What do different landscapes of the Atlantic Forest reveal about the occurrence of *Discothyrea* Roger, 1863 (Formicidae: Proceratiinae)?

Claudia Tiemi Wazema¹, Victor Hideki Nagatani¹, Débora Rodrigues de Souza-Campana¹, Fabricio Severo Magalhães¹, Ricardo Sartorello² & Maria Santina de Castro Morini¹*

¹Universidade Mogi das Cruzes, Laboratório de Mirmecologia do Alto Tietê, Núcleo de Ciências Ambientais, Centro Cívico, Av. Dr. Cândido Almeida e Souza, nº 200, CEP 08780-911, Mogi das Cruzes, SP, Brasil.
²Universidade de Mogi das Cruzes, Laboratório de Mapeamento e Análise da Paisagem, Núcleo de Ciências Ambientais, Av. Dr. Cândido Xavier de Almeida de Souza, nº 200, CEP 08780-911, Mogi das Cruzes, SP, Brasil.
*Corresponding author: Maria Santina de Castro Morini, e-mail: mscmorini@gmail.com

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Abstract: *Discothyrea* is a genus composed of specialist predatory species rarely recorded and with little known biology. Specimen collection is usually associated with preserved native vegetation. In this work, we explore the landscape of sites with occurrence of *Discothyrea* seeking to improve knowledge about the natural history of this genus. Species of *Discothyrea* were recorded in ten Atlantic Forest sites. We analyzed the landscape around the place of occurrence of each species using a 500-m buffer. We classified the landscape as heterogeneous and homogeneous according to the percentage of natural (native vegetation), urban, and rural areas. We found 67 specimens of *Discothyrea*; 59 of them were *D. sexarticulata*, occurring in 88% of the fragments. There were also eight specimens of *D. neotropica* occurring in 12% of the fragments. The results show that *D. sexarticulata* can be found in homogeneous landscapes with anthropic influence (0-51% of rural area and 0-68% of urban area). *Discothyrea neotropica* is found in heterogeneous landscapes with a dominant presence of native vegetation (between 74-95%). The results improve knowledge on the biology of *Discothyrea* mainly in relation to the vicinity of occurrence sites. In addition, the results indicate that regional studies are important to understand species ecology.

Keywords: Hypogeic species; mosaic of landscapes; cryptic habit; conservation.

O que diferentes paisagens da Floresta Atlântica nos mostram sobre a ocorrência de *Discothyrea* Roger, 1863 (Formicidae: Proceratiinae)?

Resumo: *Discothyrea* é um gênero composto por espécies predadoras especialistas, raramente registradas e com biologia pouca conhecida. A coleta de espécimes geralmente está associada à vegetação nativa preservada. Neste trabalho exploramos a paisagem de locais com ocorrência de *Discothyrea*, buscando incrementar o conhecimento sobre a história natural do gênero. Espécies de *Discothyrea* foram registradas em dez áreas de Mata Atlântica. A paisagem ao redor do local de ocorrência de cada espécie foi analisada, usando um *buffer* de 500 m. Classificamos a paisagem em heterogênea e homogênea de acordo com a porcentagem de área natural (vegetação nativa), urbana e rural. Encontramos 67 espécimes de *Discothyrea*; 59 de *D. sexarticulata*, em 88% dos fragmentos. Oito espécimes de *D. neotropica*, em 12% dos fragmentos. Nossos resultados mostram que *D. sexarticulata* pode ser encontrada em paisagens homogêneas e sob influência antrópica, com 0-51% de área rural e 0-68% de área urbana; e *D. neotropica* em paisagens heterogêneas, com presença dominante de vegetação nativa (entre 74-95%). Nossos resultados trazem um aporte de conhecimento à biologia de *Discothyrea*, principalmente em relação às adjacências do local de ocorrência. Além disso, nossos resultados indicam que estudos regionais são importantes ferramentas para o conhecimento da ecologia das espécies.

Palavras-chave: Espécie hipogeica; mosaico de paisagens; hábito criptico; conservação.
Introduction

The genus Discothyrea Roger, 1863, has 50 species distributed in tropical areas of the southern hemisphere. The occurrence area extends from the midwestern and southeastern United States to northern Argentina. It is found mainly in the Neotropical Region, especially in tropical forest areas (Antweb 2020, Antwiki 2020). There are eight species recorded in the Neotropical Region (Sosa-Calvo & Longino 2007, Antmaps 2020), five in Brazil [Discothyrea denticulata Weber, 1939, D. horni Menozzi, 1927, D. humilis Weber, 1939, D. sexarticulata Borgmeier, 1954 and D. neotropica Bruch, 1919], and two in the State of São Paulo [D. sexarticulata and D. neotropica (Delabie et al. 2015, Antmaps 2020)].

