FLORAL VISITORS AND POLLEN ASPECTS OF *Jacaranda mimosifolia* D. Don (Bignoniaceae) IN AN URBAN LANDSCAPE

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Abstract

The reproductive biology of plants in urban environments remains largely unexplored. This study investigated *Jacaranda mimosifolia*, a species of great landscape relevance in Brazil, focusing on floral visitor diversity and pollen characteristics. Observations were made in two urban areas with different population densities of the species. Visits from bees (*Apis mellifera*, *Bombus* sp., *Euglossa* sp., *Trigona* sp.) and hummingbirds (*Amazilia* sp.) were recorded, with *Bombus* sp. being the main pollinator. Other visitors acted predominantly as "nectar robbers". An isolated specimen received fewer visits, suggesting that simultaneous flowering of multiple individuals favors a higher visitation rate. The pollen is tricolpate, prolate, perforate, and dispersed as monads, with an average of 1,126 grains per anther and a pollen-ovule ratio of 56:1, typical of species with biotic pollination. Despite the urban context, *J. mimosifolia* showed a high visitation rate and viable pollen. However, visitor diversity was lower compared to previous studies, highlighting the need for further research on plant reproduction in anthropized environments.

Keywords: Nectar Robbers; Palynology; Pollen Viability; Pollination; Reproductive Biology.

VISITANTES FLORAIS E ASPECTOS POLÍNICOS DE *Jacaranda mimosifolia* D. Don (Bignoniaceae) EM UMA PAISAGEM URBANA

Resumo

A biologia reprodutiva de plantas em ambientes urbanos permanece amplamente pouco explorada. Este estudo investigou *Jacaranda mimosifolia*, uma espécie de grande relevância paisagística no Brasil, com foco na diversidade de visitantes florais e nas características do pólen. As observações foram conduzidas em duas áreas urbanas com diferentes densidades populacionais da espécie. Foram registradas visitas de abelhas (*Apis mellifera*, *Bombus* sp., *Euglossa* sp., *Trigona* sp.) e beija-flores (*Amazilia* sp.), sendo *Bombus* sp. o principal polinizador. Outros visitantes atuaram predominantemente como "ladrões de néctar". Um indivíduo isolado recebeu menos visitas, sugerindo que a floração simultânea de múltiplos indivíduos favorece maior taxa de visitação. O pólen é tricolpado, prolato, perfurado e disperso em mônades, com média de 1.126 grãos por antera e razão pólen-óvulo de 56:1, típica de espécies com polinização biótica. Apesar do contexto urbano, *J. mimosifolia* apresentou alta taxa de visitação e pólen viável. Entretanto, a diversidade de visitantes foi menor em comparação a estudos anteriores, ressaltando a necessidade de mais pesquisas sobre reprodução vegetal em ambientes antropizados.

Palavras-chave: Biologia Reprodutiva; Palinologia; Polinização; Ladrões de Néctar; Viabilidade Polínica.

INTRODUCTION

Plant reproductive biology seeks to understand the associations between floral attributes, sexual systems, modes of reproduction, pollination, phenology, and the dispersal of fruits and seeds (Rech et al. 2014). Research in this field began in the 17th century with controlled pollination experiments, particularly in temperate climate species (Barrett, 2010). On the other hand, tropical plants are often overlooked in this type of approach, as well as ecological studies in urban areas (Barrett, 2010).

Bignoniaceae comprises 79 genera and approximately 800 species with a pantropical distribution, with few representatives in temperate regions (Lohmann et al. 2022). Its flowers can have floral attributes associated with pollination by bees, bats, birds, moths, and butterflies (Lopes et al. 2002; Machado; Vogel 2004; Bittencourt; Semir 2006). Jacarandeae is the first lineage to diverge within the family (Olmstead et al. 2009) and is divided into four sections: *Jacaranda* sect. *Nematopogon*, which includes species with a staminode divided at the apex and a spathaceous calyx; *Jacaranda* sect. *Copaia* with monothecal anthers and a cup-shaped calyx; *Jacaranda* sect. *Jacaranda* with monothecal anthers and a campanulate calyx; and *Jacaranda* sect. *Dilobos*, which features bithecal anthers and a cup-shaped calyx (Ragsac et al. 2019).

