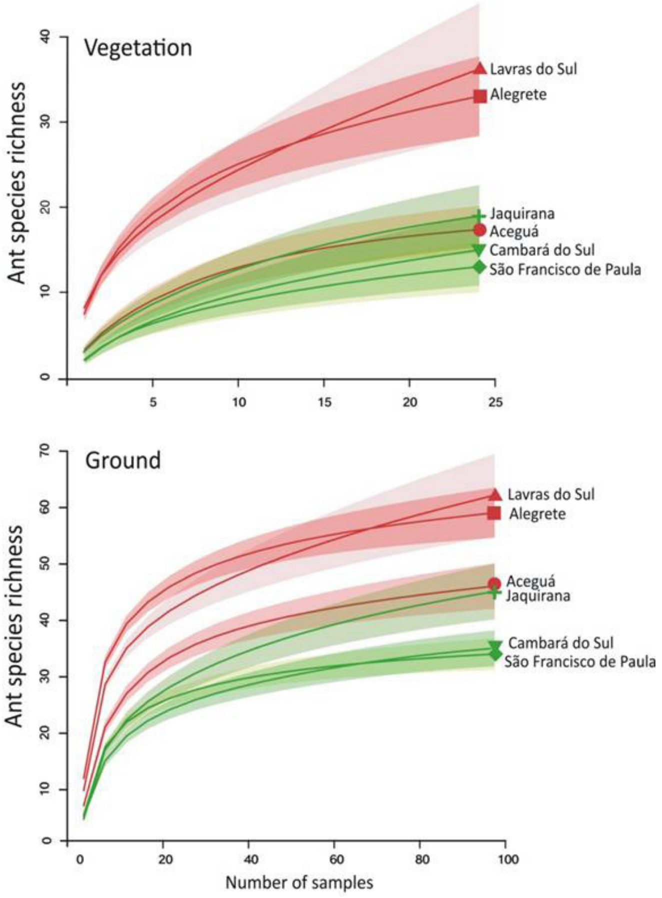

Fig 3. Incidence-based rarefaction for vegetation (24 sampling units by sweep net) and ground samples (96 pitfall traps) in six grassland sites in Rio Grande do Sul state, Brazil, under a multinomial model, with 95% unconditional confidence intervals (shaded area, bootstrap with 9999 replications). Alegrete, Lavras do Sul and Aceguá belong to Pampa grasslands (red) and Cambará do Sul, São Francisco de Paula and Jaquirana to highland grasslands (Atlantic Forest biome) (green).

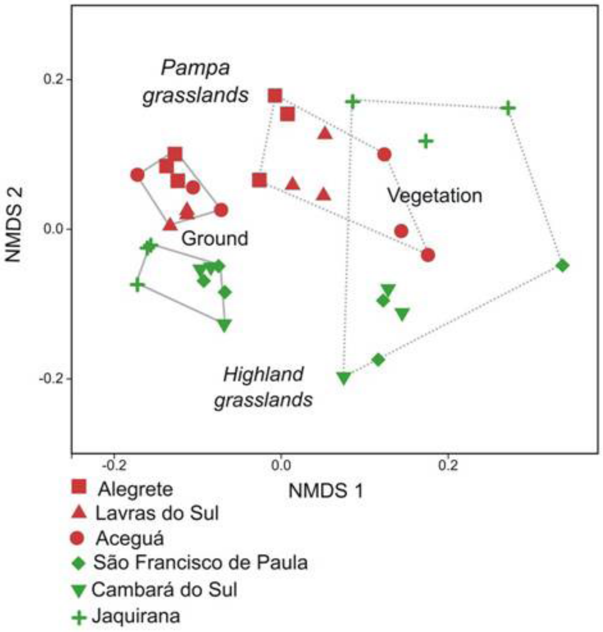

Fig 4. NMDS ordination of grassland sampling sites ant species compositions (presence/absence) with Jaccard similarity index. Pampa grasslands (red): Aceguá (circle), Alegrete (square), Lavras do Sul (triangle); Highland grasslands (green, Atlantic Forest biome): Cambará do Sul (triangle), Jaquirana (plus sign), São Francisco de Paula (diamond).

