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High visitation rate of oil bees may increase pollination efficiency of *Couepia uiti* in Pantanal wetland

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Abstract – Pollen transfer to the stigma is the primary mechanism of sexual reproduction in plants. Among bee species that act as pollen vectors and pollinators, attributes such as floral visitation rate, synchrony with the receptive phase of the flower, compatibility between flower shape and foraging behavior, and morphological traits are often used to infer pollination efficiency. Herein, we evaluate visitation frequency and behavior of bee species on *Couepia uiti* (Chrysobalanaceae) flowers in the southern Pantanal, Brazil. Additionally, after experimental manipulation of flower visitation and by accompanying pollen tube growth in the pistil after one single visit to the flowers, we were able to pinpoint the roles of *Centris spilopoda* and *Apis mellifera* on the pollination of *C. uiti*. *Centris spilopoda* was the most important pollinator of *C. uiti* in the study area. In fact, we found that this oil bee might enhance plant reproductive success by almost 40% after a single visit. In addition to its high abundance, the efficiency of *C. spilopoda* as pollinator may be explained by its tight synchrony with pollen liberation and stigmatic receptivity of flowers of *C. uiti*.

bee activity and stigmatic receptivity / floodplain / Centridini / pollen tube / single visit

1. INTRODUCTION

Pollination is the mechanism whereby plants with flowers are sexually reproduced (Endress 1994; Proctor et al. 2003). For pollination to be effective (i.e., pollen tube growth occurs and reaches the ovules leading to their fertilization), pollen grains need to be transferred to stigma(s) that are reproductively compatible (Hiscock and Mcinnis 2003). Some plants are self-compatible (Brauner and Gottlieb 1987; Toräng et al. 2017),

but many species depend on xenogamic pollen for reproduction (Vilas Boas et al. 2013; Zazan et al. 2014). Xenogamic pollination, also known as cross-pollination, can influence fruit quality and seed viability (Schneider et al. 2009; Chautá-Mellizo et al. 2012; Stein et al. 2017).

Bees are one group of animals that are responsible for increased plant reproduction success via cross-pollination (Ollerton et al. 2011) and pollinate several species, especially in tropical areas (Endress 1994; Roubik 1989). An efficient bee pollinator may present, besides synchrony with the receptive phase of the flower, morphological characteristics that are compatible with flower shape, while flowers offer resources that are compatible with the physio-behavioral requirements of bees. Centridini bees (Hymenoptera: Apidae) have pantropical distribution (Moure et al. 2012) and present tight interaction with plants species that offer oil as

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floral resource (Vogel 1974, 1990; Vinson et al. 1997; Teixeira and Machado 2000; Martins et al. 2013). These bees are potential pollinators of several wild nectariferous and polliniferous species (e.g., Fischer and Gordo 1993; Aguiar et al. 2003; Aguiar and Gaglianone 2008), including some economically important plant crops such as *Anacardium occidentale* (Freitas and Paxton 1998; Holanda-Neto et al. 2002), *Bertholletia excelsa* (Cavalcante et al. 2018), *Malpighia puniceifolia* and *M. emarginata* (Raw 1979; Oliveira and Schlindwein 2009, respectively), *Passiflora alata* and *Solanum lycopersicum* (Gaglianone et al. 2010; Gaglianone et al. 2018), *Psidium guajava* (Giannini et al. 2015), and *Trifolium pratense* (Macfarlane 2018).

Plants attract pollinators at a long distance through scents/odors as well as by the number of flowers, known as floral display. Other traits such as flower shape, flower color(s), and sugar concentration in the nectar may work as short distances attractants (e.g., Waser and Ollerton 2006; Ebeling et al. 2008). By presenting a greater floral display, plants may receive more floral visitors and, consequently, enhance fruit and seed production (Kearns and Inouye 1993; Mayfield et al. 2001). Information on fruit/seed set, as well as analysis of pollen tube growth in the style and ovary, are measurements employed to evaluate pollinator efficiency in several plant species (e.g., Motten 1986; Richards 1986; Zhang et al. 2015).

Pollen tube growth in the pistil results from the deposition of compatible pollen grains onto the stigmatic region of the flower. Despite polarized and continuing growth towards the ovule after emergence in the stigma, pollen tube growth can be interrupted a posteriori due to late acting self-incompatibility (Seavey and Bawa 1986). However, even with late incompatibility, the presence of a pollen tube indicates the pollinators' efficiency in carrying pollen to the stigma.

