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Urban green refuges influence pollen foraging and trophic plasticity of stingless bees

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Abstract

The present study evaluated the foraging of stingless bees (Meliponini) nesting at the Poeta Torquato Neto Campus in Teresina-PI, aiming to characterize the explored pollen spectrum and the temporal patterns of foraging activity in an urban environment. The research was conducted between October and December 2024, involving three nests established in natural tree cavities. Pollen loads transported in the workers' corbiculae were collected at different times and subjected to classical palynological analysis to identify the visited botanical families. The results evidenced a predominance of Myrtaceae, followed by Bignoniaceae, Turneraceae, and Anacardiaceae, indicating a selective yet flexible exploitation of available floral resources. Foraging activity was concentrated in the morning period, suggesting behavioral adjustment to microclimatic conditions and the temporal dynamics of pollen supply during the dearth period. The findings demonstrate that the campus acts as an ecological micro-refuge within an urban matrix, providing trophic resources and nesting substrates for the colonies. This study expands the understanding of the trophic niche and behavioral plasticity of stingless bees in anthropized landscapes and provides insights for management strategies and the conservation of native pollinators in urban environments.

Introduction

Stingless bees (SLB), belonging to the Meliponini tribe (Apidae), constitute one of the most diverse and ecologically relevant groups of pollinators in tropical and subtropical regions of the globe. In Brazil, the diversity of this group is significant, with hundreds of described species and a wide range of taxa still undergoing formal characterization, highlighting a complex functional and biological diversity essential for environmental balance (Vit et al., 2024).

These bees play a central role in maintaining the structure and resilience of both natural and anthropized ecosystems, acting as key pollinators for various native tree species. Through this interaction, SLB promote fundamental gene flows, aid in forest regeneration, and ensure the stability of trophic webs, establishing themselves as vital elements for biodiversity conservation, especially in biomes under fragmentation pressure (Guimarães-Alves et al., 2025).

Bee-mediated pollination represents a key ecological process that sustains both biodiversity and agricultural production. It is estimated that approximately 80% of the world's cultivated plant species depend, to some degree, on biotic pollination, with bees being the primary vectors of this ecosystem service. Although *Apis mellifera* is widely used in agricultural systems, SLB hold singular relevance in the pollination of native flora, particularly in highly biodiverse tropical environments (Martins et al., 2023; Moraes et al., 2025).

From an ecological perspective, SLB are traditionally classified as trophic generalists; however, recent studies demonstrate that this generalism can be modulated by environmental factors, floral availability, and interspecific competition, shaping patterns of contextual selectivity (Newis et al., 2023; Nuraeni et al., 2025). The concept of trophic niche, understood as the set of resources exploited by a species within

a given environmental matrix, reveals that Meliponini can exhibit significant dietary plasticity, adjusting their pollen spectrum according to landscape structure and local phenology (Batista et al., 2022; Caesar et al., 2021).

Pollen collection is a central element in colony dynamics, as it represents the primary source of protein, lipids, vitamins, and minerals essential for larval development and the maintenance of the workforce. The nutritional quality of pollen can vary substantially among plant species, influencing the productivity and stability of colonies (Zangirolami & Santos Junior, 2022). During critical dearth periods, characterized by a reduction in floral availability, a narrowing of the trophic niche or an intensification of foraging effort is frequently observed, a phenomenon already described for Meliponini in seasonal environments (Prastiyo et al., 2023; Saud et al., 2025).

In urban landscapes, the foraging behavior of SLB is mediated by specific environmental filters, such as vegetation fragmentation, the predominance of exotic ornamental species, microclimatic alterations, and the reduction of natural nesting substrates. These filters act by selecting more available or energetically advantageous plant species, potentially influencing the composition of the pollen spectrum transported by workers (Ghazi et al., 2024; Guimarães-Alves et al., 2025). Studies conducted in urban environments indicate that, although plant diversity may be high, spatial heterogeneity and irregular phenology shape the behavioral plasticity of colonies (Gatty et al., 2025; Ramos et al., 2026).

Classical palynological analysis of the pollen loads transported in the workers' corbiculae constitutes a robust tool for characterizing the instantaneous trophic niche of colonies, allowing for the precise taxonomic identification of visited families and genera. Unlike studies based exclusively on pollen stored in pots or honey metabarcoding (Martins et al., 2023), the analysis of corbicular loads provides a direct snapshot of foraging activity at the time of collection, enabling more immediate correlations with environmental variables and diurnal activity patterns.