For grassland vegetation sampling, 13 ant species were considered dominant (more than 20% frequent in samples) in Pampa grasslands, while only four species did so in highland grasslands (Fig 5). The most dominant species in vegetation in both biomes were *Camponotus punctulatus* Mayr, 1868*,* *Brachymyrmex* sp. 1 and *Camponotus* sp. 2, but their level of dominance changed considering the biome, e.g. they comprised more than 50% of all ants in Pampa and less than 30% in highland grasslands (Fig 5).

For ground sampling, 16 ant species were considered dominant in Pampa and 10 in highland grasslands (Fig 5). *Solenopsis invicta* Buren, 1972 was the most frequent species in traps from Pampa (62%), followed by *Cyphomyrmex* gr. *rimosus* sp. 1 (54%). In highland grasslands, *Pheidole obtusopilosa* Mayr, 1887 was highly dominant, present in

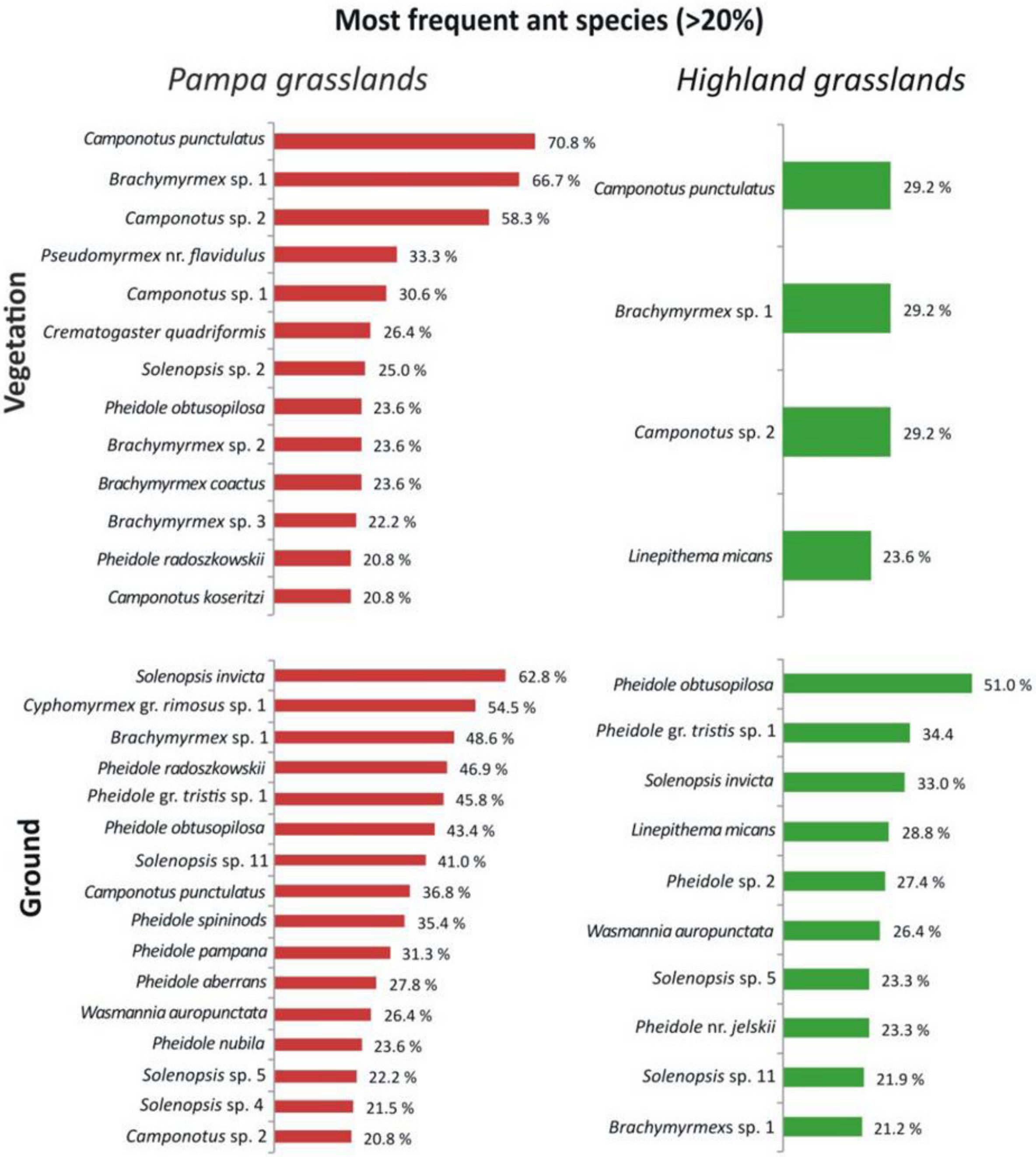

Fig 5. Most frequent ant species in Pampa and highland grasslands (Atlantic Forest biome) from vegetation and ground samples. Only species with >20% presence across all sampling units are shown.

51% of all pitfall traps (Fig 5).

Discussion

This study represents the first attempt to characterize the ant fauna from South Brazilian grasslands reaching sites distributed in two biomes sampled long-term in two target microhabitats. The survey presents the highest ant richness already recorded for these ecosystems, and also adds two new ant species records to Rio Grande do Sul state (based on Diehl et al., 2014 and specialized literature): *Trachymyrmex pruinosus* (Emery, 1906) and *Wasmannia sulcaticeps* Emery, 1894 and a new record to Brazil: *Pheidole pampana* Santschi, 1929 (Ant Maps, 2016; Ant Wiki, 2016). Furthermore, a new species was found (*Acanthoponera* sp. n., RMF unpublished data). In summary, this study reveals differences in ant community structure occurring in