Couepia uiti (Mart. and Zucc.) Benth. (Chrysobalanaceae) is a self-incompatible plant species relying in cross-pollination to produce fruits (Paulino-Neto 2007). This plant species presents wide distribution in Brazil (Grandtner and Chevrette 2013), usually occurring on floodplains of small rivers and streams (Pott and Pott 1994).

In the Pantanal, which is the world's largest tropical wetland (Assine et al. 2015), it grows as a pioneer treelet, with a height of 3–6 m and a wide canopy that nearly reaches the ground. It flowers between August and November (Pott and Pott 1994), and fruits, which are consumed by humans (Bortolotto et al. 2018), birds, and other animal species, are produced around April–May during flood season in the Southern Pantanal. Flowers are polystemonous and uniovulate. A previous study about the breeding system of *C. uiti* reported continuous nectar production with increased sugar concentration around 10:00 am. Pollen liberation starts around 9:30 am and stigmatic receptivity at 12:00 am. Manual cross-pollination resulted in 11% fruit set (xenogamy). In natural conditions, where flowers are mostly visited by honeybees, fruit set did not exceed 15% (Paulino-Neto 2007).

Herein, we present data about the foraging behavior of several bee species and evaluate the visitation rates of the most frequent species. Furthermore, as *Couepia uiti* presents several different floral visitors (Paulino-Neto 2007), we test if pollen transfer efficiency differs between two frequent bee species. To answer this question, we carried out a field experiment to evaluate the pollination success of a native solitary oil bee and the exotic eusocial honeybee by detection of pollen tube growth in the style after one single flower visit.

2. MATERIAL AND METHODS

2.1. Study area

This study was carried out in the southern Pantanal, sub-regions of Miranda and Abobral, in a partially flooded area of the Abobral river (19° 28' 34.7" S, 57° 02' 37.8" W) where trees of *C. uiti* are naturally intermingled with the shrub *Byrsonima cydoniifolia* A. Juss. (Malpighiaceae).

Field observations took place between the 18th and 21st of October 2007 between 07:00 am and 4:00 pm. From the 16th–19th of September 2008, we carried out additional observations of the same population of *C. uiti*, but we did not evaluate visitor frequencies. During both years, field observations coincided with the end of the dry season in the area.

2.2. Floral visitors and pollinators

First, flower visitors were sampled and recognized for 2 h during 4 days (total 8 h) before starting the experiment. Once we identified the four most frequent bee species visiting the flowers, we started recording their visitation frequencies. We recorded visitation between 7:00 am and 16:00, with 10 min of focal observations during each hour, totaling 100 min day⁻¹, and in 4 days totaling 400 min of observation.

We recorded the number of visits by female bees in permanent plots of 1 m² at the edge of flowering trees, with the center point at the height of the observer's eyes (1.75 m). We only recorded female visits because floral visits by males were unclear. Males were observed taking nectar and also tried to approach females during their visits. Each flowering tree was divided into four plots, one northern, southern, eastern, and western to delimitate spatial areas where bees were recorded. Plots ($n = 16$) were marked with striped tapes. All sampled plots were assorted before sampling. The plants used for counting visitation frequencies were at least 30 m apart from each other. Visitation frequencies were calculated based on the number of visits without distinction between the resource (pollen or nectar) collected by the bee. To compare the total number of visits by each of the four most frequent bee species, we used Kruskal-Wallis test, with post hoc test. To compare frequencies of those bees throughout the observation period, we used a chi-squared test. We recorded bee behavior during their visits (e.g. landing on flowers, contacting reproductive parts, collecting resources, kind of resource collected) in the field and through photographs. We categorized pollinators according to their visitation frequency and behavior as follows: Major pollinator (MP) was assigned to species which presented the highest visitation frequencies and contacted reproductive floral parts during visits; pollinator (PO) was assigned to species which contacted reproductive parts of the plants but had lower visitation frequencies than species in the previous category; and robber (RO) to floral visitors that did not contact floral reproductive structures. All bee species were assigned to their social behavior according to Michener (1974).

