Diversity of ants on Guabiroba trees (Campomanesia xanthocarpa O. Berg) of Mixed Ombrophilous Forest

Rafael Stempniak Iasczczaki

rafael.iasczczaki@ufv.br

Federal University of Vicosa Department of Entomology: Universidade Federal de Vicosa Departamento de Entomologia
https://orcid.org/0000-0002-5915-6291

Danielly Lisoski Correia

University of the State of Paraná: Universidade Estadual do Parana

Rodrigo Doline

Federal University of Parana: Universidade Federal do Parana

Roberta Holdefer

University of the State of Paraná: Universidade Estadual do Parana

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Abstract

There are many types of interactions between ants and plants, and many of these are first discovered by the observation and identification of the ants on particular plant species. Therefore, studying the diversity of ants on different strata of a plant species, for example, is indispensable to identify interactions and their ecological implications. In this study we evaluated the diversity of visitor ants on two strata of Guabiroba trees (*Campomanesia xanthocarpa* O. Berg), a tree species from Mixed Ombrophilous Forest that possess ecological, economical, medicinal, and for food importance. We sampled the ant species from lower plant strata (in the plant surroundings) by collecting them manually in 97 sites; and from higher strata through *pitfall* traps, suspended in branches and leaves of 117 Guabiroba trees. We identified 30 ant morphospecies on Guabiroba trees (Manual Sobs = 22; Sest = 46,74; Pitfall Sobs = 21; Seat = 21.89) and our results estimated increased ant richness if a higher sampling effort was applied. *Nylanderia fulva* was the most abundant species, followed by *Camponotus crassus*, *Ca. rufipes*, and *Ca. melanoticus*, which occurred very abundantly, frequently and constantly on Guabiroba trees, acording to the faunal analysis. More than half of the recorded species only occasionally visited Guabiroba trees. Our results indicated an overlap in composition of ant species from both studied strata, suggesting regular visitation of some ants and perhaps interactions between them and the plant. Our study contributes adding information on the ants from Mixed Ombrophilous Forest and ecological relations between Guabiroba trees and their visitor ants.

Introduction

Ants are social insects of success in terrestrial ecosystems (Wilson, 1971; Bolton 1995) where present high richness, abundance, biomass, broad distribution, and are engaged in diverse fundamental ecosystem services (Hölldobler and Wilson 1990). The success of the group is mainly due to their sociability, behavioral and phenotypic plasticity, and a high ability to establish interactions with other species in the environment (Wilson, 1971; Hölldobler and Wilson 1990; Oliveira and Pie 1998).

With respect to the interactions involving ants, those with plants stand out (Wheeler 1910; Del-Claro et al. 1996; Fernandez, 2003; Jazen, 1967;). Ants and plants are always related to each other, to some extent of interaction (Delabie et al. 2003; Soares et al. 1998; Delabie et al. 2006; Schütte et al. 2007; Lacau et al. 2008; Rodrigues et al. 2008; Gomes et al. 2010; Lopes et al. 2010). Ants can be detrimental, beneficial or neutral to the plants, depending on the species involved and the ecological conditions (Bronstein 1994) and plants can offer many facilities for ants to be maintained. Plants can face herbivory from ants (e.g., leaf cutting ants) (Nobúa Behrmann et al. 2017), which in turn benefit from plants; in contrast, some plants can attract and arrest predatory ants which defend plants from herbivorous attacks, because the provision of non-prey food sources (e.g., floral and extrafloral nectar) and/or refuge (Jazen, 1967; Del-Claro et al. 1996; Fernandez, 2003); ants can act as pollinators in some cases (Gómez 2000); and they may simply use plants as a substrate for foraging or nesting (Fernandez, 2003). In the face of the variability of ant-plant interactions, one of the first steps to identify them is exploring the diversity of ants present in particular plant species, because it reveals the ant species occurring and their relative abundances in the plant (Harada and Adis 1998; Yamazaki et al. 2016).