grasslands from Pampa and Atlantic Forest biomes, singling out the grasslands of Pampa as very rich ant spots. Ant fauna sampled from vegetation by sweeping net appeared to be a subset of the ant fauna sampled from the ground by pitfall trapping. Altogether, the results presented here may provide useful information for future studies and conservation efforts.

Few, if any, ant studies so far have managed to sample for longer times and used widely spaced sampling sites for

Campos ecosystems. For example, Albuquerque and Diehl (2009) surveyed 32 ant species along eight grassland sites in Cambará do Sul municipality at the highland region. Pinheiro et al. (2010) recorded 31 morphospecies in grassland-forest ecotones also at this region. Rosado et al. (2012) related 72 ant species to vineyards and adjacent grassland habitats at the Campanha region in Pampa biome. In northeastern Argentina, which represents an extension of the Campos ecosystems, Calcaterra et al. (2010) evaluated the effect of grazing on 50 ant species in savanna and grassland, while Calcaterra et al. (2014) studied fire effects on 67 grassland ant species, both at Iberá Nature Reserve. In comparison to these studies, our survey shows an expressive ant species richness (106 species) that could be clearly explained by (i) the longer sampling duration (i.e. 4 years) increasing the likelihood of finding rare or eventual species, and (ii) the broader geographic scale attained, which incorporates a greater variety of environments and management situations. Variation in site characteristics, such as latitude, altitude, soil types, climate, land management (e.g. grazing intensity and fire frequency) and vegetation physiognomy, are generally correlated to a greater variation in ant species composition, likely enhancing gamma diversity (i.e. total species richness; Schoeman & Foord, 2012).

