

STINGLEES BEES AND URBAN SPACES: AN INVESTIGATION OF THE CONDITIONS FOR ADAPTATION TO CITY BUILDINGS AND LANDSCAPING

ABELHAS NATIVAS E ESPAÇOS URBANOS: UMA INVESTIGAÇÃO DAS CONDIÇÕES DE ADAPTAÇÃO AS EDIFICAÇÕES E PAISAGISMO DAS CIDADES

ABEJAS NATIVAS Y ESPACIOS URBANOS: UNA INVESTIGACIÓN SOBRE LAS CONDICIONES DE ADAPTACIÓN A LAS EDIFICACIONES Y EL PAISAJISMO DE LAS CIUDADES

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RESUMO: As abelhas nativas sem ferrão são essenciais para a preservação dos ecossistemas, pois contribuem para a manutenção da biodiversidade e a estabilidade de fragmentos florestais. Informações sobre como essas espécies se comportam em ambientes urbanos, ainda são escassas no Brasil. Por essa razão a proposta do estudo foi investigar a ocorrência de abelhas sem ferrão na área urbana da cidade Realeza (PR), através de localização de ninhos e captura ativa e passiva. A amostragens foram realizadas entre outubro e dezembro de 2022, período de maior floração de espécies nectaríferas na região. As buscas ocorreram em cinco setores, abrangendo áreas públicas e privadas, sendo registrados aspectos da paisagem como espécies florais, características físicas das edificações e substratos de nidificação. Durante o levantamento, foram registrados 23 ninhos de *Tetragonisca angustula* e *Scaptotrigona depilis* mas também foram detectadas atividades de forrageamento de *Scaptotrigona bipunctata* e *Plebeia* spp. Os fragmentos florestais urbanos de Realeza enfrentam desafios para abelhas sem ferrão devido à urbanização e vegetação exótica. *Tetragonisca angustula* adapta-se a substratos artificiais, enquanto *Scaptotrigona depilis* depende de cavidades naturais. Casas de madeira favorecem a nidificação, e estratégias como abrigos artificiais, plantas nativas e práticas culturais sustentáveis são essenciais para conservar polinizadores e os serviços ecossistêmicos.

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Palavras-chave: Polinizadores. Fragmentos florestais. Ecossistema urbano.

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ABSTRACT: Stingless native bees are essential for the preservation of ecosystems as they contribute to the maintenance of biodiversity and the stability of forest fragments. Information on how these species behave in urban environments is still scarce in Brazil. For this reason, the aim of this study was to investigate the occurrence of stingless bees in the urban area of the city of Realeza (PR) through the localization of nests and active and passive capture methods. Sampling was conducted between October and December 2022, a period of peak nectar-producing species flowering in the region. The searches were carried out in five sectors, covering both public and private areas, and aspects of the landscape such as floral species, physical characteristics of buildings, and nesting substrates were recorded. During the survey, 23 nests of *Tetragonisca angustula* and *Scaptotrigona depilis* were recorded, and foraging activities of *Scaptotrigona bipunctata* and *Plebeia* spp. were also detected. The urban forest fragments of Realeza face challenges for stingless bees due to urbanization and exotic vegetation. *Tetragonisca angustula* adapts to artificial substrates, while *Scaptotrigona depilis* depends on natural cavities. Wooden houses favor nesting, and strategies such as artificial shelters, native plants, and sustainable cultural practices are essential for conserving pollinators and ecosystem services.

Keywords: Pollinators. Forest fragments. Urban ecosystem.

RESUMEN: Las abejas nativas sin aguijón son esenciales para la preservación de los ecosistemas, ya que contribuyen a la mantención de la biodiversidad y la estabilidad de los fragmentos forestales. La información sobre cómo estas especies se comportan en ambientes urbanos aún es escasa en Brasil. Por esta razón, la propuesta del estudio fue investigar la presencia de abejas sin aguijón en la zona urbana de la ciudad de Realeza (PR), mediante la localización de nidos y captura activa y pasiva. El muestreo se realizó entre octubre y diciembre de 2022, período de mayor floración de especies nectaríferas en la región. Las búsquedas se llevaron a cabo en cinco sectores, que abarcaron áreas públicas y privadas, registrándose aspectos del paisaje como especies florales, características físicas de las edificaciones y sustratos de nidificación. Durante el levantamiento, se registraron 23 nidos de *Tetragonisca angustula* y *Scaptotrigona depilis*, pero también se detectaron actividades de forrajeo de *Scaptotrigona bipunctata* y *Plebeia* spp. Los fragmentos forestales urbanos de Realeza enfrentan desafíos para las abejas sin aguijón debido a la urbanización y la vegetación exótica. *Tetragonisca angustula* se adapta a sustratos artificiales, mientras que *Scaptotrigona depilis* depende de cavidades naturales. Las casas de madera favorecen la nidificación, y estrategias como refugios artificiales, plantas nativas y prácticas culturales sostenibles son esenciales para conservar a los polinizadores y los servicios ecosistémicos.

Palabras clave: Polinizadores. Fragmentos forestales. Ecosistema urbano.

INTRODUCTION

The expansion of arable areas has reduced food sources and nesting sites for stingless bees, exposing these insects to direct contact with toxic substances such as pesticides and herbicides (SHIMIZU M and MOURÃO MAN, 2022). The loss of natural areas has forced species to migrate, occupy, and adapt to anthropized environments (MAY-ITZÁ WJ, et al., 2021), making it very common to observe native bees in cities nesting in hollow trees, roof crevices, electrical boxes, door frames, and wall holes (HENRIQUE dos SANTOS L, et al. 2023).

Urbanization is an activity characterized by the expansion of artificial constructions and the irreversible reduction of natural green areas, altering the dynamics of ecological services (WANG W, et al., 2020). In recent years, it has been suggested that urban environments may not be as restrictive for the occurrence of stingless bees, but such considerations must take into account scale effects, such as analyzing overall urban landscaping and small areas within that landscape, like blocks and backyards (ARAÚJO S and WIT NGMP, 2020). The success of native bees in urban environments depends on the availability of food (nectar, pollen, and water), places and materials for nesting (mud, oils, and resins), and intrinsic factors such as generalist and specialist behaviors and geographic distribution (PFEIFFER V, et al., 2023). The level of alteration in urban ecosystems impacts the complexity of city microhabitats, resulting in changes to the substrate base for nests and consequently leading to an increase in some species and local extinction of others (PRENDERGAST KS, et al., 2020).