The species are considered rare in litter probably because (1) the colonies have few individuals (Dejean & Dejean 1998, Katayama 2013, Delabie et al. 2015), (2) there is a limitation of sampling techniques (Hita- Garcia et al. 2019), and (3) the workers are very small, between 0.2 and 0.5 mm (Brandão et al. 2009). The species have a cryptic habit and the nests are generally inconspicuous (Zacharias & Dharma Rajan 2004), located in litter interstices, decaying logs (Brown 1958; Delabie et al. 2015), and under rocks (Bharti et al. 2015). Twigs from the fragmentation of tree branches are important for many ant species, especially for nesting. However, it is probably not a type of resource used by Discothyrea species in litter (see Fernandes et al. 2019a, b; 2020).

The species are specialist predators feeding almost exclusively on arthropod eggs, especially centipedes and spiders (Brown 1957, 1958, Dejean et al. 1999, Baccaro et al. 2015), but also on Tenebrio molitor Linnaeus, 1758, larvae (Wazema, personal communication). Discothyrea specimens can be collected in leaf litter using pitfall traps (Morini et al. 2007) and mainly in samples of leaf litter in forests with different phytophysionomies (Vasconcelos & Delabie 2000, Feitosa & Ribeiro 2005, Suguituru et al. 2013, Wazema et al. 2019) and at different altitudes (Santos 2008). There are records in Pinus elliottii (Pacheco et al. 2009) and eucalyptus crops (Monte et al. 2011, Suguituru et al. 2011), but these sites were surrounded by native vegetation. Lassau & Hocholi (2004) analyzed the response of ant communities to various physical and biological factors that occur in undisturbed places and recorded species of Discothyrea in low- and high-complexity habitats. In fragments of Atlantic Forest in the state of São Paulo, records of Discothyrea spp. are usually associated to preserved native vegetation (Suguituru et al. 2013, 2015).

The Brazilian Atlantic Forest has been reduced to immense archipelagos of tiny and widely separated forest fragments (Joly et al. 2014). Moreover, urban areas surround most fragments (Tabarelli et al. 2005), as well as areas with other anthropogenic activities (Ribeiro et al. 2009), such as extensive agriculture areas and eucalyptus and sugarcane crops. In a natural environment, changes caused by different land uses alter the landscape structure (e.g., by loss of biotic and abiotic resources), which in turn affect ant communities (Crist 2009). Ants are important components of the edaphic fauna (Decaëns 2010). They are considered good ecological indicators (Ribas et al. 2012, Casimiro et al. 2019), as their communities are influenced both on a local and a regional scale (Spiesman & Cumming 2008, Cumming 2011). Smaller and isolated fragments are more susceptible to species extinction and invasion by generalist species (Schoereder et al. 2004).

Generalist species affect habitats in sites with high proportions of matrix habitats in the surrounding landscape (Spiesman & Cumming 2008).

As the structure of vegetation and soil and related abiotic factors influence ant communities, the analysis of landscape fragments and their surroundings may help the implementation of conservation management plans or environmental planning (Lindenmayer et al. 2008, László et al. 2014). This is true especially when species are considered rare, as they play a fundamental role in the evolutionary adaptation of communities to changing land uses (László et al. 2014). In this work, we evaluated the landscape in the vicinity of Discothyrea occurrence sites seeking to fill gaps in knowledge about the biology of this genus. We hope to find Discothyrea in fragments of the Atlantic Forest surrounded mainly by native vegetation, as Discothyrea species are considered rare and specialized.

Materials and Methods

This study was conducted in ten sites in the following cities: São Paulo (Previdência Park), Mogi das Cruzes (Kimberly-Clark Reserve, Francisco Affonso de Mello Municipal Natural Park, Private Natural Heritage Reserve - Botujuru, and Leon Feffer Park), Mogi das Cruzes/Bertioga (Neblinas Park), Biritiba-Mirim (Biritiba-Mirim Dam), and Salesópolis (Ponte Nova Dam, Paraitinga Dam, and Ribeirão do Campo Dam) (Figure 1). All sites are part of the Atlantic Forest Domain in Southeast Brazil (Fiaschi & Pirani 2009, Colombo & Joly 2010). According to the Köppen classification, the region’s climate is mesothermal with dry winters (Cwb). The annual rainfall accumulation is 1,500 mm (Cptec-Inpe 2020).