After we recorded visitation and behavior, we collected bees using an entomological net. All collected specimens were killed in a jar with ethyl acetate vapor, mounted, and deposited into the zoological collection at the Universidade Federal de Mato Grosso do Sul (ZUFMS). Some specimens were deposited into the Coleção Entomológica Paulo Nogueira Neto, Departamento de Ecologia, Instituto de Biociências, Universidade de São Paulo. We also collected bees that we observed occasionally foraging on flowers of *C. uiti* but did not use them for frequency analysis.

2.3. Pollen viability

To analyze pollen viability, we used the cytoplasmic coloration method (Radford et al. 1974), using pollen grains collected at 10:00 am. We prepared five slides with the pollen of three flowers from different *C. uiti* individuals. Slides were prepared in the field using acetic carmine stain. We evaluated the percentage of viable grains (i.e. stained pollen grains) under a microscope.

2.4. Pollination efficiency

We performed field experiments for 4 days to evaluate the efficiency of visitors transferring pollen grains during a single visit, using the following protocol: (1) pre-anthesis flowers were bagged with non-woven pollination bags to allow floral ventilation and to prevent visitation, (2) we unbagged flowers, and (3) we allowed a single visit of a given bee (which we identified) to the flower, immediately after the visit the flower was re-bagged and tagged for analysis of pollen tube growth.

The flowers were unbagged at 12:00 am (period of stigmatic receptivity, see Paulino-Neto 2007), and single visits were recorded until 2:00 pm. We carried out this experiment for a maximum time of 2 h. Unvisited flowers or flowers that we were unable to evaluate bee visitation behavior were discarded. We collected experimental flowers in two periods: 24 h and 48 h after bee visitation. Flowers were sampled at two intervals to account for potential slow pollen tube growth, which could not be detected in the first 24 h after pollination, as well as to detect late incompatibility. Immediately after collection, flowers were

immersed in FAA 50 (formaldehyde, acetic acid, and ethanol 50%; Johansen 1940) and stored in ethanol 70%. In the laboratory, flowers were transferred and stored in alcohol 70%. Then, the pistils were isolated from the rest of the flower, placed in Beaker, and heated (95 °C) with alcohol 95%. Afterwards, the pistil was removed from alcohol and placed in a solution of 1:1 alcohol 95% and NaOH 30% for 3 min. on an electric heater (Dizeo de Strittmatter 1973). Pistils were then transferred to 50% bleach for clearing. After being diaphanized, pistils were placed on slides and stained with aniline blue 0.1% and then analyzed under a fluorescence microscope with light filter "A" (Martin 1958). These analyses were used to detect pollen tube growth in the pistil. Pollination efficiency (flowers with the presence of pollen tube in the style after a single visit) was compared between two visitors: the native *Centris pilopoda* and the exotic honeybee *Apis*

mellifera. Frequencies of pollen tube growth from periods after flower visitation (24 h and 48 h after flower visitation) were compared using chi-squared test.

3. RESULTS

We recorded 14 bee species visiting the flowers of *C. uiti*, as well as non-identified wasps and two hummingbird species. The bees searched for both nectar and pollen in the flowers. All sampled bees belong to the family Apidae, in the following tribes: Apini, Bombini, Centridini, Ericrocidini, Meliponini, Rhathymini, and Xylocopini (Table I). Six bee species (total 42%) belonged to Centridini, two species to Meliponini, while all other tribes had only one species (Table I). We recorded one single exotic species (*Apis mellifera* Linnaeus, 1758) visiting the flowers.

Table I. Bees recorded visiting flowers of *Couepia uiti* (Chrysobalanaceae) in southern Pantanal Miranda and Abobral regions, MS, Brazil.