This explanation seems to be also useful to elucidate why we sampled more ant richness (i.e. 91 spp.) in Pampa grasslands than in the highland region (i.e. 61 spp.). Since the geographic area in Pampa is larger and the sampled sites were spatially further apart, there is indeed more heterogeneity of associated habitat conditions (e.g. soil types, vegetation physiognomies, plant richness; Streck et al., 2008; Ferreira et al., unpublished data) and thus a higher probability of finding a richer associated ant community in the Pampa. Ant fauna composition was also singular between these grassland regions (sharing only 43% of the total richness), and community structure based on dominant species also shifts regarding number of dominant species and their identity. A pattern that could be draw is that ant communities from highland grasslands seemed to be more even in terms of species incidence. Altitude could certainly have an important contribution explaining ant fauna differences between the two biomes (Szewczyk & McCain, 2016). Highland grasslands in the Atlantic Forest biome are situated at altitudes from 800 to 1,000 m and have lower mean and minimal annual temperatures. Several temperature-based hypothesis emerge to explain the decline in species diversity from low to higher altitudes, for example, relating colder temperatures to decreased food resources, reduced foraging periods, and lower metabolic rates (Sanders et al., 2007; Malsch et al., 2008). Broad-scale diversity patterns in ants are likely to be supported by multiple entangled drivers, including interspecific competition, not yet comprehensively understood (Szewczyk & McCain, 2016), thus our explanations here are tentative.

Considering samplings in the ground, *S. invicta* and *C.* gr. *rimosus* sp. 1 were dominant species in Pampa, while *P. obtusopilosa* was dominant in highlands. *Solenopsis invicta* is a generalist ant, known for its high competitive ability and one of the main species of invasive ants elsewhere. Native from South America, this species was first introduced to southern United States and later to other regions of the world (Ascunce et al., 2011). Despite being responsible for damages in urban, agricultural and natural environments in non-native regions, their occurrence as the most abundant and/or frequent species is also reported in studies carried out in grassland ecosystems of Argentina (Calcaterra et al., 2010; Calcaterra et al., 2014). *Cyphomyrmex* is a fungus-farming ant. Species in this group usually nest in the ground, leaf litter and rotten logs (Mackay & Serna, 2010). However, the *rimosus* group is known by the morphological complexity of its species, and a comprehensive taxonomic revision is currently under preparation (E. Z. Albuquerque & C. R. F. Brandão, unpublished data). Ants in the genus *Pheidole* are usually generalist. They are widely distributed both global and locally and may occur from the vegetation canopy to the soil of open areas and forests (Wilson, 2003). Little is known about the biology of *P. obtusopilosa*, and its distribution is recorded from Argentina, Uruguay and Brazil (Rio Grande do Sul state) (Ant Maps, 2016; Ant Wiki, 2016).

Forty-nine ant species were found using both ground and grassland vegetation strata. Although ant nests are predominantly established in the ground for grassland ecosystems, ant use of the local vegetation depends on the foraging behavior of each species (Blüthgen & Feldhaar, 2010). Habitat structure (i.e. biomass, plant height, plant richness, and their spatial heterogeneity), and resource availability (i.e. presence of flowers, fruits, seeds, plants with extrafloral nectaries) could attract ant communities to forage and nest in the vegetation (Campos et al., 2008), which in its turn depends on the management employed on each site (Overbeck et al., 2007; Overbeck et al., 2016). Looking at the ant composition ordination diagram, a greater dispersion among vegetation samples is detected when compared to ground samples. This might be explained by the variability in vegetation structure and resources found between plots and sites. One species of *Brachymyrmex* and two species of *Camponotus* were the most frequent species foraging at vegetation in both biomes; these genera belong to subfamily Formicinae and can present arboreal habits (Baccaro et al., 2015). Marques and Del-Claro (2006) found Formicinae as dominant on the vegetation in either open or closed areas of Cerrado, especially *Camponotus* species.