Ants were collected on the litter between 2001 and 2019 using techniques such as mini-Winkler extractors (Suguituru et al. 2013, Wazema et al. 2019) and pitfalls (Morini et al. 2007). The identification was carried out using keys specific to this group (Borgmeier 1949, Fernández 2003, Jiménez et al. 2008, Eguchi et al. 2014, Xu et al. 2014, Bharti et al. 2015) and by comparison with specimens deposited at the Reference Collection of the Alto Tietê Myrmecology Laboratory (LAMAT-UMC) (Suguituru et al. 2015) of the University of Mogi das Cruzes, São Paulo, Brazil, where the vouchers of this work are deposited.

The landscape was characterized using a 500-m buffer for each species occurrence site (Figure 1). Each collection period has its own methods. Aerial images were obtained using the Landsat 8 Satellite (Bing aerial - Bing 2020 Microsoft Corporation Earthstar Geographics SIO, °Microsoft Corporation). Each buffer (n = 10) was categorized in (1) native vegetation, (2) rural, and (3) urban areas. Each class was quantified in m². The landscape of the surroundings of each collection site was classified as heterogeneous [area of native vegetation ≥ 50% (Figure 2a)] and homogeneous area [percentage of rural and urban areas ≥ 50% (Figure 2b)] (Moreira et al. 2015). The scale was 1:3,000. The software QGIS, version 2.18.19, was used (QGIS Development Team 2018). The linear models (GLM) with Poisson distribution (software R) were used to test differences in species occurrence among areas. The analyses were performed using the software Rstudio (R, version 3.6.1, R Core Team 2019) at a 5% significance level.
Figure 1. Location of collection sites. 1 - Previdência Park, 2 - Kimberly-Clark Reserve, 3 - Francisco Afonso de Mello Municipal Natural Park, 4 - Private Natural Heritage Reserve - Botujuru, 5 - Leon Feffer Park, 6 - Neblinas Park, 7 - Biritiba-Mirim Dam, 8 - Ponte Nova Dam, 9 - Paraitinga Dam, and 10 - Ribeirão do Campo Dam.

Figure 2. Characterization of the 500 m buffer and classes of the location where the species were collected. a - heterogeneous landscape (Neblinas Park, São Paulo city); and b – homogeneous landscape (Leon Feffer Park, São Paulo city).
Results and Discussion

The results show that *D. sexarticulata* is more common than *D. neotropica* in the Atlantic Forest areas of the São Paulo state. We collected 67 specimens belonging to the *Discothyrea*. *Discothyrea sexarticulata* (59 specimens) occurred in 88% of the sites, and *D. neotropica* (eight specimens) occurred in 12% (Table 1). This is probably due to the resilience of *D. sexarticulata*. The data also show that this species occurs in fragments where the surroundings have 27-92% of native vegetation, 0-51% of rural areas, and 0-68% of urban areas (Table 1). We thus suggest that *D. sexarticulata* inhabits fragments with heterogeneous surroundings comprising a higher percentage of native and homogeneous vegetation and where anthropogenic changes (e.g., urban areas and crops) are more marked. In contrast, *D. neotropica* was recorded in fragments with more preserved surroundings, with 74-95% of native vegetation, 5-26% of rural areas, and 0-1% of urban areas (Table 1). The results suggest that this species occurs in fragments of the Atlantic Forest with a heterogeneous adjacency and a higher percentage of native Atlantic Forest vegetation. However, Arcusa & Cicchino (2017) reported that *D. neotropica* also inhabits pastures in the Pampas Region, which are considered areas of low environmental complexity.

The species of *Discothyrea* are tiny and its eyes have only one ommatid (Brandão et al. 2009, Delabie et al. 2015). These characteristics and the presence of small legs (Brandão et al. 2009) should limit mobility to small extensions (Yates & Andrew 2011). Therefore, the location where the nest is found should be relevant to these species, especially a location with a greater variety of interstices (e.g., litter), which contributes to less energy expenditure during foraging (Kaspari & Weiser 1999). Adjacent areas must affect the fragment’s temperature and humidity (Lima-Ribeiro 2008), especially in areas considered small (Magnago et al. 2015). Changes in the natural environment can affect *Discothyrea* species at (1) a local scale, where the lack of humidity is a limiting factor for small species that forage in interstices of leaf litter (Kaspari 1996), and (2) a landscape scale, as landscape changes affect rare species more than common species (László et al. 2014). In this context, we suggest that *D. sexarticulata* may be more sensitive to changes than *D. neotropica*. However, the results of the analyses (Table 2) show that a larger number of samples is necessary, especially of *D. sexarticulata*.