Tribe	Species	Gender	SB	Resource	Behavior	KR
Apini	<i>Apis mellifera</i> Linnaeus 1753	F	Eusocial	N/P	PO	C
Bombini	<i>Bombus</i> sp.	F	Primitively eusocial	N	PO	O
Centridini	<i>Centris aenea</i> Lepeletier, 1841	F	Solitary	N	PO	O
	<i>Centris pilopoda</i> Moure, 1969	F/M	Solitary	N/P	MP	C
	* <i>Centris vittata</i> Lepeletier 1841	F/M	Solitary	N	PO	R
	<i>Epicharis nigrita</i> (Friese 1900)	F/M	Solitary	N	PO	O
	* <i>Epicharis zonata</i> Smith 1854	F	Solitary	N	PO	R
	<i>Epicharis xanthogastra</i> Moure and Seabra 1959	F/M	Solitary	N/P	PO	C
Ericrocidini	<i>Mesoplia rufipes</i> (Perty 1833)	F	Cleptoparasite	N	PO	C
Meliponini	<i>Nannotrigona aff. melanocera</i> (Schwarz, 1938)	F	Eusocial	N/P	RO	C
	<i>Trigona spinipes</i> (Fabricius, 1793)	F	Eusocial	P	RO	O
	<i>Paratrigona lineata</i> (Lepeletier 1836)	F	Eusocial	P	RO	O
Rhathymini	<i>Rhathymus bicolor</i> Lepeletier and Serville, 1828	F	Cleptoparasite	N	PO	O
Xylocopini	<i>Xylocopa</i> sp.		Facultative social	N	PO	O

Gender (M = male, F = female), social behavior (SB), floral resource (N = nectar, P = pollen), and the distinction between visitors (MP = main pollinator, PO = pollinator, RO = robber); KR = kind of species record (C = recorded during censuses, O = occasional, R = rare)

*Species sampled in 2008

Some bees visited flowers at 07:00 am when they were observed collecting nectar. At this time, bees searched for resources from non-functional flowers from the previous day or new flowers beginning anthesis. The most frequent visitors were *Centris spilopoda* Moure, 1969 (Centridini), followed by *Epicharis xanthogastra* Moure and Seabra, 1959 (Centridini), *Apis mellifera* (Apini), and *Nannotrigona aff. melanocera* Schwarz, 1938 (Meliponini) (Figure 1). Males of some bee species were also recorded foraging for nectar on

the flowers of *C. uiti* during our censuses and may have occasionally acted as pollinators (Table 1).

3.1. Behavior during floral visitation

Centris spilopoda and *E. xanthogastra* visited flowers by landing frontally on the reproductive structures to access the hypanthium after bending stamens and pistil laterally. During the visit, they contacted the anthers with the ventral region of the meso and metasoma, as



Figure 1. Pollinators and floral visitors of *Couepia uiti* in the Pantanal of Miranda-Abobral, Mato Grosso do Sul, Brazil. Females of *C. spilopoda* visiting flowers of *C. uiti* (upper and middle pictures). Female of *Epicharis xanthogastra* collecting nectar from the flower (bottom picture on the left side) and *Nannotrigona aff. melanocera* stealing pollen (bottom picture on the right side).

well as the legs (Figure 1). *Apis mellifera* was observed foraging for pollen and rarely accessed the hypanthium. During visits, *A. mellifera* landed laterally and frontally on anthers and collected pollen grains with the first pair of legs, while walking on the anthers. Pollen grains adhered to the ventral region of the mesosoma and metasoma and transferred to the corbicula during flight. *Nannotrigona aff. melanocera* visited flowers by landing directly on the anthers where it collected pollen, with pollen grain transfer like honeybees. Due to its small size, the bee easily accessed the hypanthium but rarely contacted the stigmatic region. Large unidentified (species level) bees, such as *Xylocopa* and *Bombus*, were occasionally recorded above sampling plots, drinking nectar in a trap line route from plant canopies. Social behavior, role in pollination, and resources collected from flowers are summarized in Table 1.

3.2. Visiting frequency

Among the four most frequent bee species, we recorded a total of 1061 visits to flowers of *C. uiti*. Over 78% of the visits were performed

exclusively by *C. spilopoda* ($n = 829$, $X = 83 \pm 43.5$ visits h^{-1}), followed by *E. xanthogastra* ($n = 134$, $X = 13.4 \pm 12.6$ visits h^{-1}), *A. mellifera* ($n = 67$, $X = 6.7 \pm 7.66$ visits h^{-1}), and *N. aff. melanocera* ($n = 31$, $X = 3.1 \pm 4.93$ visits h^{-1}). The overall visiting frequency was significantly different (Kruskal-Wallis test, $\chi^2 = 27.177$, $df = 3$, $p < 0.001$) and frequency of *Centris spilopoda* significantly higher than all other pollinators ($p < 0.05$, for all comparisons) (Figure 2).