Due to the variability in habits of living and behavior, ants may occupy different strata in a determined environment (Longino and Nadkarni 1990; Arruda et al. 2016), also because there are fundamental differences in the structure, temperature, humidity and other factors associated to the strata. For example, the community of ants from the canopy in rainfall forests is quite different from that found in the soil or leaf-litter, surrounding the trees in the same environment (Longino and Nadkarni 1990; Brühl et al. 1998). This indicates that interactions between plants and ants may be strata specific in large trees (Longino and Nadkarni 1990), and therefore, exploring different strata is fundamental for a better understanding of the ant species associated to the plant. In the present paper we accessed the ant fauna from two strata of a particular plant species, *Campomanesia xanthocarpa* O. Berg. (Myrtaceae), by using two different sampling methods in a Mixed Ombrophilous Forest fragment. We encompassed in more details the ant species community and also checked possible strata preference according to the information on the richness and abundance of ants.

Campomanesia xanthocarpa, also known as Guabirobeira, Guabiroba, Guabiroba-do-mato, or Guavirova, is a very common and important arboreal species native from Brazil, deciduous and of occurrence mainly in Mixed Ombrophilous Forests from the South of the country (Oliveira et al. 2023). This species contributes strongly with leaf litter and macronutrient production in the forests where occurs, occupying ecological importance in the structure of such ecosystems (Marques, 2008). Further, its cultivation is indicated for the regeneration of degraded areas or those contaminated with petroleum (Backes and Irgang 2002; Gogosz, 2008). Economic importance is also attributed to *C. xanthocarpa.* The fruits produced by this tree are used in sweets, liquors, ice cream, and for juice preparation (Freitas et al. 2008; Barbieri et al. 2017); the leaves have medicinal properties (Viecili et al. 2014; Leandro et al. 2021); and its flowers possess a high melliferous potential (Backes and Irgang 2002). In addition, guabiroba wood has a long natural durability and is used for gadget production, in addition to be used as firewood and for charcoal production (Backes and Irgang 2002).

Despite the information aforementioned, no study has investigated potential implications of interactions with ants to this important plant species in the South of Brazil. Indeed, to the best of our knowledge, no information related to ants and *C. xanthocarpa* is available in the literature. Thus, aiming at provide the first study on this topic, we aimed to investigate the ant richness and abundance present on *C. xanthocarpa* trees in a Mixed Ombrophilous Forest fragment in the South region of Brazil. This study provides results that might be used in future researches exploring specific interactions of ants and *C. xanthocarpa* and their ecological implications.

Material And Methods

Study Area

The study was carried out in a fragment of Mixed Ombrophilous Forest, in the municipality of Paula Freitas, state of Paraná (26º26’19.8” S e 50º57’46.9” W 26°11'59.57"S 50°56'14.91"W) (Fig. 1). The climate in the region, according to the Koppen classification, is subtropical, humid, and mesothermal, with mild summers, rainfall well distributed throughout the year, and severe, frequent frosts (Maack 1968). The annual rainfall exceeds 1,400 mm and the temperature ranges from 13ºC to 21ºC, with an annual average of 17ºC (WREGE et al. 2012).

The site where the study was carried out is approximately 94,062.381 m². It contained about 300 individuals of *C. xanthocarpa* randomly distributed among other vegetation or as isolated trees (in more open environments). Other common plant species (trees) found in this area are *Cedrella fissilis* Vellozo, *Nectandria* sp., *Cinamodendron dinisii* Schwacke, *Sebastiania commersoniana* (Baill.) L.B. Sm. & Downs, *Mimosa scabrella* Bentham, *Ocotea porosa* (Nees & C. Mart.) (Imbuia) and *Araucaria angustifolia* Kuntze, among others.

Sampling methods

We randomly selected 117 *C. xanthocarpa* trees of at least 50 years of age (Diameter at breast height/ DBH verification > 15 cm, (dos Santos et al. 2017) for the study. Ant individuals were sampled between October 2017 and May 2018 by using two methodologies from the selected trees: manual sampling (employed on 97 trees) and suspended *pitfall* traps (on 117 trees). For manual sampling, we searched actively for ants on each selected tree, and their surroundings, during 20 minutes. Leaf-litter surrounding the roots, trunks and low branches were explored towards ants by using tweezers, brushes, and tubes for packaging the individuals (Fernandez, 2003). In the case of suspended *pitfall* trap samplings (Agosti 2000), we used PET bottles previously cut, about 10 cm high, containing 70 mL ethanol 70%, without any type of baits for ant attraction. In each selected tree, we set a trap near the trunk or leaves, in the highest plant region as possible (± 5 m high). Each trap was kept active for 72 hours and the biological material was then collected.