Two ant species showed association with vegetation stratum (*M. gallicola* and *P.* nr. *flavidulus*). *Myrmelachista* is considered an exclusively arboreal genus (Longino, 2006), nesting in cavities and dry twigs of living trees, and is rarely found foraging on the ground (Nakano et al., 2013; Baccaro et al., 2015). Species in this genus may also develop associations with host plants, extrafloral nectaries or associated aphids. *Pseudomyrmex* is also a predominantly arboreal genus; it builds nests on tree twigs or hollow trunk cavities, and forages predominantly in the vegetation (Baccaro et al., 2015). Both *M. gallicola* and *P.* nr. *flavidulus* were found in different plots at sites from Pampa and highland region (Appendix 1). A high number of plant species associated to the grass matrix, including shrub, treelet and even pioneer woody species, could be hosting ant populations of *M. gallicola* and *P.* nr. *flavidulus*. As our sweeping net sampling included all vegetation found within the plots, further studies should specifically investigate details on host plants, and a possible relation to plot grazing management applied.

This study contributes with the overall description of ant diversity and composition from different sites and biomes in the South Brazilian grasslands. The dynamics of the grassland ant communities along time (i.e. four years), which is a significant dimension of ecological studies contributing to consolidation of diversity patterns, and the ant responses to different grassland managements (i.e. grazing exclusion, traditional grazing and conservative grazing) applied to our study sites will be approached in details in a further manuscript. The conservation planning of biodiversity encompasses a variety of knowledges; one of the first and more fundamental aspects is surveying biodiversity, providing spatially consistent information on surrogate taxa and habitats. Information gained from this study could be used in future research, and may help design a regional plan for grassland conservation and restoration, for example, helping definitions of areas to be protected or serving as reference sites for restoration. We emphasize that grassland biodiversity conservation efforts should consider different strategies for each biome, in order to maximize biodiversity conservation. Furthermore, the creation of conservation units in the Pampa biome is urgently needed, since the current conservation units in the Atlantic Forest biome cannot preserve all biodiversity associated to South Brazilian grasslands.

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Appendix 1. List of species recorded in sites of Long-Term Ecological Research program (LTER/PELD Campos Sulinos) in Rio Grande do Sul state, Brazil (1 - Aceguá; 2 - Alegrete; 3 - Lavras do Sul; 4 - Cambará do Sul; 5 - São Francisco de Paula; 6 - Jaquirana). Numbers represent the total number of occurrences in the two biomes (Pampa and Atlantic Forest) and strata (G - ground; V - vegetation).