Jacaranda Juss. is a genus of trees and shrubs with a Neotropical distribution, comprising 49 species (Lohmann et al. 2022). In Brazil, there are 39 species, of which 34 are endemic (Lohmann et al. 2022). *Jacaranda mimosifolia* D. Don, commonly known as "jacarandá-mimoso", is native to Argentina, Bolivia, and Paraguay, and is exotic to Brazil (Ragsac et al. 2019; Lohmann et al. 2022), but it is of great importance for ornamental use in urban landscaping (Alves et al. 2010; Freitas et al. 2020). The flowers of *J. mimosifolia* are self-incompatible and have a purple color, with a mild fragrance produced by osmophores located in the staminode (Alves et al. 2010).

It is known that exotic species with floral resources can attract native pollinators, which can have competitive or facilitative effects on coexisting native plants (Cuadra-Valdés et al. 2021). For *J. mimosifolia*, it has been reported that medium to large-sized native bees from Brazil are efficient pollinators due to their behavior during visits (Alves et al. 2010; Rana; Chauhan, 2012). The analysis of floral visitors and pollen viability is essential to elucidate issues regarding pollinator behavior and reproductive success (Dafni; Firmage 2000; Kay et al. 2020; Xu et al. 2022).

Based on this context, this study aimed to analyze aspects of the reproductive biology of *J. mimosifolia* individuals in an urban

environment in Lavras, Minas Gerais State, Brazil, where such research has not been conducted before. The tree is widely used as an ornamental plant in the city, making it an important subject for understanding its ecological interactions in urban landscapes (Chagas Júnior, 2010). In addition, we sought to answer the following questions: 1. What are the main floral visitors of *J. mimosifolia* in urban environments? 2. Does the proximity between *J. mimosifolia* individuals in full bloom influence the floral visitation rate and pollen characteristics?

MATERIAL AND METHODS

Floral Visitors

We conducted 60 hours of observations between November 5, 2022, and November 13, 2022, on three *Jacaranda mimosifolia* individuals located in an urban area of Lavras - Minas Gerais State, Brazil (-21.225743S, -44.977815W for individuals one and two; -21.227560S, -44.976637W for individual three). The observation times ranged from 5:40 AM to 7:00 PM.

At the first geographic coordinate, the individuals were part of a group of 30 trees of the same species positioned close to each other, while at the second coordinate, only two *J. mimosifolia* trees were observed. The two geographic coordinates are separated by 240 meters.

We used the classification of floral visitors proposed by Alves-dos-Santos et al. (2016), which considers effective pollinators (those that frequently contact both anthers and stigmas and carry abundant pollen grains); occasional pollinators (those that carry pollen grains upon contacting stigmas but in low abundance and with reduced visitation frequency); and robbers (those that do not directly contact anthers or stigmas, limiting their activity to collecting nectar or pollen).

We performed an analysis of variance (ANOVA) to check for significant differences in the number of visits between individuals, followed by Tukey's test for multiple comparisons. The distribution and variation of visits were visualized through graphs generated with the ggplot2 package (Wickham, 2016) in the R environment (R Core Team, 2024).

The inflorescences were collected to describe floral morphology, and the flowers were exposed to a dark chamber illuminated by UV light to assess reflectance, a factor frequently associated with pollinator attraction (Chen et al. 2020).

Pollen Morphology and Viability

Flowers were fixed in Carnoy 3:1 (ethanol and acetic acid) and stored at -4°C. Pollen viability was assessed using 2% acetocarmine and 2% Lugol’s iodine stains (Guerra; Souza 2002). Additionally, pollen tube germination tests were performed on a culture medium with different sucrose concentrations (5%, 10%, 15%, and 20%). A

total of 100 pollen grains per slide were evaluated across five slides per stain/treatment.

Pollen characterization was carried out using the categories of Punt et al. (2007) and with the use of acetocarmine. Measurements were taken from 100 pollen grains across the three individuals using the free software ImageJ. Finally, the number of pollen grains produced per anther was counted with the aid of a Neubauer chamber, using 10 anthers from 10 flowers for each individual, in order to determine the pollen:ovule ratio. Slides were examined under a light microscope (Carl Zeiss, AxioLabA1), equipped with a digital camera (AxioCam ICc1).