The collected ants were taken to the Insects Ecology Lab, Biological Science Sector from the State University of Paraná, Campus União da Vitória. Ants were sorted and identified to the lowest possible taxonomic level, using taxonomical keys according to Bolton (1995). When species level was not possible to be identified we standardized the specimens as morphospecies, according to the ant collection already present in the lab. All ant individuals collected in this research were deposited in the collection of ants from the same lab.

Statistical analysis

Data was organized as records of ant occurrence for each sampling method and tree (Longino et al. 2002). The ant communities present in manual sampling and suspended *pitfall* trap were estimated and compared using rarefaction curves and Chao 2 estimator. Further, Pielou's Evenness index and paired t-test were applied. These analyses were carried out by using the 'Vegan' package in R (R Development Core Team, 2022).

Furthermore, all ant species collected by both sampling methods were analyzed using a faunistic analysis to define the respective dominance and abundance classes. Abundance was obtained through the mean and standard error of the overall number of records for each ant species from each tree (Garcia and Corseuil 1998). We determined the confidence interval as 5% and 1% probability so that the following abundance classes were established: va = very abundant (occurrence between the upper limits of CI at 1%), a = abundant (occurrence between the upper limits of CI at 5% and 1%), c = common (occurrence within CI at 5%), d = disperse (occurrence between the lower limits of CI at 5% and 1%) and r = rare (occurrence below the lower limit of CI at 1%) (Thomazini and Thomazini 2002). The relative frequency of species results from the division of the number of occurrences of the species by the total number of collected individuals (Dajoz, 1973). We compared this result with the confidence interval, generating data as infrequent, frequent, and very frequent. The recognition of the species constancy requires the percentage of times in which the species occurred within the sampling universe. We checked the species record number in each sampling episode (Dajoz, 1973). The species were grouped as constant, accessory, and accidental, respectively, when present in more than 50% (W), between 25% and 50% (Y), and up to 25% (Z) samplings (Bodenheimer 1955).

The faunistic analysis was carried out using AnaFau Software (Moraes et al., 2003). We defined the dominance at 10% representativeness in samplings (Novotný and Basset 2000). The dominant species were those which presented a relative percentage frequency higher than 10% (fr > 0.1).

Results

We identified a total of 30 ant species (morphospecies) on *C. xanthocarpa* trees. Twelve of them were Formicinae and nine Myrmicinae, with *Camponotus* (n = 9) and *Pheidole* (n = 4) the richest genus, respectively (Table 1). From the total occurring species, 22 were sampled with manual sampling and 21 with suspended *pitfall* traps, while 13 of them were shared by both sampling methods. *Nylanderia fulva* (Mayr, 1862) stood out, making up more than 47% of the records in manual sampling and classified as super-dominant, super-abundant, super-frequent, and super-constant (Table 1). Further, the rarefaction curves did not stabilize for any of the sampling methods (Fig. 2), mainly for manual sampling and chao 2 indicated a higher richness of ants than that we observed: Manual Sobs = 22; Sest = 46,74; Pitfall Sobs = 21; Seat = 21.89.

Table 1
Faunistic analysis and relative frequency (%) of ant species collected on *Campomanesia xanthocarpa* O. Berg, in a Mixed Ombrophilous Forest, using suspended *pitfall* traps or manual sampling in the period between October 2017 and May 2018.