University campuses embedded in urban matrices represent microenvironments of strategic ecological relevance by functioning as structured green refuges. These spaces offer relatively high tree diversity and lower disturbance intensity compared to the surrounding urban density, mitigating foraging constraints imposed by habitat fragmentation (Arena et al., 2026). However, knowledge regarding the pollen spectrum of stingless bees in these spaces is still limited, especially in the Brazilian Northeast, where pronounced climatic seasonality and periods of floral scarcity impose additional physiological and behavioral challenges on colonies (Gatty et al., 2025).

Given this scenario, the present study aimed to characterize the trophic niche and flight activity patterns of SLB nesting at the Poeta Torquato Neto Campus (UESPI) in Teresina-PI during the dearth period. Through palynological analysis of the pollen loads transported in the corbiculae and behavioral monitoring, we sought to identify the composition of the pollen spectrum and evaluate the influence of daily environmental variations on the collection effort. This work provides essential insights into the trophic plasticity of meliponines in anthropized environments and for the planning of urban green infrastructures aimed at pollinator conservation.

Material and Methods

Study Area

The institutional floristic survey records 602 arboreal individuals distributed across 49 botanical species, representing high plant diversity for an urban environment. This floristic heterogeneity creates favorable conditions for a continuous supply of floral resources, such as pollen and nectar, as well as potential substrates for nesting in natural cavities, making the campus a suitable microenvironment for investigations into plant-pollinator interactions within an anthropized context.

From an ecological perspective, the site can be characterized as an urban green refuge, where the arboreal structure and plant species diversity function as environmental filters that modulate the availability of trophic resources for stingless bees.

Temporal Delimitation and Georeferencing of Nests

Sampling was conducted between October and December 2024, a period corresponding to the transition between the dry season and the beginning of the regional rainy season, characterizing a floral dearth context for part of the local vegetation.

Field activities took place on distinct days, between 07:00 and 15:00 h, covering the primary periods of diurnal foraging activity for stingless bees.

Nest prospecting was performed through a systematic survey of the campus area, with occurrence points georeferenced using the GPS Field Area Measure application (version 1.0.2). Four sites with the presence of stingless bee (SLB) nests were identified, all located in natural tree cavities.

However, one of the georeferenced points could not be included in the pollen load sampling due to physical access restrictions to the nest; therefore, three actively accessible colonies were considered for analysis. The spatial distribution of the nests is represented in Fig. 1.

Pollen Load Sampling

Pollen loads were collected directly from workers returning to the nests with visible pollen in their corbiculae. To facilitate the selective capture of these bees, the nest entrance was temporarily obstructed for a short period, minimizing interference with colony dynamics.

Workers were individually captured using a large-diameter fine-mesh net, allowing for the exclusive selection of individuals carrying evident pollen loads. For each accessible nest, at least three independent pollen loads were collected to ensure a minimum representation of the floral spectrum exploited at the time of sampling.

Pollen removal was performed manually using sterile entomological forceps, ensuring that only the pollen load was removed without harming the individuals. After collection, the bees were immediately released and reintegrated into the colony to reduce behavioral or physiological impacts.

Each sample was individually placed in sterile acrylic microtubes, properly labeled with the nest's georeferencing information, date, time, and collection point, ensuring traceability and sample control. The spatial distribution of the sampling points is presented in Fig. 2.

Subsequently, the samples were stored under controlled conditions and transported to the Laboratory of New Technology Generation (GERATEC), located on the campus, where they underwent palynological identification procedures.

Preparation of Palynological Slides

The collected pollen loads were prepared for microscopic analysis using classical palynological techniques with a gelatinous medium as a support. The following materials were employed for slide mounting: glycerin jelly, Petri dishes, glass slides, coverslips, sterile entomological forceps, and clear nail polish for sealing.

Initially, a homogeneous layer of jelly was prepared in a Petri dish to serve as a fixation matrix for the pollen material, providing stable support and adequate optical visualization. Subsequently, a standardized fragment of the jelly was transferred to the center of a previously cleaned glass slide.

A small fraction of the pollen load was then deposited onto the surface of the jelly using sterile forceps, being carefully dispersed to avoid clumps that could hinder morphological observation. To ensure integrity and prevent cross-contamination, each sample was handled with an exclusive instrument.