Ant species composition	**Pampa biome**						**Atlantic Forest biome**					
Sites	**1**		**2**		**3**		**4**		**5**		**6**	
Strata	**G**	**V**	**G**	**V**	**G**	**V**	**G**	**V**	**G**	**V**	**G**	**V**
Dolichoderinae												
Dorymyrmex pyramicus (Roger, 1863)	0	0	11	4	3	1	1	0	0	0	3	2
Dorymyrmex sp. 1	0	0	0	0	0	0	0	0	0	0	1	0
Dorymyrmex sp. 2	0	0	0	0	1	0	0	0	0	0	0	0
Dorymyrmex sp. 4	0	0	0	5	0	1	0	0	0	0	0	0
Dorymyrmex sp. 5	0	0	1	0	0	0	0	0	0	0	0	0
Dorymyrmex sp. 6	0	0	0	1	0	0	0	0	0	0	0	0
Gracilidris pombero Wild & Cuezzo, 2006	12	0	0	0	2	0	0	0	0	0	0	0
Linepithema micans (Forel, 1908)	2	0	39	6	16	3	48	13	11	1	24	3
Linepithema sp. 2	0	0	0	1	4	10	0	0	0	0	0	0
Tapinoma sp. 1	6	0	49	1	0	1	0	0	1	0	0	0
Dorylinae												
Acanthostichus quadratus Emery, 1895	0	0	0	0	0	0	0	0	0	0	1	0
Neivamyrmex sp. 4	0	0	0	0	1	0	0	0	2	0	3	0
Neivamyrmex sp. 5	0	0	0	0	1	0	0	0	0	0	0	0
Neivamyrmex sp. 6	0	0	0	0	0	0	0	0	0	0	1	0
Ectatomminae												
Ectatomma edentatum Roger, 1863	5	0	0	0	5	0	0	0	0	0	6	0
Gnamptogenys rastrata (Mayr, 1866)	0	0	1	0	6	0	0	0	2	0	1	0
Gnamptogenys striatula Mayr, 1884	1	0	0	0	34	0	2	0	0	0	18	0
Formicinae												
Brachymyrmex coactus Mayr, 1887	0	0	18	12	10	5	0	0	0	0	0	0
Brachymyrmex sp. 1	42	10	62	23	36	15	43	6	17	13	1	2
Brachymyrmex sp. 2	11	5	15	3	29	9	15	3	10	1	4	1
Brachymyrmex sp. 3	0	0	7	10	6	6	0	0	0	0	0	0
Brachymyrmex sp. 4	0	0	4	1	1	0	0	0	0	0	0	0
Brachymyrmex sp. 5	0	0	1	0	0	0	0	0	0	0	0	0
Camponotus koseritzi Emery, 1888	0	0	0	0	21	15	0	0	0	0	0	0
Camponotus punctulatus Mayr, 1868	58	19	23	17	25	15	5	7	11	10	8	4
Camponotus rufipes (Fabricius, 1775)	0	0	3	0	0	0	0	0	0	0	0	0
Camponotus sp. 1	11	2	23	16	22	4	4	0	2	0	23	10
Camponotus sp. 2	3	3	21	22	36	17	0	0	0	0	24	21
Camponotus sp. 4	0	0	0	0	2	0	0	0	0	0	0	0
Camponotus sp. 5	0	0	0	0	1	0	0	0	2	0	0	0
Camponotus sp. 6	0	0	0	1	0	1	0	0	0	0	0	2
Myrmelachista gallicola Mayr, 1887	0	0	0	2	0	11	0	1	0	0	0	3
Nylanderia fulva (Mayr, 1862)	11	0	0	0	31	1	0	0	0	0	6	0
Heteroponerinae												
Acanthoponera sp. n. 1	0	0	0	0	0	0	0	0	0	0	2	1
Myrmicinae												
Acromyrmex ambiguus (Emery, 1888)	8	2	0	0	17	5	0	0	0	0	0	0
Acromyrmex coronatus (Fabricius, 1804)	2	0	0	0	0	0	4	0	7	0	0	1
Acromyrmex heyeri (Forel, 1899)	21	6	1	1	1	0	1	2	3	2	0	0

Appendix 1. List of species recorded in sites of Long-Term Ecological Research program (LTER/PELD Campos Sulinos) in Rio Grande do Sul state, Brazil (1 - Aceguá; 2 - Alegrete; 3 - Lavras do Sul; 4 - Cambará do Sul; 5 - São Francisco de Paula; 6 - Jaquirana). Numbers represent the total number of occurrences in the two biomes (Pampa and Atlantic Forest) and strata (G - ground; V - vegetation). (Continuation)