TAXON	**Manual**					***Pitfall***				
	***Domin.**	**Abund.**	**Freq.**	**Const.**	**%**	***Domin.**	**Abund.**	**Freq.**	**Const.**	**%**
DOLICHODERINAE										
Dorymyrmex brunneus (Forel, 1908)	ND	c	F	Y	0.033	ND	r	IF	Z	0.008
Dorymyrmex pyramicus (Roger, 1863)	ND	r	IF	Y	0.005					
Linepithema humile (Mayr, 1868)	ND	d	IF	Y	0.011					
Linepithema micans (Forel,1908)	ND	va	VF	Y	0.049	ND	r	IF	Z	0.012
Linepithema pulex (Wild, 2007)	ND	r	IF	Y	0.005					
FORMICINAE										
Camponotus crassus (Mayr,1862)	ND	va	IF	Y	0.049	D	va	VF	W	0.194
Camponotus melanoticus (Emery, 1894)						D	va	VF	W	0.110
Camponotus rufipes (Fabricius, 1775)	ND	va	VF	Y	0.049	D	va	VF	W	0.177
Camponotus sp. 1	ND	r	IF	Y	0.006					
Camponotus sp.2						ND	r	IF	Z	0.012
Camponotus sp. 3	ND	c	F	Y	0.033	ND	r	IF	Z	0.012
Camponotus sp. 4	ND	r	IF	Y	0.005	ND	d	IF	Z	0.020
Camponotus sp. 5						ND	c	IF	Y	0.031
Camponotus sp. 6						ND	r	IF	Z	0.008
Myrmelachista arthuri (Forel, 1903)	ND	r	IF	Y	0.005					
Myrmelachista catharinae Mayr, 1887						ND	d	IF	Z	0.016
Nylanderia fulva (Mayr,1862)	SD	sa	SF	W	0.478	D	va	VF	W	0.220
MYRMICINAE										
Acromyrmex rugosus (Smith, F., 1858)	ND	va	VF	Y	0.043	ND	d	IF	Y	0.020

Where: Domin. = Dominance: SD = superdominant, D = dominant, ND = non-dominant. Abund. = Abundance: sa = superabundant, va = very abundant, a = abundant, c = common, d = disperse, r = rare. Freq. = Frequency: SF = super-frequent, VF = very frequent, F = frequent and IF = infrequent. Const. = Constancy: W = constant, Y = accessory and Z = accidental.* Dominance NOVOTNY, BASSET, (2000). %= relative frequency.

TAXON	Manual					*Pitfall*				
	*Domin.	Abund.	Freq.	Const.	%	*Domin.	Abund.	Freq.	Const.	%
Crematogaster distans (Mayr,1870)	ND	va	IF	Y	0.087	ND	va	VF	W	0.091
Cyphomyrmex rimosus (Spinola, 1851)	ND	r	IF	Y	0.005					
Pheidole sp. 1	ND	r	IF	Y	0.005					
Pheidole sp. 2	ND	r	IF	Y	0.005	ND	r	IF	Z	0.028
Pheidole sp. 6	ND	c	F	Y	0.022					
Pheidole sp. 11						ND	c	F	Y	0.008
Solenopsis invicta (Buren,1972)	ND	va	VF	Y	0.082	ND	c	F	Y	0.043
Wasmannia lutzi Forel, 1908						ND	r	IF	Z	0.004
PONERINAE										
Hypoponera sp.3						ND	r	IF	Z	0.008
Pachycondyla striata Smith, F., 1858	ND	r	IF	Y	0.005	ND	r	IF	Z	0.008
Neoponera crenata (Roger, 1861)	ND	r	IF	Y	0.005					
PSEUDOMYRMECINAE										
Pseudomyrmex sp. 1	ND	d	IF	Y	0.011	ND	c	F	W	0.071
Where: Domin. = Dominance: SD = superdominant, D = dominant, ND = non-dominant. Abund. = Abundance: sa = superabundant, va = very abundant, a = abundant, c = common, d = disperse, r = rare. Freq. = Frequency: SF = super-frequent, VF = very frequent, F = frequent and IF = infrequent. Const. = Constancy: W = constant, Y = accessory and Z = accidental.* Dominance NOVOTNY, BASSET, (2000). %= relative frequency.										

The ants *Linepithema micans* (Forel,1908), *Camponotus crassus* (Mayr, 1862), *Camponotus rufipes* (Fabricius, 1775), *Acromyrmex rugosus* (Smith, F. 1858), *Crematogaster distans* (Mayr, 1870) and *Solenopsis invicta* (Buren, 1972) occurred in both sampling methods. They were classified as non-dominant, very abundant, very frequent and accessory species in manual sampling, and as dominant, very abundant, very frequent and constant species in suspended *pitfall* trap. *Pseudomyrmex* sp.1 was non-dominant in both sampling methods, varying from disperse to common, infrequent to frequent and from accessory to constant.