Immediately after, a coverslip was positioned over the material, and the slide was subjected to light, controlled heating to promote partial fusion of the jelly. This procedure allowed for the elimination of air bubbles and the uniform distribution of pollen grains across the optical field.

After cooling, the edges of the coverslip were sealed with clear nail polish, forming a continuous peripheral seal. The slides remained at rest for 5 to 10 minutes until completely dry, ensuring the stability of the preparation and preventing external contamination.

Palynological Identification and Analytical Approach

The prepared slides were analyzed under a light microscope, and the pollen grains were identified based on morphological features such as size, shape, aperture type, exine ornamentation, and symmetry. Taxonomic identification was performed by comparison with specialized palynological literature and reference atlases.

The adopted methodological approach allows for the characterization of the instantaneous pollen spectrum exploited by the workers at the time of collection, providing insights into the trophic niche of the colonies in an urban environment.

Considering that the analyzed nests are naturally distributed across the campus and that no experimental manipulation of the colonies occurred, the study design is characterized as observational and descriptive. Thus, the data were processed using descriptive statistics to qualitatively characterize the taxonomic composition of the collected pollen and general foraging patterns.

Results and Discussion

Pollen load samples were obtained from three stingless bee nests located in natural tree cavities at the Poeta Torquato Neto Campus. The nests were established in arboreal individuals of Melicoccus bijugatus Jacq. (Nest 1), Syzygium cumini (L.) Skeels (Nest 2), and Phytolacca dioica L. (Nest 3), as illustrated in Fig. 3. All nests exhibited the typical tubular entrance structure constructed with cerumen, evidencing active colonies adapted to the natural substrate available in the campus's urban environment.

The use of natural cavities as nesting substrates reinforces the widely described pattern for Meliponini, whose colonies depend on tree hollows for establishment and structural protection. Studies conducted in urban and peri-urban environments demonstrate that the availability of mature trees with natural cavities is a determining factor for the persistence of stingless bees (SLB) in anthropized landscapes (Guimarães-Alves et al., 2025; Ghazi et al., 2024). Thus, the presence of tree species with the potential for cavity formation on campus suggests that the vegetation structure acts not only as a source of trophic resources but also as essential support for colony maintenance.

From an ecological perspective, nesting in trees distributed across an urban green fragment indicates that the campus functions as a structural micro-refuge within the urban matrix. The maintenance of colonies in environments with diverse tree cover is associated with greater trophic stability and an expansion of the explored ecological niche, favoring broader and more flexible foraging patterns (Batista et al., 2022; Ramos et al., 2026). This scenario suggests that, even within an urban context, SLB maintain adaptive strategies compatible with those observed in natural environments.

Furthermore, the choice of different tree species as nesting substrates may reflect behavioral plasticity in the selection of available cavities, a recurring characteristic in Meliponini faced with the structural heterogeneity of the landscape (Newis et al., 2023). In urban environments, where the supply of cavities may be limited or irregular, this behavioral flexibility represents an important mechanism of ecological resilience.

The foraging activity of the stingless bees showed variation throughout the morning period, as illustrated in Graph 1. A higher collection frequency was observed in the early morning hours, between 07:40 and 09:05 h, with two sampling units recorded per interval. After this period, there was a reduction in activity

at 09:12 h, followed by a brief increase at 09:35 h and a further decrease at 09:42 h. The trend line indicates a gradual decline in collection over the observed time.

The observed pattern suggests that foraging intensity is concentrated in the early hours of the day, a behavior widely recorded for Meliponini across different environmental contexts. Studies demonstrate that the external activity of workers tends to be maximized during periods of moderate temperature and higher availability of newly secreted nectar, decreasing as temperatures rise throughout the morning (Prastiyo et al., 2023; Nuraeni et al., 2025). This behavior is associated with the optimization of energy expenditure, as high temperatures can increase the metabolic cost of flight and accelerate dehydration.

From the perspective of optimal foraging theory, the concentration of activity within specific time windows indicates behavioral adjustment to microclimatic conditions and the dynamics of floral supply. In urban environments, where the microclimate may be modulated by heat islands and the structural heterogeneity of vegetation, SLB tend to exhibit behavioral plasticity, adjusting collection times according to the balance between energy cost and nutritional return (Newis et al., 2023; Ramos et al., 2026). The gradual reduction trend observed in the graph may reflect both variation in the immediate availability of resources and a response to thermal changes during the morning period.