Centris spilopoda presented two visitation peaks, the first at 11:00 am and the second at 14:00. *Epicharis xanthogastra* had the same visitation peaks as *C. spilopoda*, but lower frequency. *Apis mellifera* concentrate floral visits in the early morning. Visits by the smallest bees, *N. aff. melanocera*, were always discrete (Figure 3). Visits to flowers did not change throughout the day (from 7:00 to 16:00) within a given species (Kruskal-Wallis test, $\chi^2 = 9$, $df = 3$, $p = 0.437$).

3.3. Pollen viability and efficiency of visits

The pollen viability after accounting 1003 pollen grains was approximately 90%. Of the 40 flowers that were visited by a single

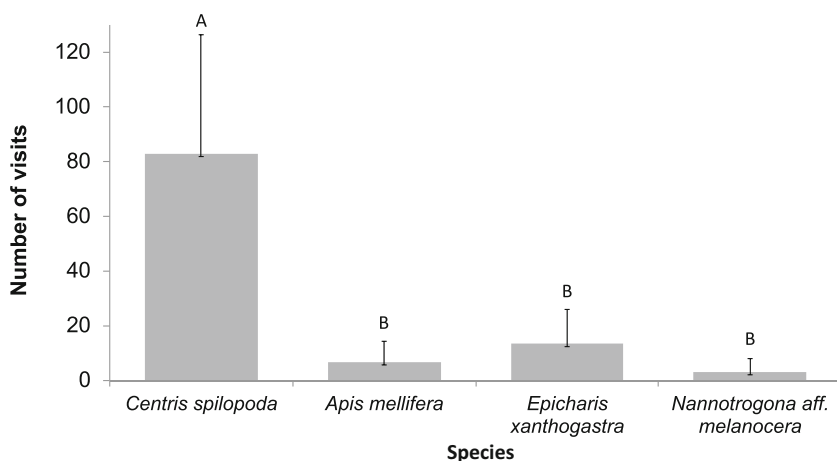


Figure 2. Number of visits of the four most frequent floral visitors (*Centris spilopoda*, *Apis mellifera*, *Epicharis xanthogastra*, and *Nannotrigona aff. melanocera*) of *Couepia uiti* flowers. *Centris spilopoda* was more frequent than all other visitors together (post hoc test, $p < 0.05$). Different letters on the bars indicate significant differences.

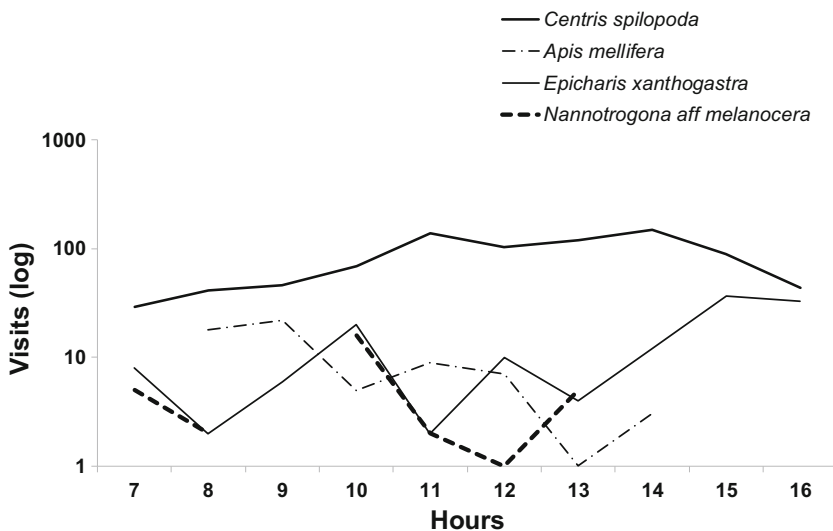


Figure 3. The visits (log) of the four floral visitors of *Couepia uiti* in 10 min periods between 7:00 and 16:00 in the Pantanal of Miranda-Abobral, MS, Brazil.

pollinator, 35 were visited by *C. spilopoda*, while the other five were visited by *A. mellifera*. We found that in 14 pistils pollen tubes grew after a single visit of *C. spilopoda*, with six in the treatment of 24 h ($n = 16$) and eight after 48 h ($n = 19$). None of the flowers visited by *A. mellifera* presented pollen tube growth (Table II). Pollination efficiency, however, was not significantly different across the native oil bee *Centris spilopoda* and the honeybee *A. mellifera* ($\chi^2 = 3.0769$, $df = 1$, $p = 0.07941$). The number of pollen tubes that grew in an interval of 24 h did not differ from the number

that grew after 48 h ($\chi^2 = 0.286$, $df = 1$, $p = 0.593$).