To minimize the impacts of urbanization on fauna and flora, it is necessary to promote urban backyards and parks with diverse vegetation, which can provide floral and nesting resources as tools for pollinator conservation in cities (DANIELS B, et al., 2020). Urban backyards can offer heterogeneous shelters to be used as refuges for native stingless bees, aiding in the maintenance of natural populations (ANDERSON M., et al., 2023). These spaces can play a strategic conservationist role in preserving and increasing native bee populations, contributing to the balance of urban flora and fauna (FELIPPSEN EA, et al., 2021).

Bees have highly specific responses to their environment, and disturbances can be reflected in changes in foraging and nesting behaviors. Stingless bees build cryptic nests in natural locations that can persist for years. However, in urban settings, short-term landscape transformations can expose nests, making colonies vulnerable (AYERS AC and REHAN SM, 2023). Therefore, studying the occurrence of native bees can reveal valuable information about the level of anthropization (SOUTHER SK, et al., 2024) and the directions of urban development needed to promote more sustainable cities.

Urban areas with green spaces and gardens provide floral resources and nesting substrates, which can help maintain the ecosystem services offered by these insects. For this reason, the focus of the research was to investigate the occurrence of native species in public squares and backyards, and the factors influencing the presence/absence of colonies based on

physical characteristics (construction type) and phytophysognomic features (gardens, parks, forest remnants) of residential areas.

METHODS

Characterization of Study Area

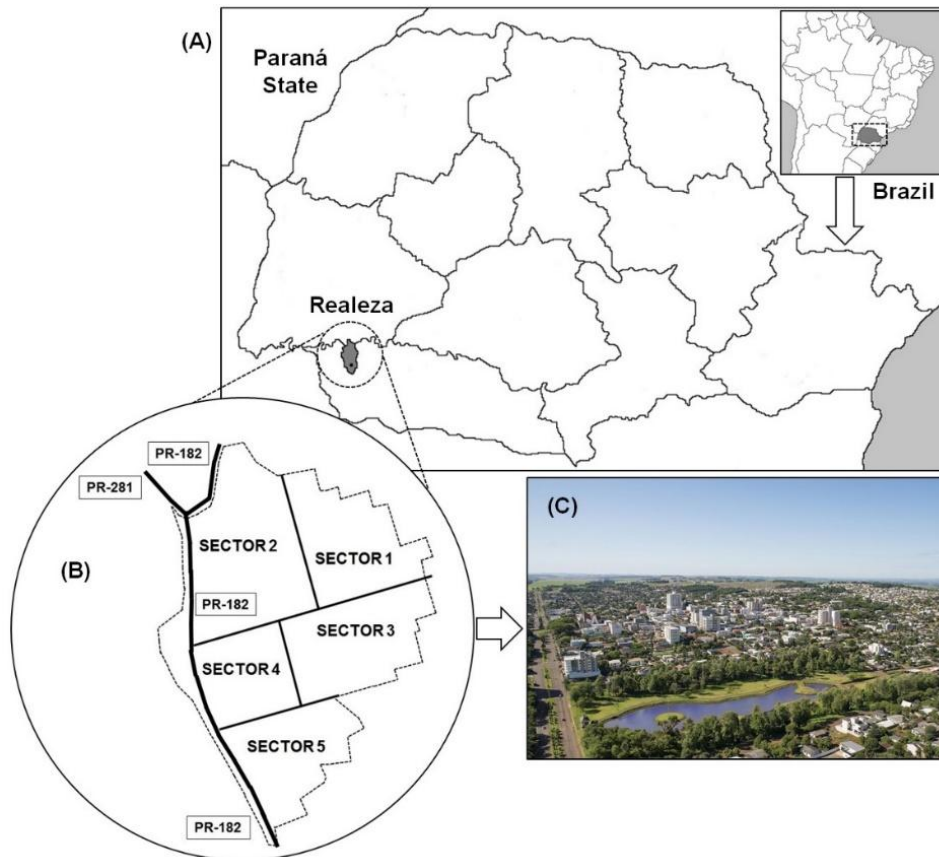
The study was conducted in the urban area of Realeza (coordinates 25°46'08"S, 53°31'57"W), a small city in southwestern Paraná, Brazil, with an area of 353.416 km² (IBGE, 2022) and an estimated population of 16,976 inhabitants, of which 72.2% reside in urban areas and 27.8% in rural areas (IBGE, 2019).

The region's climate is humid subtropical, mesothermal, with annual rainfall ranging between 1,600 mm and 1,900 mm and temperatures varying from 18°C in the coldest months to above 22°C in the warmest months (ALVARES CA, et al., 2013). The city's landscape is part of the Atlantic Forest Biome and is located in a phytogeographic transition zone between Mixed Ombrophilous Forest and Semideciduous Seasonal Forest (IBGE, 1992).

Data from the 2017 Agricultural Census on land use and occupation show that 15,375 hectares are used for crops, 10,666 hectares for pastures, 6,618 hectares consist of forests, and 78 hectares are agroforestry systems (IBGE, 2017). This configuration corresponds to the typical landscape of southwestern Paraná, described as an agricultural matrix interspersed with small forest fragments, resulting in the progressive loss of native biodiversity over the years (IAT, 2023).

Within the urban boundaries, the landscape is characterized by 76.8% of urban households and public roads with tree cover (IBGE, 2010). The city center features wide avenues with small to medium-sized trees and buildings primarily used for consumer goods and commercial services. On the outskirts, secondary roads are lined with larger vegetation, and small forest fragments can be found, some of which have been designated for the development of groves and parks (Figure 1).

Figure 1 - Characterization of study area: (A) Realeza city localization in the Paraná state; (B) Division Sectors: Sector 1 - São José, Loteamento Prandes e Cosaca; Sector 2 - Centro, Industrial e Nossa Senhora Aparecida; Sector 3 - Centro Cívico, João Paulo I e João Paulo II; Sector 4 - Cidade Universitária, Marchese e Prandes II; Sector 5 - Araxá, Universidade Federal da Fronteira Sul (UFFS), Jardim Primavera, Cohapar e Zanchet; (C) Overview of the city's landscaping and buildings.



Source: Adapted from maps of Wikipedia/Wikimedia/Open Street View, 2020; Google Earth, 2022; Viaje Paraná, 2023. PR-state highway.

Sampling Design

The sampling units for the urban zone of Realeza were established based on the division used for selective waste collection services, delineating five sectors that include both central and peripheral areas. To prevent overlap of surveyed areas, a distance of 1000 to 1500 meters was maintained between the regions selected for sampling within each sector. In these regions, five sampling points were defined, spaced more than 100 meters apart (urban spaces with vegetated backyards, and structures made of wood and masonry), with a survey radius of approximately 50 meters to investigate the occurrence of native bees according to the characteristics of the buildings and local landscaping.