Furthermore, the punctual oscillation observed at 09:35 h may indicate episodic events of greater floral availability or momentary recruitment of foragers, a phenomenon already described for Meliponini in contexts of spatially distributed resources (Batista et al., 2022). This dynamic reinforces that, even in urban fragments, colonies maintain a capacity for fine-tuning in worker allocation, evidencing ecological resilience in the face of anthropized environments.

Graph 1. Variation in the number of pollen loads collected during the morning period (07:40–09:42 h), representing the intensity of foraging activity of stingless bees (SLB) in the evaluated nests. **Source**: Prepared by the author (2026).

The highest foraging activity occurred in the early morning, with a concentration of collections during the first hours of the day. No pollen collection was recorded during the afternoon on the evaluated days, indicating a reduction in foraging activity as daily temperatures increased.

The predominance of activity in the morning period is associated with microclimatic conditions more favorable for flight, especially milder temperatures and higher relative humidity, factors that directly influence the physiological performance of the workers. Studies conducted with *Tetragonula biroi* demonstrated that the intensity of forager entry and exit is significantly higher in the early hours of the day, with a progressive decline as the temperature rises (Prastiyo et al., 2023). Similarly, analyses of behavioral patterns in Meliponini indicate that foraging is optimized within specific thermal windows, evidencing behavioral plasticity in response to environmental conditions (Nuraeni et al., 2025).

The absence of collection in the afternoon may also be related to the dynamics of floral supply. In many plant species, pollen release occurs predominantly in the early morning hours and decreases throughout

the day, which can limit the energy return of late foraging. This pattern has already been observed in seasonal tropical environments, where resource availability presents a strong temporal component (Saud et al., 2025). Thus, the behavior recorded in this study suggests a fine-tuning between floral availability and the energy cost of flight, compatible with the assumptions of optimal foraging theory.

However, when investigating the behavior of *Melipona fasciculata* also in Teresina-PI, other authors observed activity distributed throughout both the morning and afternoon, indicating a broader pattern of daily exploitation. This difference suggests that intra- or interspecific variations, as well as local microclimatic characteristics and landscape structure, can modulate the temporal range of foraging (Caesar et al., 2021). The divergence between these results reinforces that the trophic niche and the daily collection rhythm are not fixed but are shaped by interactions between climate, plant phenology, and the behavioral strategies of the colonies.

Diversity of Floral Resources

The palynological analysis of the corbicular loads revealed the presence of four botanical families in the samples collected between October and December 2024: Myrtaceae, Bignoniaceae, Turneraceae, and Anacardiaceae (Table 1). The Myrtaceae family was recorded across different nests and collection dates, demonstrating a higher recurrence within the observed pollen spectrum. The other families occurred occasionally, associated with specific times and nests.

Tabela 1. Pollen families identified in stingless bee nests

From: Botanical families identified in pollen loads collected from stingless bee nests at the Poeta Torquato Neto Campus (UESPI), Teresina, Brazil, indicating nest identification, sampling date, and collection time during the study period.

Nest	Collection Date	Collection Time	Families
1	12/07/2024	09:35	Myrtaceae and Bignoniaceae
	12/07/2024	09:42	Myrtaceae
2	12/07/2024	09:05	Myrtaceae and Turneraceae
	12/07/2024	09:12	Myrtaceae
3	26/10/2024	07:40	Anacardiaceae
	11/02/2024	08:30	Anacardiaceae
	11/09/2024	08:00	Bignoniaceae

Source

Prepared by the author (2026).

The predominance of Myrtaceae in the pollen spectrum is consistent with the pattern widely described for Meliponini in tropical environments, as this family presents high pollen production and broad distribution in both urban and forest formations. Studies utilizing both classical palynology and metabarcoding have recorded a strong representation of Myrtaceae in the diet of different stingless bee (SLB) species (Martins et al., 2023; Moraes et al., 2025). The recurrence of this family in the present study suggests significant local availability and a possible preference associated with high nutritional return.

The complementary occurrence of Bignoniaceae, Turneraceae, and Anacardiaceae indicates trophic niche breadth, albeit with low total diversity of recorded families. In urban contexts, the composition of the pollen spectrum tends to reflect environmental filters related to landscape structure and the phenology of available ornamental or arboreal species (Ghazi et al., 2024; Guimarães-Alves et al., 2025). Thus, the moderate diversity observed may be associated with both the dearth period and the spatial distribution of flowering plants on the campus.