RESULTS AND DISCUSSION

The flowers of *J. mimosifolia* are arranged in panicles averaging 30 cm in length (Figure 1a), with each inflorescence bearing up to 300 flowers. The calyx is gamosepalous and cup-shaped (Figure 1b), while the corolla is gamopetalous, lilac-colored, and features a white nectar guide (Figure 1c). The androecium consists of five stamens — four fertile and one staminode (Figure 2a). The fertile stamens are didynamous with white, monothecal anthers (Figure 2b). The staminode is larger than both the stamens and gynoecium, and bears lilac glandular trichomes (Figure 2c). The gynoecium comprises a bifid stigma (Figure 2d), a nectariferous disc (Figure 2e), and a bicarpellate ovary containing approximately 80 ovules (Figure 2f).

Within Bignoniaceae, eight distinct floral morphologies are recognized (Alcântara & Lohmann, 2010). The floral morphology of *J. mimosifolia* corresponds to the "*Anemopaegma* type", characterized by a tubular calyx, an infundibuliform corolla, and an open floral apex. This configuration is considered the ancestral condition for the family and is frequently associated with the presence of nectar guides (Alcântara & Lohmann, 2010). Although nectar guides often reflect UV light in various plant groups (Leonard & Papaj, 2011; Lunau et al., 2020), this feature was not observed in *J. mimosifolia* (Figure 1d).

At least five species of floral visitors were recorded (Table 1, Figure 3). The frequency of visits was similar for individuals one and two, whereas individual three received significantly fewer visits. ANOVA followed by Tukey's test confirmed significant differences: individual three differed from individuals one ($p = 0.020$) and two ($p = 0.014$), but no difference was found between the latter two ($p = 0.977$) (Table 1). Greater richness and frequency of floral visitors tends to occur when conspecific or heterospecific individuals bloom simultaneously (Laverty, 1992; Ghazoul, 2006; Molina-Montenegro et al. 2008; Peter; Johnson 2008; Barônio et al. 2016), which aligns with our observations (Table 1, Figure 3).

The recorded visitors included the bee species *Apis mellifera* L., *Bombus* Latreille sp., *Euglossa* Latreille sp., and *Trigona* Jurine sp.

(Figure 4a–d), in addition to hummingbirds of the genus *Amazilia* R.Lesson (Figure 4e). Among these, only *Bombus* was identified as an effective pollinator. The remaining species exhibited nectar-robbing behavior (Table 1). Compared to the findings of Alves et al. (2010), who reported a higher diversity of pollinators — including *Eulaema nigrita* Lepeletier and *Xylocopa* Latreille sp. — our study recorded a lower diversity. Nonetheless, *J. mimosifolia* displayed a higher overall visitation frequency, predominantly due to nectar-robbing species.

The "*Anemopaegma* type" floral morphology is generally associated with bee pollination, especially by euglossine bees (Alcântara; Lohmann 2010). However, *Euglossa* sp. individuals were observed only briefly between 2 PM and 3 PM, entering and exiting the corolla within about one second, without acting as effective pollinators. In contrast, *Bombus* (Figure 4a–b) visited flowers between 6 AM and 6 PM, remaining in the corolla for approximately three seconds and pollinating multiple flowers per inflorescence, as previously reported (Guimarães et al. 2008; Alves et al. 2010). No floral visitors were recorded after 6:30 PM.

Hummingbirds (Figure 3e) were active from 7 AM to 5 PM and perforated the base of the corolla to access nectar without contacting the anthers or stigma, thus acting as nectar robbers (Figure 5). Similarly, *A. mellifera* and *Trigona* sp. (Figure 4c–d) displayed typical nectar-robbing behavior, with visits recorded between 9 AM and 6 PM, and after 3 PM, respectively. Nectar robbing is commonly reported in Bignoniaceae (Navarro, 2000; Barros, 2001; Souza et al. 2004; Guimarães et al. 2008; Milet-Pinheiro; Schlindwein 2009). In *Jacaranda rugosa* A.H.Gentry, *Trigona spinipes* Fabricius was responsible for circular perforations in the corolla (Milet-Pinheiro; Schlindwein 2009). Likewise, smaller perforations observed in Figure 5a–b were caused by hummingbirds, while those in Figure 4c were attributed to *Trigona* spp.