The surveys for locating nests were conducted in private areas (inside backyards) and public areas (surrounding backyards, sidewalks, streets, nearby central flowerbeds). Sampling

was concentrated from October to December 2022, during the transition between spring and summer. Data collection and selection frequency occurred weekly, between 9:00 and 12:00 and from 15:00 to 17:00, as suggested by Souza-Júnior JBF, et al. (2019), as the period of greatest activity for stingless bees, when they leave the nest to forage. Nest location was performed through active search with a sampling effort of 5 hours/day, totaling 200 hours, with backyards visited three times a week. The search focused on areas more likely to host native bees, such as fallen trunks, thick tree trunks with visible cavities, rocks on the ground near roots, and masonry structures with fissures or holes.

During the searches, records were made of the floral species available in the backyards and their surroundings, as well as the physical characteristics of the sampled buildings. A field sheet was created for data recording. The analysis of the physical composition of the buildings and the vegetation composition of the backyards and surrounding areas considered the parameters (dimensions and indicators) shown in Table 1.

Table 1- Assessment parameters for the characterization of sampling areas.

Dimensions	Indicators
1. Configuration of forest fragments.	- Vegetation composition (native and exotic species); - Plant size (height and structure of species); - Type of vegetation (natural or altered); - Proximity to fragments.
2. Landscaping and garden structure (backyard and surroundings).	- Vegetation composition (native and exotic species); - Plant size (height and structure of species); - Garden complexity (simple or diversified).
3. Building typology.	- Masonry construction; - Wood construction; - Mixed construction.
4. Space delimitation.	- Masonry walls; - Wooden fences.
5. Flowering.	- Flowering plant species; - Number of flowering plant species.
6. Nesting.	- Number of nests; - Nesting substrate (natural or artificial).

Source: Pereira DC, et al., 2024.

Passive capture traps made from PET bottles, with volumes of 2 and 5 liters and containing a propolis attractant, were installed in areas with potential for stingless bee occurrence when no nests were observed (Figure 2). The baits were distributed in the arboreal stratum, considering the volume in relation to the intrinsic characteristics of colony size and nesting form, in order to avoid pre-selection of species. The locations for placing the traps considered direct sunlight exposure, with the devices being placed in large or medium-sized

trees or buildings in partial shade, allowing for support with ties. Capture inspections were conducted biweekly, and the baits were left in place for a period of 45 to 60 days, which was considered sufficient for the entry and establishment of a new colony (CETAP, 2023).

Figure 2 - PET-type traps installed on tree trunks for the passive capture of native bees.



Source: Pereira DC, et al., 2024.

Regardless of the method of specimen capture (active or passive), five randomly selected individuals were always collected using an entomological aspirator. The specimens were transferred with the aid of forceps to Falcon tubes (15 mL), containing cotton soaked in toxic liquid (ether), properly labeled with provenance data. The bees were temporarily stored in a humid chamber container to prevent stiffening until pinning and identification. The samples were taken to the Entomology Laboratory at the Universidade Federal da Fronteira Sul (UFFS), where taxonomic identification was performed based on taxonomic keys (ENGEL MS, et al., 2023; COSTA, 2019; SILVEIRA FA, et al., 2002). The data obtained were organized into graphs and tables using Excel software for descriptive data analysis.

Multivariate Analysis

Principal Component Analysis (PCA) was performed to inspect possible distribution relationships of the nest registration data across urban sectors, with the physical structure of residences, local landscaping, nearby forest fragments, and the occurrence of flowering. The PCA was conducted using the Paleontological Statistics (PAST) software, version 4.02 (Hammer Ø, et al., 2001), with the data being previously normalized using the following formula:

$$Z = \frac{X - \mu}{\sigma}$$

Where:

Z is the Z score (standardized value);

X is the raw or original value of the data;

μ is the mean of the data distribution;

σ is the standard deviation of the data distribution.

This normalization was used to facilitate comparisons of different data distributions or to combine data from different scales in one analysis, so that when $Z=0$, the raw value is equal to the mean; a positive Z indicates that the raw value is above the mean, and a negative Z indicates that the raw value is below the mean.

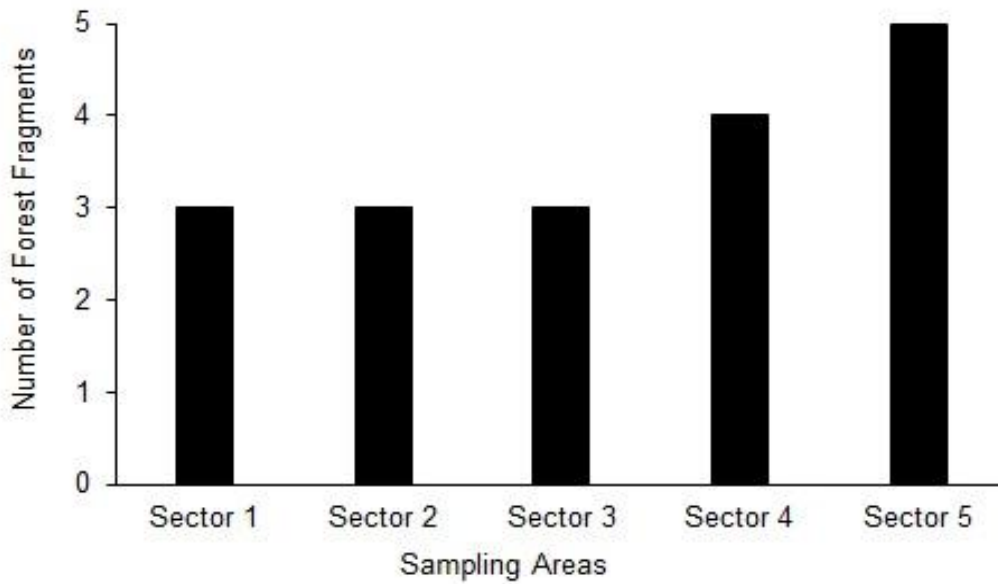
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RESULTS

Characterization of Forest Fragments and Landscaping of Backyards

The study revealed that the sampled urban area contains 18 forest fragments with potential for stingless bee refuge (Figure 3). Due to urbanization, the forest fragments in the city of Realeza are small and isolated, distributed among residential, commercial, and public spaces. This configuration hinders the formation of ecological corridors, which may affect the regeneration of areas designated for native vegetation restoration, species flow, and the overall health of the urban ecosystem.

Figure 3 - Number of forest fragments recorded in the sampling areas



Source: Pereira DC, et al., 2024.