4. DISCUSSION

We found that one single visit to flowers of the self-incompatible *Couepia uiti* may enhance pollination. This finding may be related to species-specific interactions linked to the number of individuals foraging as well as to high displacement among different flowers patches. Centridini bees were the most frequent visitors to flowers of *C. uiti*. Due to their behavioral approach, visitation rate, and successful transfer of pollen grains to stigma, these bees were considered efficient pollinators for this plant species in our study. *Centris spilopoda* was the most important pollinator due to its high abundance of individuals and visitation frequency to the flowers. This oil bee presented two visitation peaks throughout the day, which coincided with an increased solute concentration of nectar and stigmatic receptivity (Paulino-Neto 2007). This species also contacted floral reproductive structures during visits promoting cross-pollination, which resulted in pollen tube growth in the pistils. We observed pollen tube growth in 14 (out of 35) flowers that were treated with a single visit. This suggests that if

Table II. Pollen tube growth after 24 and 48 h (number of treated flowers) recorded for two pollinators in a single floral visitation event.

	<i>Centris spilopoda</i>		<i>Apis mellifera</i>	
	24 h (16)	48 h (19)	24 h (3)	48 h (2)
PTG	37.5%	42%		
WPT	62.5%	58%	100%	100%

Note that a single visit of *Centris spilopoda* can increase fruit set by 40% in *Couepia uiti*

PTG pollen tube growth, WPT without pollen tube

C. uiti does not have late incompatibility, fruit production can increase by 40% after one single visit by *C. spilopoda*.

Yet, *A. mellifera* presented lower visitation frequency, concentrated in the first hours of the morning, and probably contributes poorly to the pollination of *C. uiti*. Furthermore, this bee may negatively affect *C. uiti* reproduction since pollen removal occurs before full stigmatic receptivity at 12:00 am. For *Clusia arrudae* Planch. and Triana (Clusiaceae), 90% of the pollen grains were removed by *A. mellifera*, which reduced seed production (Carmo et al. 2004). Thus, pollen grain collection time by exotic bees, which occurs before full stigmatic receptivity, could cause a pollen deficit during the high foraging periods of effective pollinators, reducing fruit and seed set. Moreover, the early foraging behavior for pollen recorded in this study might also represent a case of competition with the other native bees, as *A. mellifera* might reduce their pollen foraging success. Pollen loss has been recognized to play an important role in nutrition of solitary bee species (Müller et al. 2006). In addition, it has been shown that depletion of resources due to foraging of honeybees affect negatively other bee species (Roubik 1978; Paine 2004; Hudewenz and Klein 2015).

Apis mellifera is known for floral constancy, generally visiting flowers that are located close together, reducing the possibility of pollen transfer between non-related individuals (Eisikowitch 1998; Karron et al. 2009). If in one hand only 15% of fruit production in natural condition has been reported to an area with high predominance of honeybees (Paulino-Neto 2007), our estimation (40%) with native pollinator exceeds the previous values in at least 2.6 times. This visitation behavior was already indicated as negatively affecting the reproduction of some plants, especially species that depend on cross-pollination (Gibbs 1988; Camillo 1996; Sáez et al. 2014). However, in some systems, *Apis mellifera* may help native pollinators enhance fruit set (e.g., Freitas and Paxton 1998).

Cross-pollination, even at low rates, is considered essential to maintain populations of self-incompatible plant species (Bawa 1974). Bees of the genera *Xylocopa* and *Bombus* visited flowers

of *C. uiti* at low frequencies, following trap line routes. Nonetheless, due to their size, the ventral portion of their bodies make broad contact with the floral reproductive structures and seem to efficiently pollinate in single visits. This pollination efficiency for the genus *Xylocopa* has already been recorded for *C. uiti* in the Nhecolândia sub-region of the Pantanal (Paulino-Neto 2007).