All individuals exhibited high pollen viability: 90.83% with acetic carmine and 92.13% with Lugol (Figure 6a–b). However, pollen grains did not germinate in media containing different sucrose concentrations, suggesting that specific substances or conditions are required to stimulate germination. Morphologically, the pollen grains were consistent with previous descriptions for *Jacaranda* spp. (Fasasi; Alluh 2019; Mallick, 2019): tricolpate (Figure 6c–d), isopolar, prolate, with perforate ornamentation (Figure 6c), and dispersed as monads. Grain size averaged 48.12 µm (SD: 34.45 µm). Pollen production per anther ranged from 910 to 1629, with an average of 1126 ± 235. This results in a pollen:ovule ratio of approximately 56:1 — typical of species pollinated by biotic vectors (Culley et al. 2002).

In summary, the *J. mimosifolia* individuals analyzed in this study showed low diversity of floral visitors but high pollen viability, even in an urban environment —

where increased pollution could theoretically reduce viability (Gottardini et al. 2004; Vasilevskaya, 2022; Cristofolini et al. 2024). Areas with more individuals blooming simultaneously received higher visitation rates. However, the limited diversity of floral visitors suggests that *J. mimosifolia* may attract a narrow range of pollinators, which could affect its pollination success. These findings highlight the species' ability to attract pollinators, but also point to potential limitations in ecological resilience due to the reduced diversity of visitors in urban environments.

Table 1: Floral visitors and their visit frequencies on *Jacaranda mimosifolia* individuals during 60 hours of observation. Different letters indicate significant differences ($p < 0.05$) according to Tukey's test.

Species	**Behavior**	**Frequency of visits in individual 1**	**Frequency of visits in individual 2**	**Frequency of visits in individual 3**
Amazilia sp	robbers	18 (13.23%)	36 (25.17%)	2 (6.25)
Apis mellifera	robbers	42 (30.88%)	49 (34.26%)	7 (21.87%)
Bombus sp.	pollinator	27 (19.85%)	12 (8.39%)	8 (25%)
Euglossa sp.	robbers	15 (11.09%)	26 (18.18%)	3 (9.37%)
Trigona sp.	robbers	34 (25%)	20 (13.98%)	9 (28.12%)
Total		136	143	32
Mean		27 **a**	28 **a**	6 **b**