The urban forest fragments predominantly feature species typical of the seasonal semideciduous forest and native trees adapted to the Atlantic Forest biome. Many of these species are medium-sized and belong to secondary regeneration stages, with their vegetation composition altered by the introduction of ornamental and exotic species traditionally used in urban landscaping in various cities across southwestern Paraná (Table 2). Natural spaces are confined to the city's surroundings, forming small belts or internal plots in the form of parks, smaller reserves, or areas of mandatory preservation, such as Permanent Preservation Areas (APPs). The urban landscape is characterized by intermediate spaces, concentrating forest fragments that serve as refuges for biodiversity impacted by habitat loss in the surrounding rural areas. However, urban expansion and inadequate management have reduced these formations to small, increasingly isolated vegetation islands, diminishing connectivity between fragments and compromising essential ecological processes, such as pollination and seed dispersal.

Table 2 - Main plant species present in the forest fragments of the urban area.

Botanical Family	Native Species (Scientific and Common Names)
Araucariaceae	<i>Araucaria angustifolia</i> (Bertolini) Kuntze, 1898 (Pinheiro-do-Paraná)
Bignoniaceae	<i>Handroanthus albus</i> (Chamisso) Mattos, 1948 (Ipê-amarelo)
Bignoniaceae	<i>Handroanthus impetiginosus</i> (Martius ex Candolle) Mattos, 1838 (ipê-roxo)
Anacardiaceae	<i>Schinus terebinthifolia</i> Raddi, 1820 (Aroeira-pimenteira)
Fabaceae	<i>Libidibia ferrea</i> (Martius ex Tulasne) Queiroz, 2009 (Pau-ferro)
Myrtaceae	<i>Campomanesia xanthocarpa</i> (Martius) Berg, 1857 (Guabiroba)
Sapindaceae	<i>Allophylus edulis</i> (Saint-Hilaire, Jussieu & Cambessèdes) Radlkofe1825 (Chal-chal)
Fabaceae	<i>Peltophorum dubium</i> (Sprengel) Taubert, 1892 (Canafistula)
Myrtaceae	<i>Eugenia uniflora</i> Linnaeus, 1753 (Pitanga)
Meliaceae	<i>Cedrela fissilis</i> Vellozo, 1825 (Cedro)
Lauraceae	<i>Ocotea puberula</i> (Richard) Nees, 1836 (Canela)
Exotic Species (Scientific and Common Names)	
Bignoniaceae	<i>Tecoma stans</i> (Linnaeus) Jussieu ex Kunth, 1818 (Ipê-de-jardim)
Fabaceae	<i>Cenostigma pluviosum</i> var. <i>peltophoroides</i> (Benth) Gagnon & Lewis, 2016 (Sibipiruna)
Strelitziaceae	<i>Ravenala madagascariensis</i> Sonnerat, 1782 (Árvore-do-viajante)
Arecaceae	<i>Archontophoenix cunninghamiana</i> Wendland & Drude, 1875 (Palmeira-real)
Bignoniaceae	<i>Jacaranda mimosifolia</i> Don, 1822 (Jacaranda)
Oleaceae	<i>Ligustrum lucidum</i> Linnaeus, 1758 (Ligustro ou Alfeneiro)
Lythraceae	<i>Lagerstroemia indica</i> (Linnaeus) Persoon, 1806 (Resedá)
Chrysobalanaceae	<i>Licania tomentosa</i> (Benth) Fritsch, 1935 (Oiti)
Chrysobalanaceae	<i>Ficus benjamina</i> Linnaeus, 1753 (Ficus)

Source: Pereira DC, et al., 2024.

The highest concentration of green areas was found in sectors 4 and 5, where urban subdivisions are more recent and located near one of the city's exit routes. The city's older sectors (1, 2, and 3) contain fewer forest units (three each), which tend to be smaller in size but exhibit a more heterogeneous profile. In these older sectors, nests of *Scaptotrigona depilis* (Moure, 1942) and *Tetragonisca angustula* (Latreille, 1811) were identified, species known for their adaptability to urban environments. Although a higher number of stingless bee species were expected in sectors 4 and 5 due to the larger number of forest fragments (four and five units, respectively), no nests were found. However, in sector 5, foraging activity was detected for species such as *Scaptotrigona bipunctata* (Lepelletier, 1836) (Tubuna), *Plebeia* sp. Schwarz, 1938 (Mirins), and *Apis mellifera ligustica* Spinola, 1806, and *Apis mellifera mellifera* Linnaeus, 1758 (Italian and German honey bees, respectively).

In residential environments, there is a tendency to use exotic ornamental plants for landscaping in yards. The botanical profile of domestic spaces and surrounding areas revealed the presence of four native species and 14 exotic species used in landscaping (Table 3). During sampling, it became evident that gardens with a higher number of flowering species exhibited greater foraging activity by stingless bees, particularly *T. angustula*.

Table 3 - Main plant species used in landscaping the backyards in the sampling areas.

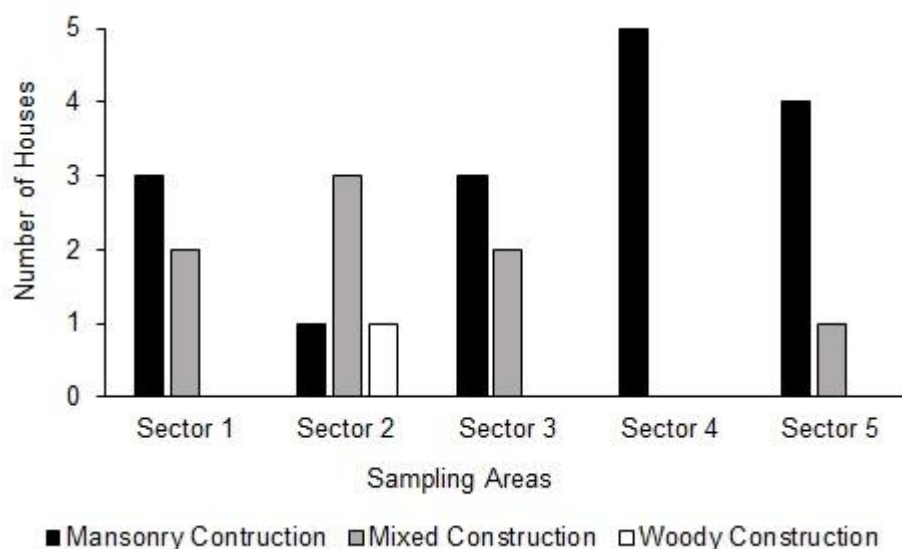
Botanical Family	Native Species (Scientific and Common Names)
Nyctaginaceae	<i>Bougainvillea glabra</i> Linneaus, 1753 (Primavera)
Orchidaceae	<i>Cattleya labiata</i> Lindley, 1824 (Orquídea-comum ou Rainha-do-sertão)
Poaceae	<i>Stenotaphrum secundatum</i> Aiton, 1811 (Gramma-santo-agostinho)
Poaceae	<i>Paspalum conjugatum</i> Raddi, 1820 (Capim-azedo)
Exotic Species (Scientific and Common Names)	
Acanthaceae	<i>Thunbergia alata</i> Thunberg, 1794 (Amarelinha)
Apiaceae	<i>Daucus carota</i> Linneaus, 1753 (Capim-cenoura-brava)
Arecaceae	<i>Phoenix dactylifera</i> Linnaeus, 1753 (Tamareira-de-jardim)
Asteraceae	<i>Leucanthemum vulgare</i> Lamarck, 1779 (Margarida)
Begoniaceae	<i>Begonia semperflorens</i> Linneaus, 1753 (Begonia)
Commelinaceae	<i>Tradescantia pallida</i> (Rose) Hunt, 1975 (Coração-roxo)
Fabaceae	<i>Delonix regia</i> (Kalm) Hooker, 1832 (Flamboyant)
Lamiaceae	<i>Lavandula</i> sp Linneaus, 1753 (Lavanda)
Liliaceae	<i>Lilium</i> sp Linneaus, 1753 (Lírio)
Magnoliaceae	<i>Magnolia grandiflora</i> Linnaeus, 1759 (Magnólia-branca)
Malvaceae	<i>Abutilon striatum</i> Candolle, 1824 (Lanterninha chinesa)
Musaceae	<i>Musa acuminata</i> Colla, 1820 (Banana-prata)
Poaceae	<i>Zoysia japonica</i> Steudel, 1854 (Gramma-esmeralda)
Rosaceae	<i>Rosa</i> spp. (Rosa-comum)