Mesoplia rufipes made few visits to the flowers of *C. uiti*, feeding on nectar only. According to Roubik (1989), bees of this genus are rarely found on flowers. Our record of this species and *Rathymus bicolor*, both cleptoparasites (Snelling 1984), visiting *C. uiti* flowers, may be due to the underground nests of *Centris* spp. and *Epicharis* spp. in the habitat surrounding the study area. Besides adequate soil structure for nest aggregation (SB pers. observ.), the high flux of Centridini in the area, and consequent cleptoparasites, may have been caused by the co-occurrence and simultaneous flowering of *C. uiti* and *Byrsonima cydoniifolia* (Malpighiaceae). The synchronized flowering of these two species provided nourishment for individuals and brood of Centridini bees, potentially making them abundant in the area. Nectar and pollen offered by *C. uiti* can feed adult and immature bees, and the oil offered by *B. cydoniifolia* flowers can be used to waterproof material to construct brood cells (Vinson et al. 2006), which is important for bees nesting in floodplain areas and for the diet of immature bees (Alves-dos-Santos et al. 2007).

4.1. Pollen tube growth after single visits

We recorded pollen tube growth in 40% of the 35 pistils with one single visit by *C. spilopoda*. After 24 h of pollen transfer to the stigma, the pollen germinated on the stigma and pollen tube growth already occurred in the pistil (see supplementary material, Fig. S1). No significant difference between the number of pollen tubes at 24 h and 48 h may indicate that this species does not present late incompatibility, or that late incompatibility occurs even later (after 48 h). The limited number of pistils that presented pollen tube growth (14 out of 35) could be due to the fact that only part of pollen on visitors (*C. spilopoda*) originated from a non-related

individual. Thus, low frequency of pollen tube growth could be a result of lack of pollen deposition on the stigma during the visit (pollinator inefficiency) or incompatibility between the pollen donor and receiver (Bedinger et al. 2017) potentially due to displacement behavior among flower patches. We also emphasize that ovary incompatibility (Gibbs 1988, 2014) was not evaluated since flowers were removed before fruit formation. Although the observation of pollen tube growth is an appropriate method for studying pollinator/pollen transfer efficiency (Motten 1986; Richards 1986), our findings are not conclusive regarding the effects on the reproduction success of *C. uiti*. So, high visitation frequency (or movement) of pollinators, as observed in our study, may increase the deposition of pollen grains onto the stigma (Sáez et al. 2014) and the chances of effective cross-pollination (Gorenflo et al. 2017). Since *C. uiti* flowers are polystemonous, they produce a large amount of pollen and, since they are uniovulate, they only need one viable pollen grain to be pollinated. We did not have evidence (0%) of pollen tube growth from a single visit of *A. mellifera*. Our findings support the importance of native species as pollinators of xenogamous species; however, further studies are needed to clarify the pollination success mediated by native pollinators and the negative effects of the exotic *A. mellifera* on *Couepia uiti*.

Self-incompatible uniovulate flowering plants can theoretically develop fruit after non-related conspecific pollen grains are deposited onto stigmas if pollen grain and stigma are functional. Although we did not find significant differences between pollen transfers resulting in pollen tube growth in the stigma, none of the flowers visited by honeybees were pollinated. In addition, its massive foraging behavior adjusted mainly to the period of pollen presentation and before stigmatic receptivity may characterize honeybees as resource thieves, affecting negatively plant reproduction. On the other hand, *C. spilopoda* presented tight synchrony with stigmatic receptivity, high abundance of foragers, and therefore was the most important pollinator of *Couepia uiti*. In fact, we found that this oil bee might enhance plant reproductive success by almost 40% after one single visit.

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AUTHOR CONTRIBUTIONS

SB and ACA conceived the study and designed experiment; SB performed experiments and analysis; SB and GMP analyzed pollen tube; SB, AP, and ACA led the writing of the study. All authors read and approved the final manuscript.

COMPLIANCE WITH ETHICAL STANDARDS

Conflict of interest The authors declare that they have no conflict of interest.

Un taux de fréquentation élevé des abeilles à huile peut augmenter l'efficacité de la pollinisation de *Couepia uiti* dans les zones humides du Pantanal

Activité des abeilles et réceptivité stigmatique, plaine inondable, Centridini, tube pollinique, visite unique

Hohe Visitationsrate von Ölbienen könnte die Bestäubungseffizienz an *Coupeia uiti* in Feuchtgebiete vom Pantanal erhöhen

Bienenaktivität und Aufnahmefähigkeit des Stigmas, Überschwemmungsgebiet, Centridini, Pollenschläuche, Einzelbesuche

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