The faunistic analysis indicated prevalence of non-dominant, rare, infrequent and accidental or accessory species in both sampling methods. Some of these species are exclusive for each sampling method. For those ants which presented such classification, seven out of nine were of exclusive occurrence for manual sampling, while in suspended *pitfall* traps four out of eight were exclusive. We found three non-dominant, common, frequent and accessory species in manual sampling: *Dorymyrmex brunneus* (Forel, 1908), *Camponotus* sp. 3 and *Pheidole* sp. 6. In suspended *pitfall* trap, four common and frequent with a variated constancy species occurred: *Pheidole* sp.11; *S. invicta*; *Hypoponera* sp.3 and *Pseudomyrmex* sp.1.

We observed higher Evennes's in the suspended *pitfall* trap (E = 0.8030) occurrences in comparison to manual sampling one (E = 0.6590). Part of the sampled fauna tended to occupy the upper spaces of the studied plant. For example, *C. melanoticus*, *Camponotus* sp.2, *Camponotus* sp.5, *Camponotus* sp.6, *Myrmelachista catharinae* Mayr, 1887, *Pheidole* sp.11, *Wasmannia* *lutzi* Forel, 1908 and *Hypoponera* sp.3, which were captured only with suspended *pitfall* traps. Other ant species as *D. pyramicus*, *Linepithema humile*, *L. pulex*, *Camponotus* sp.1, *M. arthuri*, *C. rimosus*, *Pheidole* sp.1, *Pheidole* sp.6 and *N. crenata* apparently occupied only the lowest strata of *C. xanthocarpa* trees, also indicated by the exclusive occurrence on the manual sampling method. We assumed that those species shared between two sampling methods, maintained activity for all over the studied plant.

Discussion

Our results revealed a diversified ant fauna visiting *C. xanthocarpa* trees, and to the best of our knowledge, this is the first study to identify the ants associated to this important plant species. Some ant species found in *C. xanthocarpa* trees were very abundant during all period of sampling in both studied strata, revealing potential interactions with the plant, which remains to be explored, considering our data as basis. Furthermore, our data revealed some extent of strata preference by part of the ant fauna studied, showing species which occurred exclusively on the higher or lower parts of the plant we studied. Our study also contributes to the understanding of ants from Mixed Ombrophillous Forest in the South region of Brazil (Junqueira et al. 2001; Maciel et al. 2011; Iasczczaki et al. 2021; Carneiro et al. 2022).

We were not capable of reach stabilization in the rarefaction curves of species in our study, indicating that our sampling effort was possibly not sufficient to collect all ant species, mainly those from the manual sampling method. Nonetheless, this is a quite common phenomena for ants, because these insects are naturally rich in species, presenting many rare species, and clustered distribution (Wilson, 1971Hölldobler and Wilson 1990; Oliveira and Pie 1998) what makes almost impossible to sample all the species present in a given habitat. With our sampling efforts, we revealed that Formicinae and Myrmicinae were the richest groups in our study. This is likely due to the high richness of these ant groups in the Neotropical region (Fernandez, 2003); varied nesting and food gathering strategies; they are of easy sampling (Ward 2014) and; the most diverse ant subfamilies in Mixed Ombrophilous Forests (Fernandez, 2003; Lopes et al. 2010; Lutinski et al. 2016; Franco and Feitosa 2018)).

We adopted two ant sampling methods in order to obtain better results in our faunistic analysis (Longino et al. 2002): one manual, active sampling method and a passive arboreal pitfall trap. We noticed that our methods complemented each other in terms of species collected, because some species occurred exclusively for each method. The active manual-sampling method increases the likelihood of finding species of hard access in micro-habitats in a given moment. In contrast, the suspended *pitfall* trap enhances the temporal window of sampling to 72 h from its set, revealing species in different periods of the day and night (Agosti 2000). We did not use baits to collect ants in our traps, because it could influence the results by selection of ants attracted to, and competition and/or exclusion between the species in the area (variety in feeding habits) (Hahn and Wheeler 2002). Therefore, we presumed low disturbance from our sampling method in the ant species obtained in *C. xanthocarpa* trees, with more realistic results (Yusah et al. 2012).