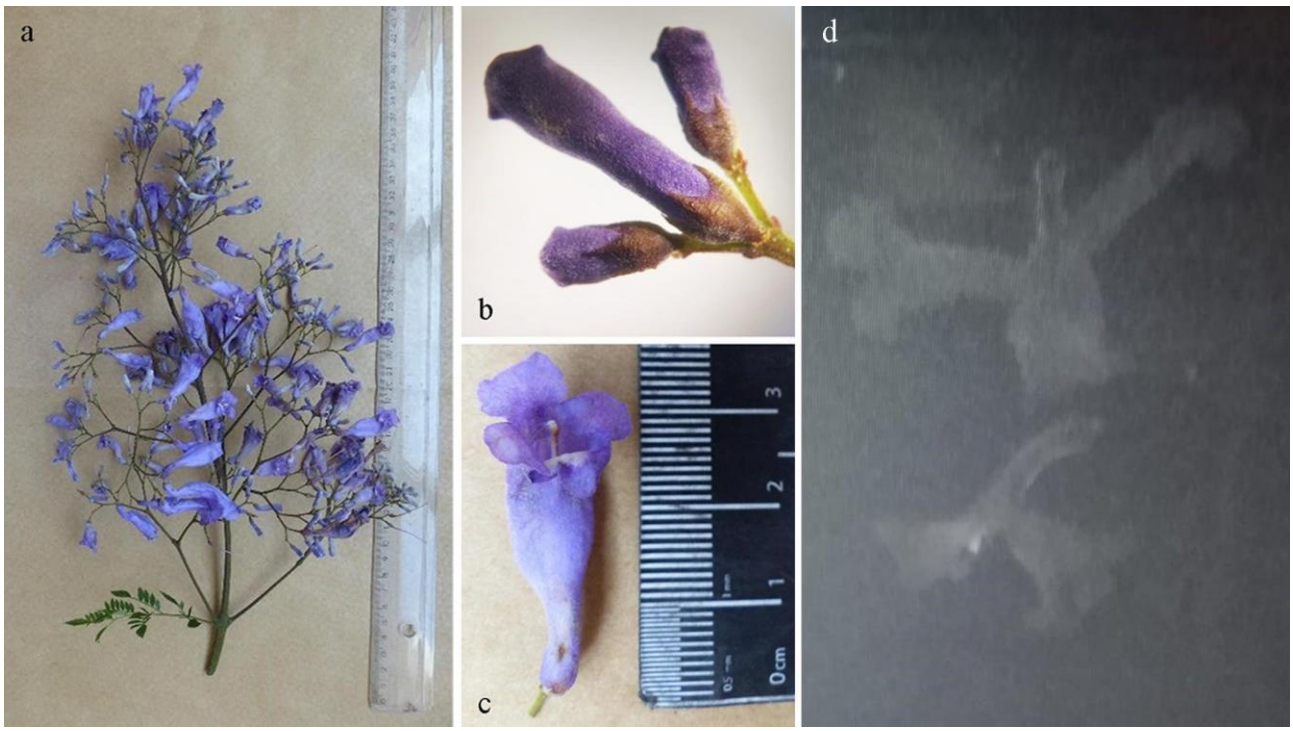


Figure 1 – Morphological aspects of *Jacaranda mimosifolia*. **a** Inflorescence. **b** Flower buds. **c** Flower. **d** Flowers exposed to UV rays

Source: Authors.

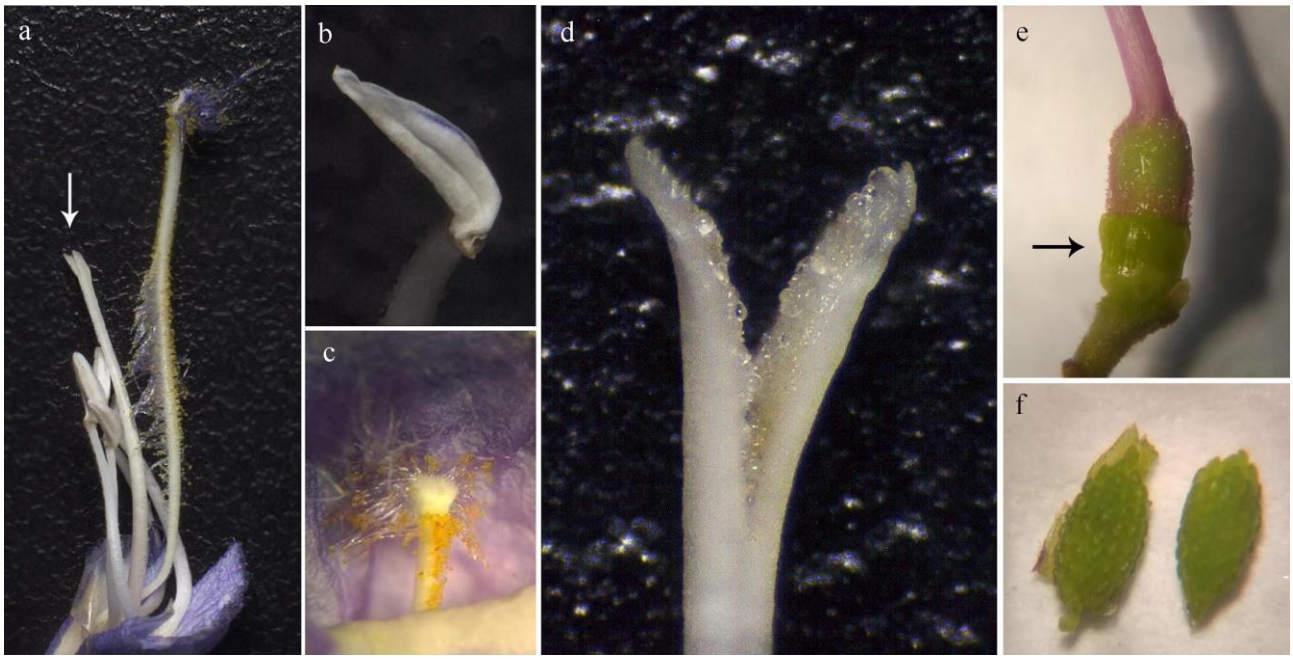


Figure 2 – Floral parts of *Jacaranda mimosifolia*. **a** Androecium and gynoecium (arrow indicates gynoecium). **b** Monothecal anther. **c** Details of the staminode. **d** Bifid stigma. **e** Ovary (arrow indicates nectary disk). **f** Carpels with ovules

Source: Authors.