From an ecological perspective, the presence of multiple families, even in small numbers, reinforces the pattern of functional generalism typical of SLB, which is nonetheless modulable according to local availability. Studies conducted in seasonal environments demonstrate that the trophic niche may undergo narrowing during periods of lower floral supply, concentrating on the most abundant or energetically advantageous species (Saud et al., 2025; Batista et al., 2022). In this sense, the spectrum recorded in this study may reflect an adaptive adjustment to the environmental conditions prevailing during the analyzed period.

Incidence of Botanical Families

The incidence of the plant families identified in the pollen loads is represented in Graph 2. The Myrtaceae family exhibited the highest relative frequency, being recorded in samples from Nests 1 and 2. Bignoniaceae was identified in a lower proportion, occurring in Nests 1 and 3. Turneraceae and Anacardiaceae showed punctual records, restricted to Nests 2 and 3, respectively.

Graph 2. Incidence of botanical families identified in pollen loads collected from stingless bee (SLB) nests, expressed as the number of records per family during the sampling period. **Source**: Prepared by the author.

The higher representativeness of Myrtaceae suggests a strong contribution of this family to the trophic spectrum of the analyzed colonies. Species such as *Psidium guajava*, *Myrciaria cauliflora*, and *Eugenia uniflora* are frequently utilized by Meliponini due to their high pollen production and broad availability in urban environments. Studies based on metabarcoding and classical palynology demonstrate the recurrent dominance of Myrtaceae in the diet of different stingless bee (SLB) species, reinforcing its ecological relevance as a strategic resource (Martins et al., 2023; Moraes et al., 2025). The high incidence observed in this study may reflect both local abundance and favorable nutritional return.

The presence of Bignoniaceae, although less frequent, indicates complementarity in the exploitation of floral resources. Species such as *Tecoma stans* are widely distributed in urban landscapes and known for their attractiveness to pollinators. Although nectar represents the primary floral reward for this family, the pollen collection recorded in this study evidences that SLB exploit multiple available resources, expanding their dietary plasticity according to environmental supply. This pattern is compatible with the contextual generalism described for Meliponini in heterogeneous environments (Batista et al., 2022; Ghazi et al., 2024).

The punctual occurrences of Turneraceae and Anacardiaceae indicate opportunistic exploitation of specific resources, possibly associated with the momentary phenology of these species during the sampling period. In seasonal contexts, the trophic niche breadth may undergo temporal variations, concentrating on species with active flowering during the analyzed period (Saud et al., 2025). The presence of *Turnera subulata*, for instance, has already been recorded as a relevant resource for SLB in tropical urban environments, especially due to floral accessibility and the simultaneous supply of pollen and nectar (Nuraeni et al., 2025).

The limitation in the total number of collected samples was associated with the low incidence of pollen loads transported by workers, with a frequent return of individuals without visible pollen in their corbiculae. This pattern may indicate a reduction in the immediate availability of resources or a greater investment in nectar collection during the evaluated period. Studies conducted in urban and seasonal environments demonstrate that the intensity of pollen collection can oscillate significantly as a function of climatic conditions and plant phenology, reflecting adaptive adjustments of the colonies to environmental constraints (Prastiyo et al., 2023; Ramos et al., 2026).

Conclusion

Stingless bees nesting at the Poeta Torquato Neto Campus exhibited a pollen spectrum predominantly composed of Myrtaceae, with complementary records of Bignoniaceae, Turneraceae, and Anacardiaceae, evidencing a selective yet flexible exploitation of the floral resources available in the urban environment. Foraging activity was concentrated in the morning period, indicating a behavioral adjustment to microclimatic conditions and the temporal dynamics of floral supply during the evaluated dearth period.

The results demonstrate that the campus acts as an ecological micro-refuge, simultaneously providing nesting substrates and trophic resources essential for colony maintenance. Thus, this study expands the understanding of the trophic niche and behavioral plasticity of stingless bees (SLB) in urban landscapes of the Brazilian Northeast, reinforcing the relevance of planning and conserving urban green areas for the sustainability of native pollinators.

Declarations

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Declaration of competing interest

The author has no conflicts of interest to declare.