We noticed that the highest richness of ants was found between November and December in our study, and this period coincides with the flowering and fruit production period of *C. xanthocarpa* (HomczinskI et al. 2017). This process usually lasts between 15 and 20 days in *C. xanthocarpa* (Lisbôa et al. 2011), and it may have attracted more ant species and increased their activity on the plant due to nectar, pollen, fruits and other food sources (e.g., preys) present during this period (RICO-GRAY 1989; Rico-Gray 1993).

Non-dominant, rare, infrequent, accidental, or accessory species in the faunistic analysis were the majority in our study, and it might reflect ants that occasionally visit the studied plant. Some ant species, *D. pyramicus* for example, are visitor of nectaries (Cruz et al. 2019), and as aforementioned, part of our study was carried out during the flowering period of *C. xanthocarpa*. Moreover, many ant species are naturally rare in the environment (Pinto-Coelho, 2008): *C. rimosus* is characterized by having quite small nests with relatively few individuals (Wetterer 2012); *Hypoponera* sp.3 and *N. crenata* are both specialized predators' solitary-foraging species (Delabie et.al, 2015). Such characteristics can potentially explain the relatively low occurrence of these ants in our study.

We observed differences in species Evenness between the two sampling methods, but our results suggest that four ant species dominated the whole plant studied: *N. fulva*, *C. crassus*, *C. rufipes*, and *C. distans*. These ants probably determined the distribution of other species present in *C. xanthocapa* plants studied here and may potentially present interactions with the plant and the visiting fauna and, deserving close attention in future researches. We speculate this because ants from *Camponotus* and *Crematogaster* genera are frequently found as dominant in canopies (Floren and Linsenmair 1998) where they are aggressive towards competitors and can monopolize areas and food sources rapidly (Hölldobler and Wilson 1990). Beyond that, the canopy of *C. xanthocarpa* is elongated, complex and dense with small irregularities in its shape (Lorenzi 2008), and it may provide structure to sustain nests and prey for several ant species, including non-native ants as *N. fulva*. This is an exotic ant species strongly adapted to tropical or sub-tropical regions and may occupy a series of different places for building its highly populated nests (Kumar et al. 2015).

Many other arboreal ant species occurred in *C. xanthocarpa*, such as those from *Pseudomyrmex*, *Crematogaster* and *Pheidole* suggesting that this plant can function as an important refugia and host for nesting and foraging of these species. Futhermore, because *C. xanthocarpa* is a plant species responsible by a high leaf-litter production in Mixed Ombrophilous Forests, it may provide shelter, structure and microclimate for ant species common in leaf-litter or ground (Silva et al. 2011). Our study also revealed high abundance of occurrence of leaf-cutting ants, such as *A. rugosos*, and the constancy of this species along our samplings reveals herbivory activity on floral and vegetative tissues of *C. xanthocarpa*. Future studies should evaluate the probable impacts of the herbivory activity by these ants for the plant. Considering our results, these leaf-cutting ants may be potential pests for *C. xanthocarpa* trees (Goncalves 1957; Nobúa Behrmann et al. 2017).

The trees selected for this study were 50 years old or more and due to this age, the trees provide some other observable traits that may facilitate the occurrence of ant species not tipically observable in the canopies of trees. Thick and twisted trunks, for example, from which a quite complex canopy emerges with many branches where bryophytes, pteridophytes, and epiphytes grow freely, in addition to organic matter accumulated under the roots and mats that these plants form (pers. obs.). Such traits favor the occurrence of ants in the canopies, even those typically ground or leaf-litter nesting species such as *W. lutzi*, *Pheidole* spp., and *M. arthuri* (Longino and Nadkarni 1990; Yanoviak et al. 2011).

Our study provides important information and add to the understanding of ants and a particular plant species occurring in the South region of Brazil. Our findings can be used for improving the understanding of interactions between the studied plant and the ant fauna. This is relevant because *C. xanthocarpa* has economic importance for its region of occurrence and also plays ecosystem services for the Mixed Ombrophilous Forest (Marques, 2008). Thus, *C. xanthocarpa* supports ant species by providing food and shelter, and ants may protect the plant against herbivorous insects, as well as helping in nutrient cycling and perhaps pollination, but the impacts of leaf-cutting ants on this plant also needs more detailed studies. Therefore, the interaction between this tree species and the associated ant fauna has intrinsic importance in helping to maintain biodiversity as well as ecosystem balance (Marques, 2008).