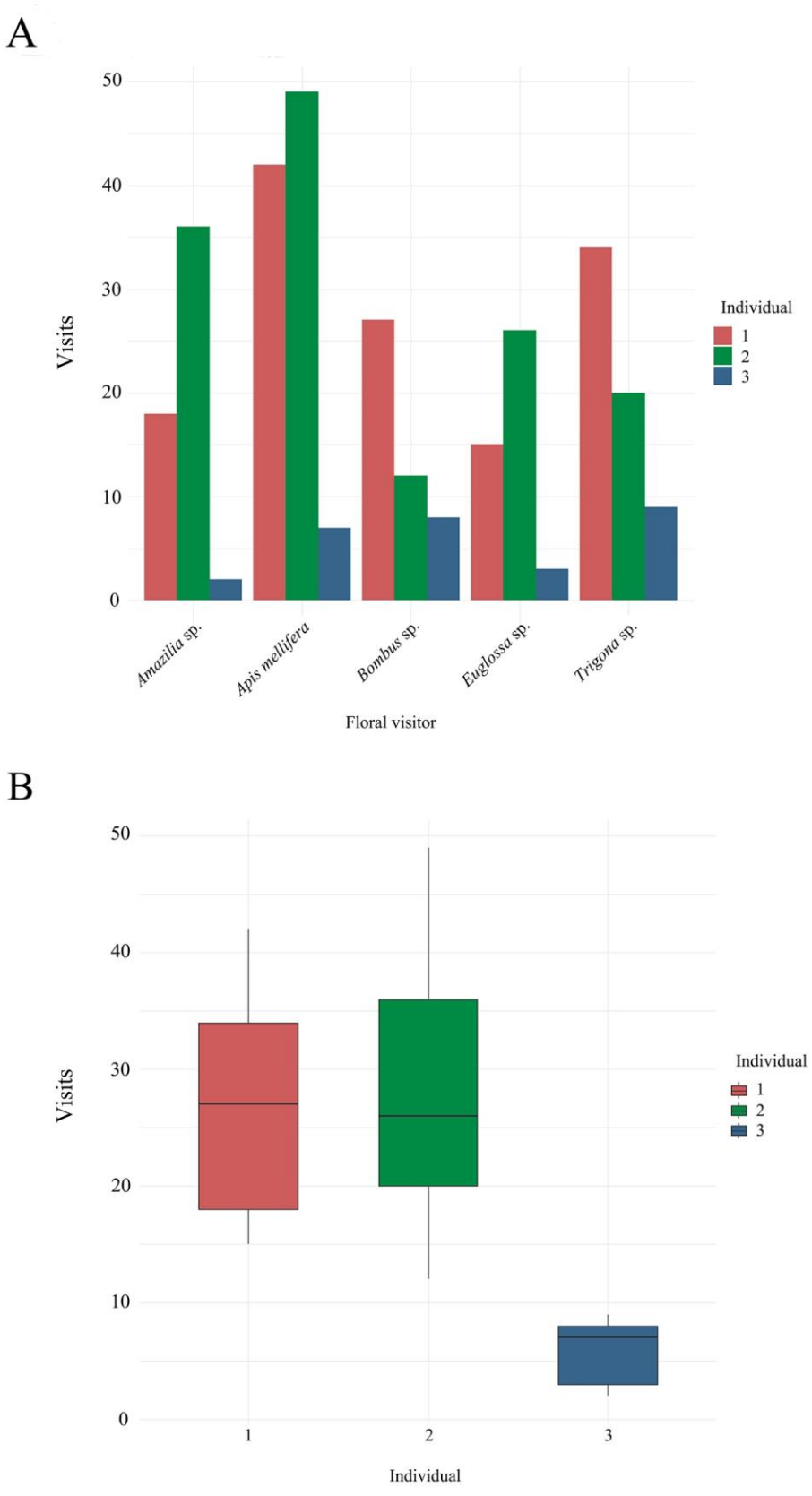


Figure 3 – Floral visits in *Jacaranda mimosifolia*. (a) Number of visits by different floral visitors to three individuals of *J. mimosifolia*. (b) Distribution of the number of floral visits among individuals of *J. mimosifolia*.

Source: Authors.