Source: Pereira DC, et al., 2024.

Residential Buildings Typologies

The results for the building typologies showed variation in the materials used for residential construction, with three types of buildings detected: wood, masonry, and mixed (Figure 4), all containing masonry walls delimiting the private space.

Figure 4 - Building typologies in the sampled areas.



Source: Pereira DC, et al., 2024.

In sectors 1, 2, and 3, where the presence of nests was recorded, there was one wooden house, six mixed houses, and 18 masonry houses, showing a trend toward more modern buildings with low permeability to the entry of stingless bees into the internal environments. In the wooden house, two nests of *T. angustula* were detected, one in the door frame and the other in a crack in the baseboard of the house. In the mixed and masonry houses, colonies of *T. angustula* and *S. depilis* were found mainly in wall cracks and tree trunk cavities. These results suggest that wooden houses have a higher probability of containing hives within the structure of the house itself, as in other structures, the internal permeability of the buildings is lower for the nesting process, limiting the occurrence of stingless bees to external residential environments. Permeability can be understood as the ability of an environment to allow circulation, feeding, and nesting of stingless bees. It is a metric that considers the density of native vegetation, the number of ecological corridors, the distance between habitat fragments, and safe shelters for hive installation—factors that are critical for the pollination process.

Number of plants in the flowering stage

During the sampling period, between 22 and 38 plants in the flowering stage were recorded per sector, including tree vegetation, shrubs, ground herbaceous plants, and small to medium-sized ornamental native and exotic plants, totaling 126 plants with active floral whorls (Table 4). The data show a relatively wide distribution, with most sectors having between 22 and 24 plants, while sectors 3 and 5 have significantly higher values. Sectors 1, 2, and 3 show a certain uniformity in the number of plants and environmental conditions, while sectors 4 and 5 maintain a more diverse vegetative structure, with a higher number of native plants, as these areas are more recently occupied. This variability between sectors may be associated with environmental factors, such as soil quality and usage, light availability, the level and form of urbanization, and differences in the management of each area.

This description helps to identify patterns and compare sector performance in terms of flowering, providing a starting point for more detailed analyses that can predict the level of permeability and support capacity for stingless bees.

Stingless Bee Nesting

A total of 23 nests were located at points in sectors 1, 2, and 3 (Table 4), represented by specimens of *T. angustula* (Jataí) and *S. depilis* (Canudo). Among the 18 nests of *T. angustula*

located, 15 were found within the backyards, and three in surrounding trees, six on natural substrates and nine on artificial ones (walls and wooden fences). Three other nests were found in the surroundings of the backyards, on natural substrates (tree cavities). The number of *S. depilis* nests was much lower compared to *T. angustula*, as only five nesting sites were recorded, four of which were installed in cavities of old, large tree trunks, two inside the backyards on artificial substrates (walls), and two on natural substrates (tree cavities). In the surroundings of the backyards, three nests were also recorded, but in older trees.

When comparisons are made regarding the occurrence of these species according to their colonization preferences, *T. angustula* clearly shows a preference for backyards, possibly due to the presence of cultivated vegetation or structures favorable for nesting. *S. depilis* demonstrates greater flexibility, with records both in the backyards and in the surroundings, though in smaller numbers.

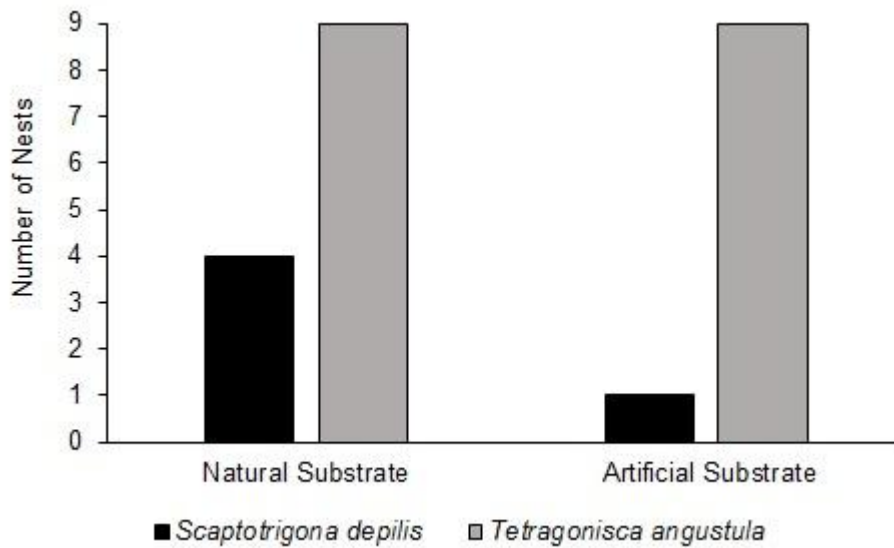
Table 4 - Number of stingless bee nests and flowering plants in the sampling areas.