DECLARATION OF INTERESTS

 The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

 The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

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Graphs

Graphs 1 and 2 are available in the Supplementary Files section

Figures

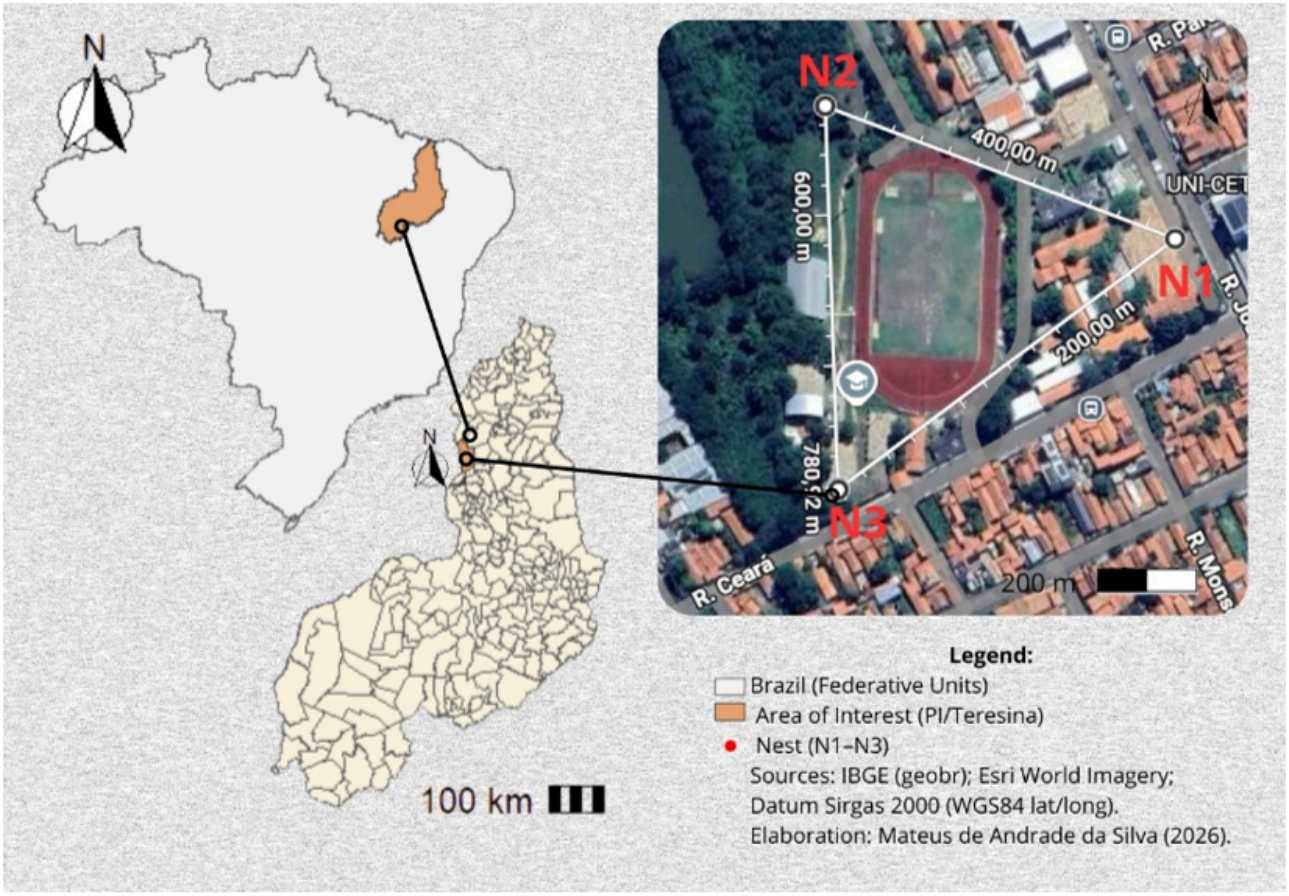


Figure 1

Geographic location of stingless bee (SLB) nests at the Poeta Torquato Neto Campus, State University of Piauí (UESPI), Teresina, Piauí, Brazil. The map displays the study area at national (Brazil), regional (Teresina municipality), and local scales, indicating the georeferenced nests (N1–N3) embedded within the urban matrix. **Elaboration**: Mateus de Andrade da Silva (2026).



Figure 2

Microtubes containing pollen loads collected from stingless bee (SLB) workers, properly labeled with date, time, and nest identification, ensuring traceability and sample control for palynological analysis. **Source:**Prepared by the author (2026).



Figure 3

Stingless bee (SLB) nests established in natural tree cavities at the Poeta Torquato Neto Campus, showing the use of arboreal hollows as nesting substrates and the characteristic tubular entrance

structures built with cerumen. **Source:** Prepared by the author (2026).

Supplementary Files

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