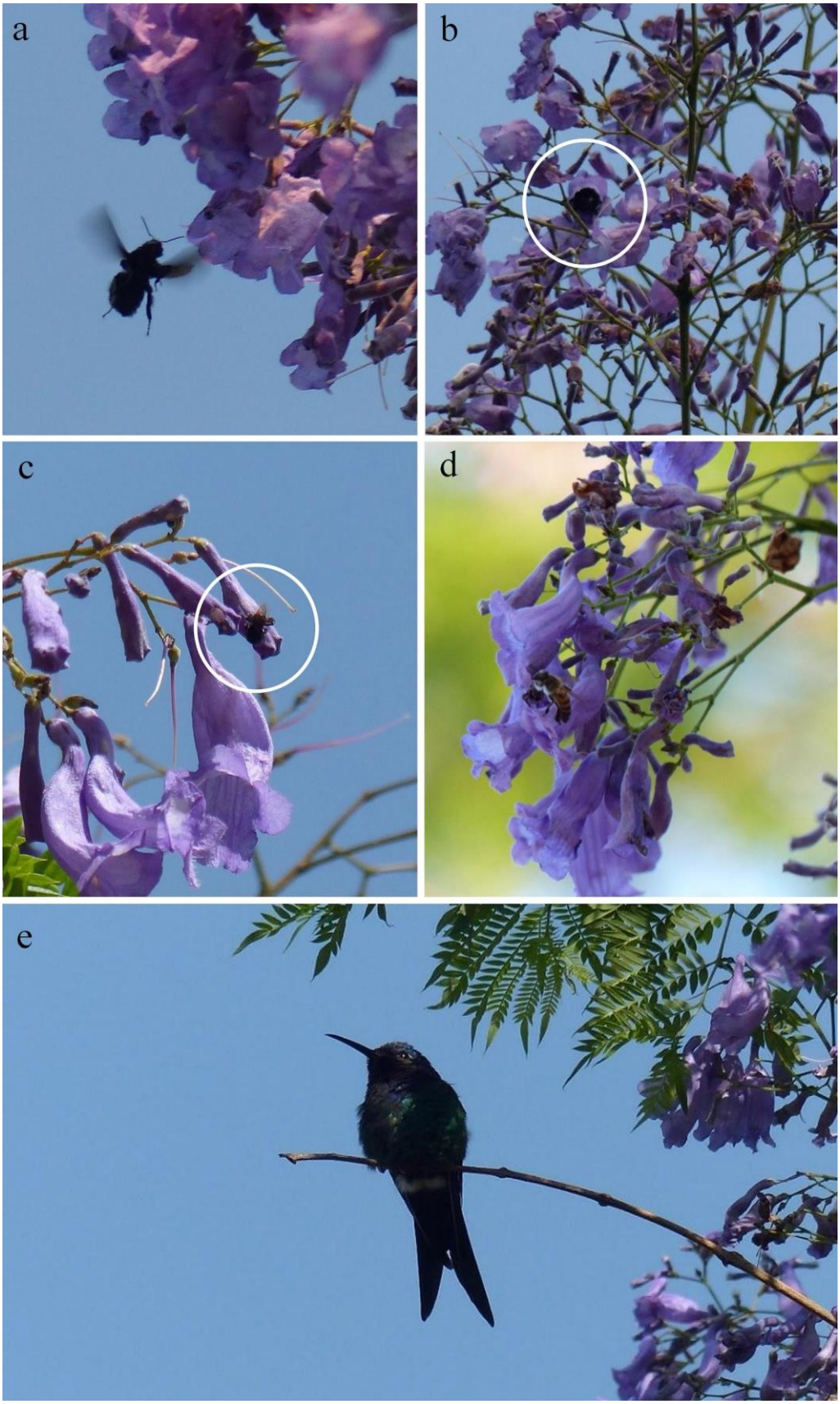


Figure 4 – Main floral visitors of *Jacaranda mimosifolia*. **a-b** *Bombus* sp. **c** *Trigona* sp. **d** *Apis mellifera*. **e** *Amazilia* sp. Photos: Katherine Lorena Rivera Hernández

Source: Authors.



Figure 5 – Flowers of *Jacaranda mimosifolia*. **a** Perforations at the base of the corollas indicated by arrows. **b** Detail of the perforation. **c** Perforated floral bud. Photos: Katherine Lorena Rivera Hernández

Source: Authors.