Declarations

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Competing interests

The authors declare no conflicts of interest. This research requires no ethical approval for the use of animals.

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Figures

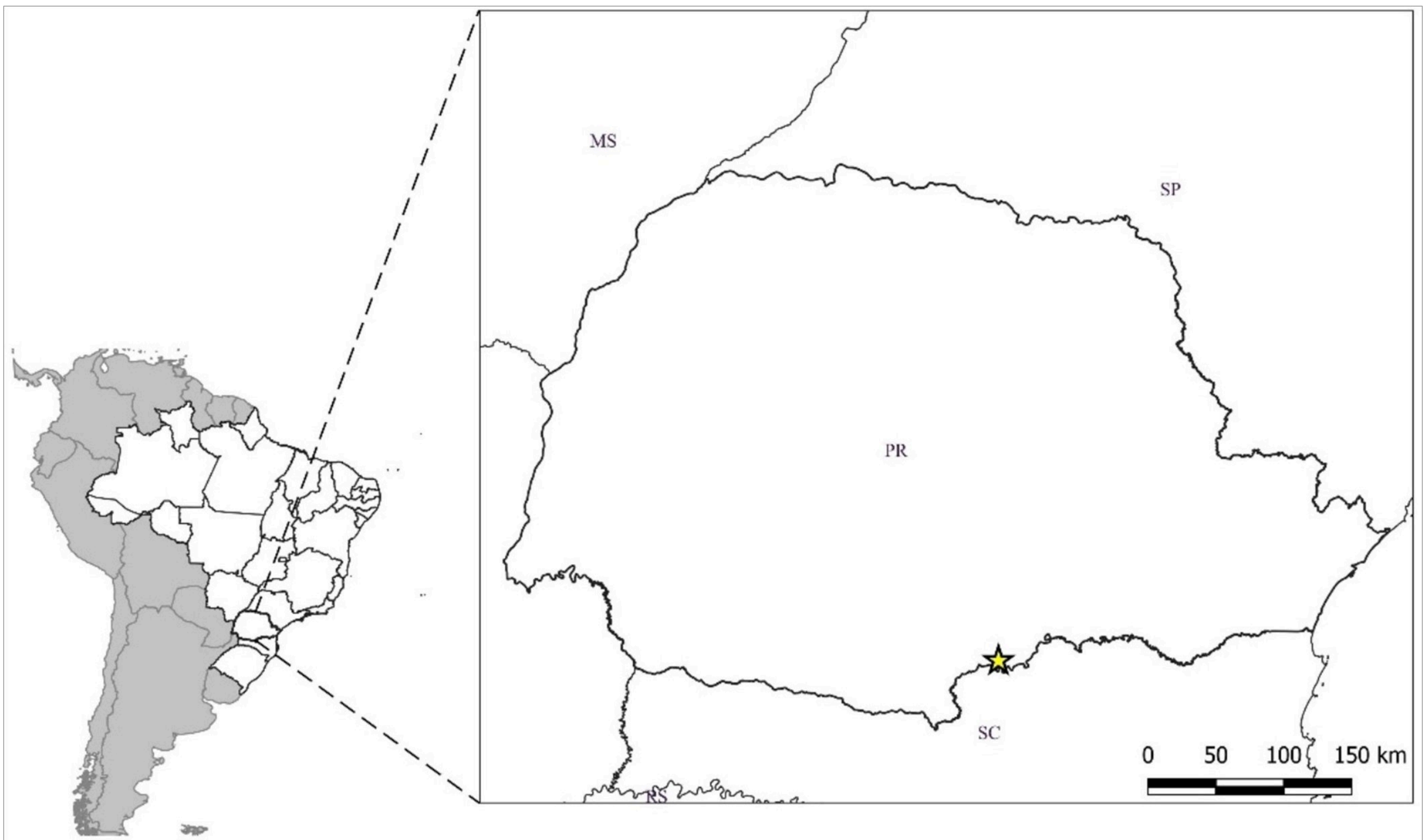

Figure 1

Study area (yellow star) where suspended *pitfall* traps and manual sampling were used to collect ants on *Campomanesia xanthocarpa* trees in the period between October 2017 and May 2018 (26°11'59.57"S 50°56'14.91"W, Paula Freitas, state of Paraná).