Sampling Area	Number of Nests				Total of Nests	Number of Flowering Plants
	<i>S. depilis</i>		<i>T. angustula</i>			
	Backyard	Surroundings	Backyard	Surroundings		
Sector 1	1	1	5	0	7	22
Sector 2	1	0	6	3	10	22
Sector 3	0	2	4	0	6	34
Sector 4	0	0	0	0	0	24
Sector 5	0	0	0	0	0	38
Total	2	3	15	3	23	126

Source: Pereira DC, et al., 2024.

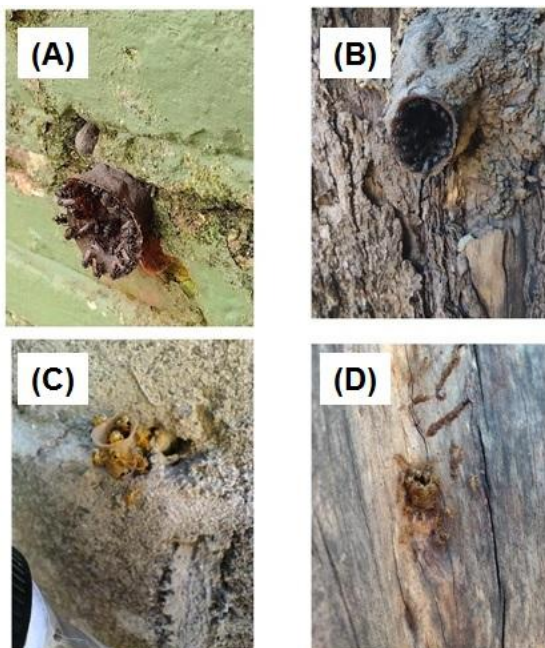
The results of the observations regarding the substrates used for nesting demonstrated the versatility of *T. angustula* in using artificial or natural shelters, while *S. depilis* seems to adapt better to natural shelters (Figure 5 e 6). The bee *S. depilis* showed a behavior of greater dependence on specific characteristics of the natural environment for nesting (presence of cavities in trees, trunks, or other wooden structures), as there is a lower availability of suitable artificial substrates for this species. The equivalent proportion of nests of *T. angustula* in both types of substrate showed its flexibility in substrate choice, equally using natural and artificial materials to build its nests. Furthermore, field observations recorded the ease with which *T. angustula* infiltrates artificial structures in the urban environment, such as cracks, fissures, and cavities in walls, fences, posts, or other masonry structures.

Figure 5 - Nesting substrate types for stingless bee.



Source: Pereira DC, et al., 2024.

Figure 6 - Substrates used for nesting by stingless bees in urban environments: (A) *Scaptotrigona depilis* in artificial substrate; (B) *Scaptotrigona depilis* in natural substrate; (C) *Tetragonisca angustula* in artificial substrate; (D) *Tetragonisca angustula* in natural substrate.



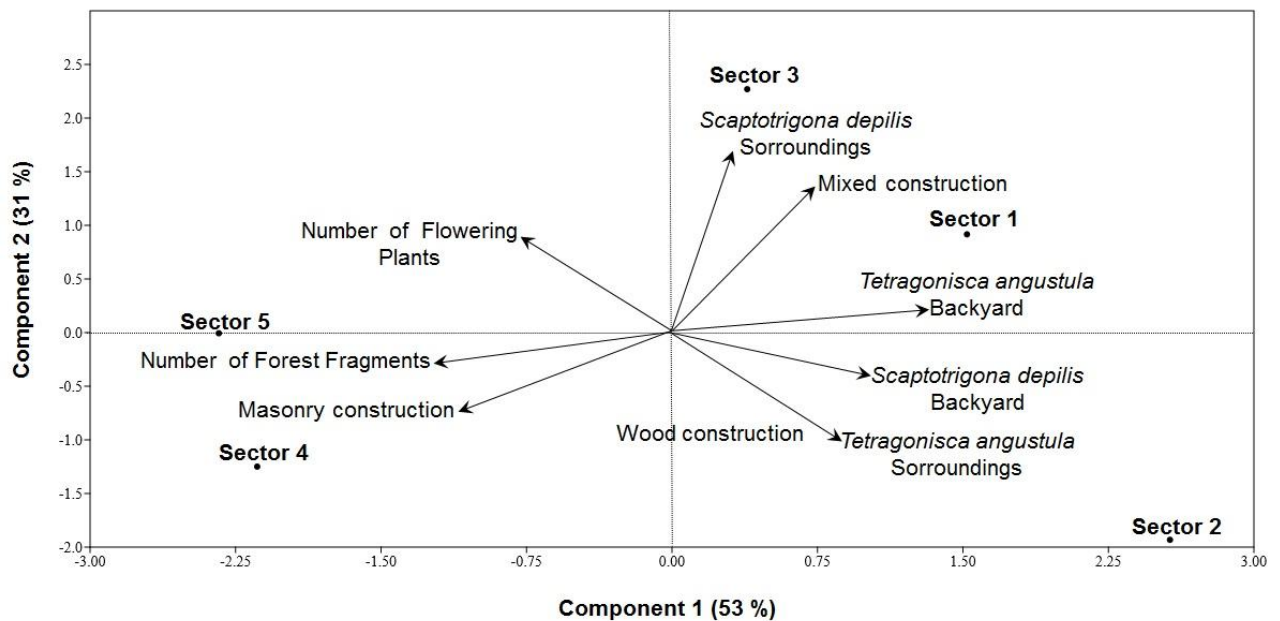
Source: Pereira DC, et al., 2024.

Species distribution analysis

The ordination of sampling points in the Principal Component Analysis (PCA) and associated variables such as species occurrence in the sectors, number of nests inside the yard

and surroundings, number of forest fragments, flowering plant species, and number of residences according to building typology, showed that axis 1 accounted for 53% of the data variability, while axis 2 accounted for 31%, resulting in a cumulative explanation of 84% (Figure 7).

Figure 7 - Principal Component Analysis (PCA) for the distribution of stingless bee nests and environmental variables of the urban ecosystem.



Source: Pereira DC, et al., 2024.

The principal component analysis revealed that sectors 1 and 3 are associated due to their similar characteristics, such as a higher number of mixed houses and nests of *S. depilis* occurring in the surroundings, and *T. angustula* inside the yards. In relation to sector 2, a distancing was observed due to the registration of the wooden building typology and the higher number of nests of *S. depilis* in the yard and *T. angustula* in the surroundings. The higher number of masonry houses and forest fragments made sector 4 distinctly separate from the other sectors. On the other hand, the higher number of flowering plant species separated sector 5 from the others in the principal component analysis.