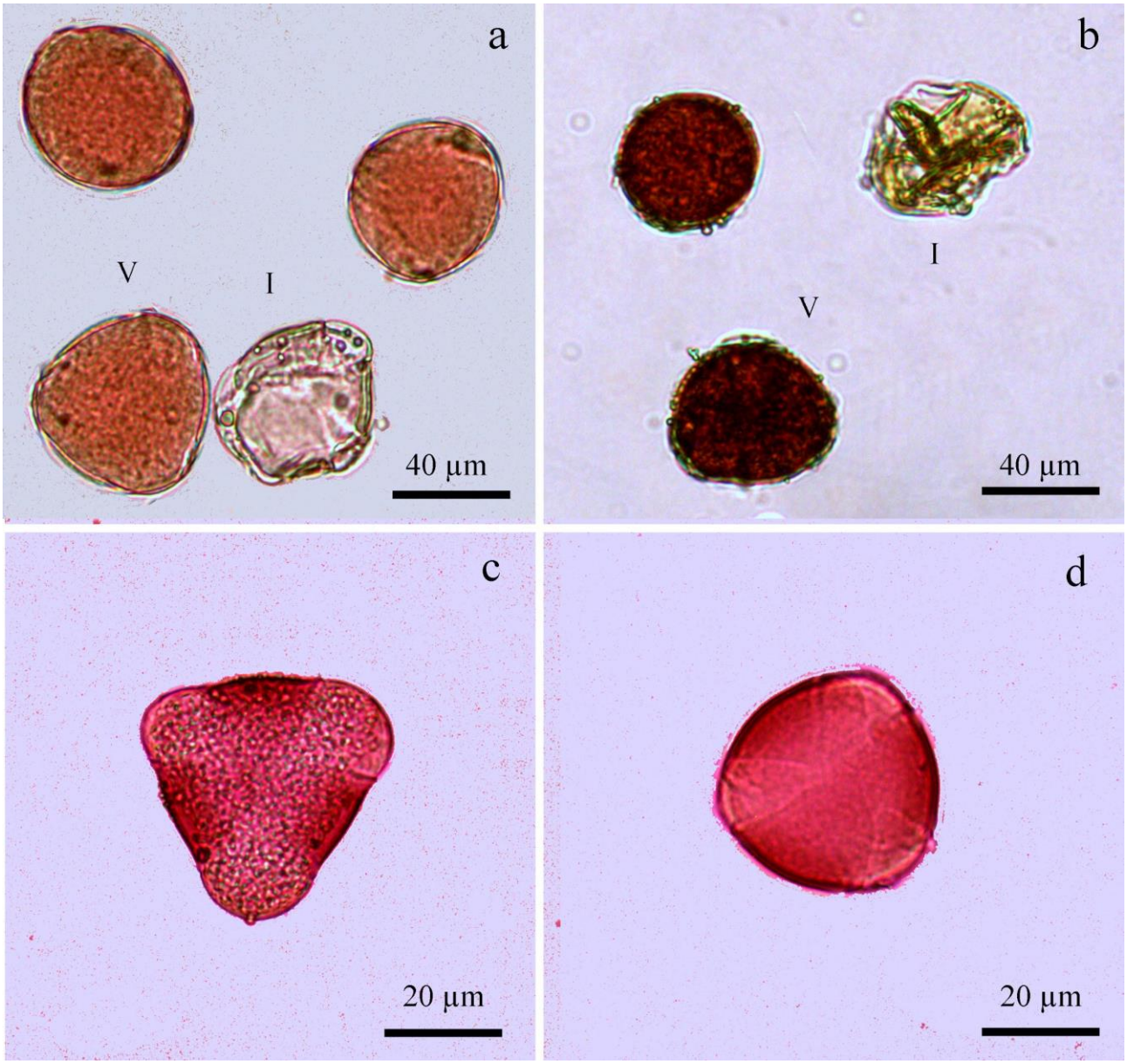


Figure 6 – Pollen viability for *Jacaranda mimosifolia* with dyes **a** 2% acetocarmine **b** 2% Lugol. Pollen grains in **c** equatorial view and **d** polar view

Source: Authors.

Author contributions

A. L. A. Chaves: Observation of Visitors, Pollen analysis, Conceptualization, Writing, Review and Editing. **K. L. R. Hernández:** Observation of Visitors, Writing, Review and Editing. **J. C. F. Silva:** Observation of Visitors, Review and Editing. All authors read and approved the final manuscript.

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Competing Interests

The authors declare that they all agree with this publication. There is no conflict of interest of any kind.

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