Table 1. Collection site, landscape composition, and number of specimens according to *Discothyrea* species.

| Sites | Sites characterization | Coordinates | Landscape composition (%) | Landscape types | Number of specimens |
|-------|------------------------|-------------|---------------------------|-----------------|---------------------|
|       |                        | Latitude    | Longitude                 | Native vegetation | Rural area | Urban area | D. neotropica | D. sexarticulata |
| 1-Previdência Park | Urban park | 23°34'00"S | 46°43'00"W | 37 | 0 | 63 | Homogeneous | 1 |
| 2-Kimberly-Clark Reserve | Anthropized fragment of Atlantic Forest | 23°26'52"S | 46°14'48"W | 59 | 8 | 33 | Heterogeneous | 9 |
| 3-Francisco Affonso de Mello Municipal Natural Park | Conservation units | 23°29'22"S | 46°11'55"W | 92 | 8 | 0 | Homogeneous | 1 |
| 4-Private Natural Heritage Reserve - Botujuru | Conservation units | 23°28'59"S | 46°09'49"W | 49 | 51 | 0 | Homogeneous | 21 |
| 5-Leon Feffer Park | Urban park with Atlantic Forest native vegetation | 23°31'49"S | 46°13'26"W | 27 | 5 | 68 | Homogeneous | 1 |
| 6-Neblinas Park | Conserved Atlantic Forest area | 23°44'40"S | 46°09'43"W | 95 | 5 | 0 | Heterogeneous | 5 |
| 7-Biritiba Mirim Dam | Conserved Atlantic Forest fragment | 23°35'54"S | 46°05'06"W | 74 | 26 | 0 | Heterogeneous | 4 |
| 8-Ponte Nova Dam | Conserved Atlantic Forest fragment | 23°36'04"S | 45°58'10"W | 80 | 19 | 1 | Heterogeneous | 14 |
| 9-Paraitinga Dam | Conserved Atlantic Forest fragment | 23°31'28"S | 45°57'01"W | 51 | 49 | 0 | Heterogeneous | 2 |
| 10-Ribeirão do Campo Dam | Conserved Atlantic Forest fragment | 23°34'10"S | 45°49'57"W | 74 | 26 | 0 | Heterogeneous | 1 |

Specimens total: 8 59
Therefore, this study significantly contributes to the knowledge on the biology of Discothyrea. There are few studies on the natural history and behavior of species of this genus possibly because they have cryptic habits and small sizes. The results show that *D. sexarticulata* and *D. neotropica* occur in fragments with different surroundings, suggesting a greater resilience of *D. sexarticulata*. Our study indicates that landscape scales can be important structuring forces on local communities. We report relevant technical knowledge for future studies on landscape ecology and ant communities in Atlantic Forest areas. Furthermore, we demonstrate the importance of regional studies as a tool for understanding species ecology. As most of the Atlantic Forest is composed of forest fragments smaller than 50 ha (Ribeiro et al. 2009), corresponding to vegetation islands within a matrix with several types of anthropogenic activity (e.g., crops, paved roads, dense buildings, railroads, and mining), studies of this nature are highly relevant for the conservation of species.

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### Tables

**Table 2.** Summary of GLM with a transect-level random intercept (ten transects, one in each area). Landscape classes selected according to landscape composition (%) of areas: NV-U (Native vegetation + urban area), NV-R-U (Native vegetation + rural area + urban area) and NV-R (Native vegetation + rural area).

| Predictors | Estimated | Standard Error | Z   | p   |
|------------|-----------|----------------|-----|-----|
| Intercept  | -2.438    | 1.000          | 0.000 | 1.000 |
| NV-U       | 6.280     | 1.414          | 0.000 | 1.000 |
| NV-R-U     | 1.846     | 1.026          | 1.799 | 0.072 |
| NV-R       | 1.482     | 1.022          | 1.449 | 0.147 |

**Model: Occurrence of *D. sexarticulata* ~ Area + (1 | transect)**

| Predictors | Estimated | Standard Error | Z   | p   |
|------------|-----------|----------------|-----|-----|
| Intercept  | -1.830    | 5.718          | -0.003 | 0.997 |
| NV-U       | -4.912    | 8.086          | 0.000 | 1.000 |
| NV-R-U     | 1.790     | 5.718          | 0.003 | 0.998 |
| NV-R       | 1.848     | 5.718          | 0.003 | 0.997 |

**Model: Occurrence of *D. neotropica* ~ Area + (1 | transect)**
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