These results suggest that the establishment of *S. depilis* is favored in areas outside the yard, such as proximity to more natural areas or the availability of suitable cavities outside residential properties. The variation in mixed structures (combined masonry and wood constructions) may contribute to some stingless bees finding intermediate shelter and

protection conditions compared to those in the natural ecosystem, enabling the installation of nests near the surroundings or even inside the yards.

The general data on the occurrence of *S. depilis* nests revealed a tendency for nest establishment in the surrounding area, but specifically in sector 2, nests were mainly found in the yard. This species is a more robust and larger stingless bee compared to *T. angustula*.

DISCUSSION

The forest fragments in Realeza, although isolated and small, contain native plant species typical of the Seasonal Semideciduous Forest, combined with exotic species used in urban landscaping. This heterogeneous composition, amplified by urbanization, highlights the challenge of forming effective ecological corridors, limiting genetic flow and the regeneration of native vegetation. This environmental disconnection directly affects species dependent on vegetation for nesting, such as stingless bees, which would benefit from greater ecological continuity. The alteration of the natural landscape due to territorial expansion and the increasing population concentration in urban areas has significantly contributed to a decline in the number of native stingless bee colonies (BERINGER J, et al., 2019), reducing nesting sites and food availability (JAFFÉ R, et al., 2019). Field surveys showed that even in areas with a higher number of forest fragments, nests of native bee species commonly found in urban ecosystems in Brazil (LIMA dos SANTOS SL, et al., 2020), such as *T. angustula* (Jataí), *S. depilis* (Canudo), *S. bipunctata* (Tubuna), *Nannotrigona testaceicornis* (Lepeletier, 1836) (Iraí), *Melipona quadrifasciata* Lepeletier, 1836 (Mandaçáia), *Frieseomelitta varia* Lepeletier, 1836 (Marmelada), and *Plebeia* spp. Schwarz, 1938 (Mirins), were not found. Thus, the presence of forest fragments in the urban and peri-urban landscape, expected to be a positive factor for the nesting of diverse stingless bees, did not reflect suitable microhabitats for colony establishment.

The configuration of these fragments is a selective factor, directly impacting the type of food resources available (varieties producing more nectar, pollen, or resins) for bees, safe shelters, competition with other pollinators, and predation risks. This suggests that residential areas with less intensive management, even in older sectors, provide better conditions for housing species like *S. depilis* and *T. angustula*. Similarly, the predominance of exotic plants in backyards reflects a gap in connecting landscaping practices with environmental sustainability, evidenced by increased foraging activity in gardens with native flowering species. This highlights the need to expand green areas in urban ecosystems with native species, resulting in

a feedback loop of pollination for seed production and ecosystem services related to the regeneration of regional native vegetation. TAYE RR (2020) asserts that the presence of large native tree species within forest fragments is essential for increasing the density of stingless bees, as they provide trunk cavities that can be used for nesting. The author also emphasizes that native plant species offer a range of floral rewards that make them more attractive to pollinators. COSTA-MACEDO CR, et al. (2020) also highlight that the best host trees for some stingless bee species are those with larger trunk circumferences, containing holes or cavities located more than 1.5 meters above the ground. In agricultural areas, the choice of nesting sites may be limited by the absence of trees with robust trunks (>15 cm), which are essential for providing safe shelters for nesting (TORNYIE F and KWAPONG PK, 2015). The reduction in the availability of trees in agricultural landscapes, combined with practices such as bush burning and wild honey collection, poses a significant threat to the survival and abundance of stingless bees. GONZÁLEZ-CHAVES A, et al. (2024) support these ideas, adding that it is not the tree species that is a key factor in nest selection, but rather their size and density in the environment. Thus, urban landscape designs should consider the preservation of pre-existing species and the introduction of species, preferably native ones, that meet these requirements (WEBER M, et al., 2023; CHACON K and GRECO S, 2022).

The loss or alteration of nesting substrates can lead to an increase in some stingless bee species and the extinction of others because biological responses to urban ecosystem changes are shaped by foraging and nesting habits. VIEIRA KM, et al. (2016) proposed that species developing strategies such as cryptic nest construction in pre-existing cavities (tree hollows, ground holes, termite mounds, or abandoned bird nests) are more likely to adapt to urban forest fragment areas. In this context, the self-maintenance of stingless bees in highly fragmented Atlantic Forest areas with scarce nesting sites can be optimized through artificial shelters (ARENA MVN, et al., 2018). Without a doubt, urbanization has posed significant challenges to native stingless bees, particularly regarding the availability of microhabitats and floral resources. However, these species have demonstrated high adaptability, utilizing a variety of artificial substrates, such as walls, fences, and poles, for nesting, in addition to exploiting gardens and urban forest fragments for food and shelter (ALBERNAZ JM, et al., 2020).

The predominance of exotic ornamental plants in gardens highlights a landscaping trend that prioritizes aesthetics over ecological functionality. However, it was observed that gardens with greater diversity and a higher number of flowering species attracted increased

foraging activity, especially from *T. angustula* (Jataí). This suggests that even in urbanized environments, the provision of floral resources can mitigate the negative impacts of reduced native vegetation and contribute to maintaining pollinator populations. Floral diversity is essential for interactions between pollinators and plants, ensuring food resource availability and colony development (VARASSIN IG, et al., 2021). Antonini and Martins (2022) suggest that urban sectors with higher density and richness of tree species, especially native ones, have greater potential to support stingless bees. These authors also argue that the permeability of the surrounding matrix (type of construction, presence of gardens, vegetation structure, size, and quality of forest fragments) influences migration between patches, affecting stingless bee dispersion in the urban ecosystem. Therefore, promoting flower-filled yards, urban parks, and forest fragment preservation are indispensable strategies for sustaining these populations and the associated ecosystem services, such as agricultural pollination (ARENA MVN, et al., 2023; PERSSON AS, et al., 2022). Diverse gardens and yards provide essential food resources for stingless bees, as well as protection against climatic fluctuations, such as temperature, humidity, and wind variations (KALUZA BF, et al., 2016). The internal garden layout is another important condition for foraging activity in domestic spaces, as it can be more or less attractive to stingless bees depending on its composition. A study by Mohamad WSNW, et al. (2024) observed the behavior of stingless bees concerning three garden designs (covered, natural, and mixed) and their relationship to bee permeability in domestic environments. The results suggest that in tropical areas subject to variations between rainy and intensely hot periods, the mixed design, characterized by a combination of open and closed areas, provides the best support capacity.