Ant species composition	**Pampa biome**						**Atlantic Forest biome**					
Sites	**1**		**2**		**3**		**4**		**5**		**6**	
Strata	**G**	**V**	**G**	**V**	**G**	**V**	**G**	**V**	**G**	**V**	**G**	**V**
Acromyrmex landolti (Forel, 1885)	0	0	0	0	0	0	0	0	0	0	1	0
Acromyrmex lobicornis (Emery, 1888)	14	1	2	0	0	0	0	0	0	0	0	0
Cephalotes incertus (Emery, 1906)	0	0	0	0	0	2	0	0	0	0	0	0
Crematogaster quadriformis Roger, 1863	6	1	36	16	4	2	0	0	0	0	0	0
Crematogaster sp. 1	1	2	0	0	0	0	0	0	0	0	0	0
Crematogaster sp. 2	0	0	1	0	8	1	0	0	0	0	0	0
Crematogaster sp. 3	0	0	0	1	0	0	0	0	0	0	0	0
Crematogaster sp. 4	0	0	1	0	0	0	0	0	0	0	0	0
Cyphomyrmex gr. *rimosus* sp. 1	55	0	33	1	69	0	5	0	16	0	31	0
Cyphomyrmex transversus Emery, 1894	0	0	39	0	0	0	0	0	0	0	0	0
Megalomyrmex gr. *silvestrii* sp. 1	0	0	1	0	0	0	0	0	0	0	0	0
Megalomyrmex sp. 2	1	0	0	0	0	0	0	0	0	0	0	0
Mycetophylax nr. *lilloanus* (Kusnezov,1949)	0	0	1	0	0	0	0	0	0	0	0	0
Pheidole gr. *fallax* sp. 1	2	0	3	0	0	0	0	0	0	0	0	0
Pheidole gr. *tristis* sp. 1	75	4	32	1	25	3	41	6	56	2	2	0
Pheidole gr. *tristis* sp. 2	0	0	4	0	0	0	0	0	0	0	0	0
Pheidole aberrans Mayr, 1868	35	0	28	1	17	0	4	0	1	0	2	0
Pheidole breviseta Santschi, 1919	2	0	6	1	46	0	19	1	2	0	10	0
Pheidole cavifrons Emery, 1906	0	0	17	0	31	0	23	0	8	0	23	0
Pheidole nr. *jelskii* Mayr, 1884	0	0	1	0	8	0	2	0	13	0	52	1
Pheidole nubila Emery, 1906	25	0	21	0	22	1	0	0	0	0	0	0
Pheidole obtusopilosa Mayr, 1887	43	5	32	1	50	11	48	3	73	6	26	2
Pheidole pampana Santschi, 1929	33	0	22	2	35	4	19	1	10	0	23	0
Pheidole radoszkowskii Mayr, 1884	29	1	53	7	53	7	12	0	16	0	17	1
Pheidole nr. *rufipilis* Forel, 1908	0	0	0	0	0	0	0	0	0	0	1	0
Pheidole spininods Mayr, 1887	1	0	54	1	47	0	0	0	0	0	0	0
Pheidole sp. 1	0	0	0	0	38	0	17	0	35	0	2	0
Pheidole sp. 2	1	0	0	0	36	0	30	1	44	1	5	0
Pheidole sp. 3	0	0	10	1	0	0	0	0	0	0	36	1
Pheidole sp. 4	1	0	1	0	5	0	0	0	0	0	0	0
Pheidole sp. 13	0	0	0	0	0	0	0	0	0	0	1	0
Pogonomyrmex naegelii Emery, 1878	0	0	0	0	0	0	0	0	1	0	15	0
Solenopsis invicta Buren, 1972	52	2	57	2	72	0	19	0	71	6	5	1
Solenopsis sp. 2	4	3	8	10	22	5	7	2	9	0	0	0
Solenopsis sp. 3	0	0	0	0	0	0	5	0	0	4	1	5
Solenopsis sp. 4	7	0	46	4	9	1	0	0	0	0	4	0
Solenopsis sp. 5	8	0	27	0	29	0	33	0	16	0	18	2
Solenopsis sp. 6	0	0	0	0	0	0	0	0	0	0	1	0
Solenopsis sp. 7	13	0	3	0	0	0	0	0	0	0	0	0
Solenopsis sp. 8	1	0	16	0	0	0	0	0	0	0	0	0
Solenopsis sp. 9	0	0	1	0	6	0	3	0	1	0	2	0
Solenopsis sp. 10	13	0	29	0	0	0	0	0	0	0	0	0