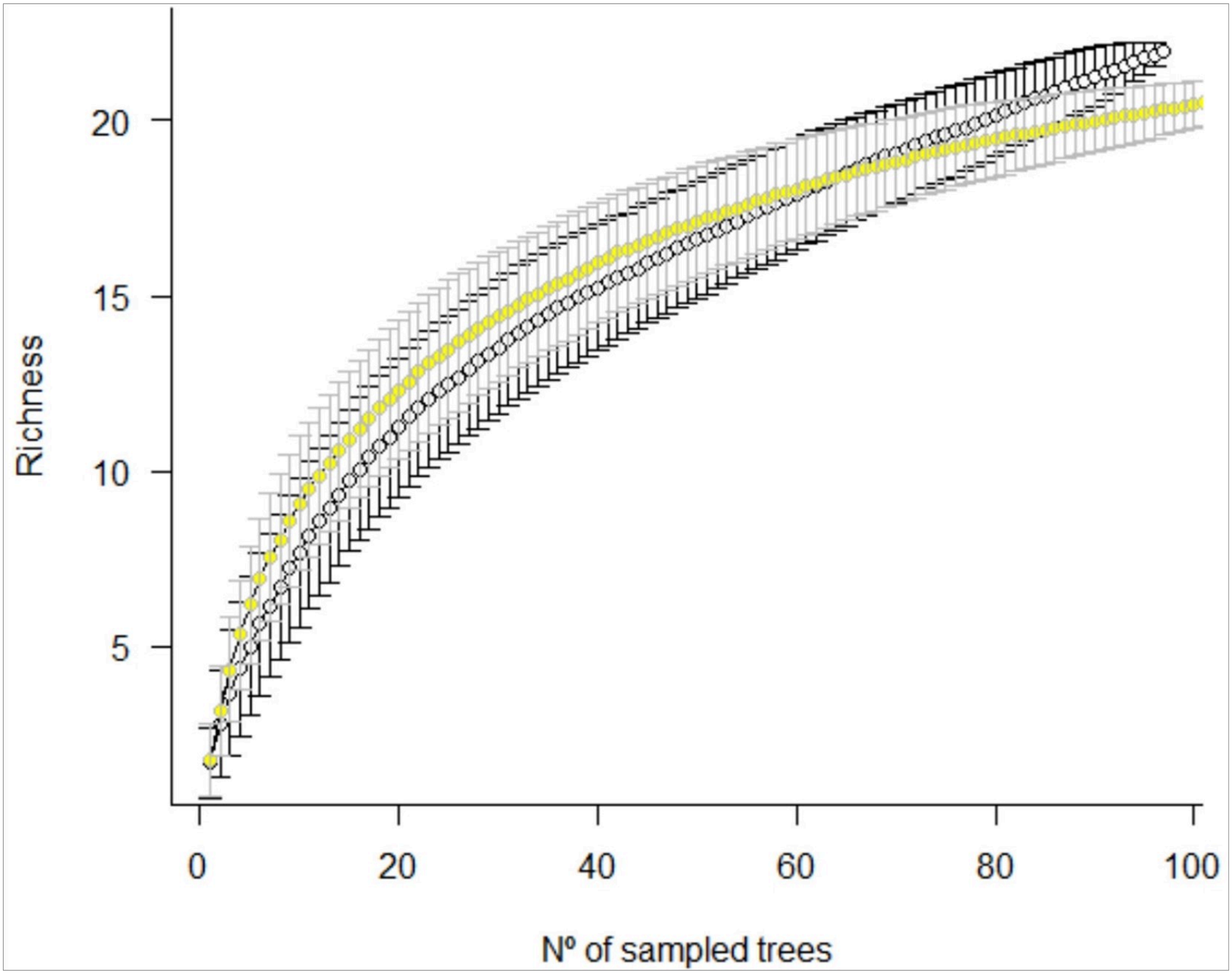

Figure 2

Rarefaction curve of ants sampled on *Campomanesia xanthocarpa* using manual sampling (black open circles) and suspended *pitfall* traps (yellow filled circles), in the period between October 2017 and May 2018, in an area located in the municipality of Paula Freitas, state of Paraná (26°11'59.57"S 50°56'14.91"W).