Regarding substrate selection for nest establishment, there is a certain selectivity among stingless bee species recorded in the urban environment, which may indicate nesting adaptation in the absence of natural microhabitats, leading to the use of artificial and synthetic structures. *T. angustula*, for instance, shows high adaptability to urban environments, occupying both natural cavities like tree trunks and artificial substrates such as wall crevices and wooden structures (SIQUEIRA ENL, et al., 2012). Its limited flight range, under 0.6 km, likely contributes to its preferential distribution in backyards where floral resources are closer. On the other hand, *S. depilis* depends more heavily on natural cavities, such as mature trees or trunks (WITTER S, et al., 2023), which favors its distribution in surrounding areas where these elements are more present. The hive architecture of Canudo seems to benefit from installation

in tree trunk cavities, facilitating colony thermal regulation and bee movement (OLIVEIRA-LIMA FV, et al., 2013).

Research by Biral de Faria L, et al. (2012) on the foraging behavior of *S. depilis* in urbanized areas indicates its high plasticity in exploiting floral resources across different habitats, including distant forest fragments. This species is recognized for its exceptional ability to collect pollen in urban forest areas (KRAEMER de MOURA M, et al., 2024), suggesting that nesting sites do not necessarily need to be located near floral resources. This medium-sized species (0.55 cm), with a more robust body and more aggressive behavior, typically has a lower incidence in anthropized environments when compared to *T. angustula*, which is smaller (0.40 cm), more delicate, and gentler (ABELHA, 2024). In a study by Araújo E, et al. (2004) involving 12 species of stingless bees, positive correlations between body size, wing size, and foraging range were evident, meaning that the larger the size, the greater the ability to explore more distant locations. Larger species exhibit a greater capacity to withstand events that limit flight for resource collection, particularly strong winds and heavy rains. Furthermore, stingless bee species display varying abilities to maneuver around artificial structures such as buildings, power poles, and heavy vehicle traffic. These factors in urban landscapes interfere with foraging behavior and the time it takes to return to the hive, whether following pre-established flight routes or not (KASIERA W, et al., 2023).

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The research data also revealed a variation in the distribution regarding the occupation of nesting space (backyards and surroundings) for both species, which was evident through Principal Component Analysis (PCA). When *S. depilis* occupies the backyards, *T. angustula* prefers to establish itself in the surroundings, which may reflect variations in competitive success between the species in the competition for substrates. The partitioning of resources among different stingless bee species is related to their specific need for pollen resources, which is a decisive factor for coexistence with other species in the same territory. Silva-Rodrigues C, et al. (2020) supports this idea and suggests that this phenomenon fits the exclusion principle model, where overlapping resource use leads to competition, and the greater the overlap, the higher the exclusion of species, until only one remains in the area.

The permeability of urban environments for stingless bees can be significantly influenced by the construction materials of houses, as these materials can determine the availability of nesting substrates and the microclimate in urban areas. Results have shown that in newer developments, where houses feature modern, less organic architecture, there is a

tendency not to find stingless bee nests. Greater environmental permeability is often associated with wooden structures and the presence of nearby vegetation, which favor the establishment of nests. Wooden residences usually provide more spaces for internal nesting, while mixed and masonry houses restrict access to peridomestic areas (backyards and surroundings). In this case, the type of material used in house construction has direct impacts on the level of permeability, either optimizing or limiting the occupation of architectural structures by stingless bees (SURIAWANTO N, et al., 2017). Thus, the architectural characteristics of buildings and their surroundings play a fundamental role in the distribution of stingless bee species in urban environments. These patterns highlight the importance of preserving natural elements in urban spaces, such as mature trees, and planning constructions that favor species conservation.

Cultural aspects unrelated to stingless bee biology can also modify the landscape on a local scale. Residents of older wooden, mixed-material, or masonry homes often cultivate nectariferous, fruit-bearing, or vegetable species in their backyards and maintain wooden or other structures to support ornamental or edible vegetation (fruits and vegetables). This configuration creates microhabitats suitable for establishing stingless bee colonies, combining refuge zones with access to food resources (pollen, nectar, and resins). Local and landscape-level structures affect stingless bee richness; larger orchards, floral richness, their interactions, and forest cover within a 1 km radius increase the likelihood of finding greater species richness (WAYO K, et al., 2020). Integrating human cultural practices with biological or ecological processes (bioculture) represents a promising pathway for agroecological education, indirectly contributing to pollinator conservation. Promoting and preserving the cultural heritage of gardens, orchards, and backyards can foster an interesting agroecological transition, even for modernized buildings with low permeability to stingless bees, as seen in large urban centers (MAYA EMA, et al., 2023).

Climatic conditions, such as temperature, humidity, and rainfall, also significantly influence stingless bee occurrence in urban environments, impacting their behavior. Bees, for instance, need to regulate their body temperature during flight, and factors like strong winds and rain can inhibit floral resource exploration and nesting departures (GRÜTER C, 2020; SILVA de CASTRO J, et al., 2019). These factors are crucial for colony development and new population expansion (HRNCIR et al., 2019). The study period was marked by abrupt temperature variations (18 to 28 °C) and excessive rainfall, which greatly hindered nest

detection as bee behavior shifted to a reclusive mode within colonies. Tracking nest occurrences relies on signs of foraging activity, flight paths, and movement near nest entrances. Bee departures are delayed under unfavorable environmental conditions (temperature and humidity), with workers staying longer inside the colony or reducing their daily activity time (HRISTOV M, et al., 2020). The urban environment of Realeza (PR) likely hosts a greater number of species, as foraging activities of *S. bipunctata*, known as Tubuna, and *Plebeia droryana* (Friese, 1900) and *Plebeia emerina* (Friese, 1900) (both known as Mirins), were recorded.

The findings obtained during the research demonstrate that conserving stingless bees in urban areas requires an integrated approach that includes native vegetation, ecological connectivity, and appropriate architectural characteristics. Promoting backyards with greater floristic diversity, preserving mature trees, implementing planned green spaces, and designing constructions that maintain environmental permeability are essential measures to promote stingless bee species' resilience and preserve the essential ecosystem services they provide (REMMERS R and FRANTZESKAKI N, 2024).

FINAL CONSIDERATIONS

The urban forest fragments of Realeza, while having potential to sustain biodiversity, demonstrated significant limitations as suitable habitats for stingless bees. The predominance of exotic plant species, habitat fragmentation, and urbanization compromise ecological connectivity, the availability of floral resources, and nesting sites. The high adaptability of some species, such as *T. angustula*, highlights opportunities for sustainable management strategies, including the use of artificial shelters, landscaping with native plants, and the integration of cultural practices in backyards. These actions can mitigate the impacts of urbanization, promoting the conservation of pollinators and the associated ecosystem services, such as agricultural pollination and vegetation regeneration. The connection between cultural practices and environmental sustainability, combined with the promotion of more permeable urban environments, can ensure the coexistence of stingless bees in urbanized landscapes.

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