Solenopsis sp. 11	23	3	27	3	68	2	41	1	1	1	21	0
Solenopsis sp. 12	0	0	1	1	2	0	0	0	1	0	2	0
Solenopsis sp. 13	0	0	1	0	0	0	0	0	0	0	0	0
Strumigenys emiliae Forel, 1907	0	0	0	0	1	0	0	0	0	0	0	0
Strumigenys louisianae Roger, 1863	0	0	0	0	18	2	15	1	6	0	0	0
Trachymyrmex gr. *urich* sp. 1	4	0	5	1	13	0	19	0	3	0	0	0
Trachymyrmex holmgreni Wheeler, 1925	0	0	0	0	0	0	0	0	0	0	5	0
Trachymyrmex kempf Fowler, 1982	4	0	16	1	14	0	0	0	0	0	0	0
Trachymyrmex pruinosus (Emery, 1906)	0	0	0	0	0	0	11	0		0	0	0
Wasmannia auropunctata (Roger, 1863)	14	0	1	0	61	5	26	1	27	2	23	0
Wasmannia sulcaticeps Emery, 1894	6	0	11	0	5	0	0	0	2	1	0	0
Wasmannia nr. *sulcaticeps* Emery, 1894	0	0	0	0	5	0	8	0	0	0	0	0
Wasmannia williamsoni Kusnezov, 1952	10	0	1	0	12	1	0	0	0	0	0	0
Ponerinae												
Anochetus neglectus Emery, 1894	6	0	7	0	3	0	0	0	0	0	0	0
Hypoponera sp. 1	0	0	1	0	1	1	2	0	0	0	0	0
Hypoponera sp. 2	0	0	3	0	0	0	0	0	2	0	2	0
Hypoponera sp. 3	0	0	2	0	0	0	0	0	0	0	0	0
Hypoponera sp. 4	0	0	2	0	0	0	0	0	0	0	0	0
Hypoponera sp. 5	1	0	0	0	0	0	0	0	0	0	0	0
Hypoponera sp. 6	4	0	1	0	0	0	0	0	0	0	0	0
Hypoponera sp. 7	0	0	0	0	3	0	0	0	1	0	0	0
Hypoponera sp. 8	0	0	2	0	0	0	0	0	0	0	0	0
Hypoponera sp. 9	0	0	0	0	0	0	0	0	0	0	1	0
Hypoponera sp. 10	0	0	0	0	0	0	1	0	0	0	0	0
Neoponera bucki (Borgmeier, 1927)	0	0	0	0	3	0	0	0	0	0	0	0
Pachycondyla striata Smith, 1858	0	0	0	0	0	0	0	0	23	0	1	0
Pseudomyrmecinae												
Pseudomyrmex nr. *flavidulus* (Smith, 1858)	0	1	1	12	0	11	0	0	0	0	0	10

Appendix 2. Results from Indicator Value (IndVal) analysis showing ant associations to either vegetation or ground strata. Were considered IndVal of 70 or higher as important. For all IndVal presented, p<0.001.

Species	Stratum	IndVal
Pseudomyrmex nr. *flavidulus*	Vegetation	73.1
Myrmelachista gallicola	Vegetation	70.7
Solenopsis sp. 5	Ground	98.3
Cyphomyrmex gr. *rimosus* sp. 1	Ground	96.4
Pheidole pampana	Ground	95.5
Pheidole cavifrons	Ground	91.3
Pheidole radoszkowskii	Ground	88.4
Solenopsis sp. 11	Ground	88
Solenopsis invicta	Ground	87.7
Pheidole aberrans	Ground	87

Species	Stratum	IndVal
Pheidole breviseta	Ground	85.9
Wasmannia auropunctata	Ground	82.6
Pheidole nr. *jelskii*	Ground	80.3
Trachymyrmex gr. *urich* sp. 1	Ground	80.2
Pheidole sp. 2	Ground	79.7
Pheidole sp. 1	Ground	78.2
Wasmannia sulcaticeps	Ground	76.3
Solenopsis sp. 4	Ground	73.1
Gnamptogenys striatula	Ground	70.7