Sociobiology 66(1): 66-74 (March, 2019) DOI: 10.13102/sociobiology.v66i1.3474

Sociobiology
An international journal on social insects

RESEARCH ARTICLE - BEES

Presence of *Plebeia* aff. *flavocincta* Nests in Urban Areas

MF Ribeiro, TA Taura

Embrapa Semiárido, Petrolina, Pernambuco, Brazil

Article History

Edited by
Denise Alves, ESALQ-USP, Brazil
Received 11 May 2018
Initial acceptance 20 June 2018
Final acceptance 20 September 2018
Publication date 22 April 2019

Keywords
Meliponini, nesting sites, urban ecology.

Corresponding author
Márcia de Fátima Ribeiro
Embrapa Semiárido
BR 428, Km 152, Zona Rural
CP 23, CEP 56302-100, Petrolina-PE, Brasil.
E-Mail: marcia.ribeiro@embrapa.br

Abstract

This study was performed in 2009, 2015 and 2018, at urban areas of Petrolina (state of Pernambuco) and Juazeiro (state of Bahia), at the São Francisco valley, Brazilian Northeast. Trees were identified and investigated for the presence of *Plebeia* aff. *flavocincta* nests, in three years: 2009, 2015 and 2018. Data on height at nest entrance (HNE), tree diameter at nest entrance (DNE) and tree diameter at breast height (DBH) were obtained. Trees containing nests were identified and geo-referenced. In Petrolina and Juazeiro, the percentage of bees nests in all checked trees was smaller in 2009 (3.94% and 0.56%) than in 2015 (1.92% and 5.26%) and in 2018 (21.21% and 3.66%). This increase (especially in Petrolina) suggests *P*. aff. *flavocincta* is well adapted to urban environments and food and/or nesting resources might be improved along the years. On the other hand, survival of nests was not high: only two nests found in 2009 survived up to 2018. Mortality of nests was mainly due to the cutting of trees. Simultaneously, according to observations, swarming probably occurs, what might have improved the number of nests observed in 2018. The vegetal species most used by the bees was *Prosopis juliflora* (84.38%). Considering all data, the HNE, DNE, and DHB varied among the cities and years, but the differences were not significant in most of the cases. In Juazeiro, two nests were found also in a wall and another one in a pipe showing the diversity of nesting habits of the species. It is remarkable that despite high levels of stress (caused by noise, pollution, and human interferences) these bees are able to survive and swarm at urban areas.

Introduction

Stingless bees make their nests in a variety of substrates, such termite mounds, spaces in walls or rocks, cavities made by other animals, etc. (Roubik, 2006). However, most of them nidify inside hollows of trees. This is the case of a small stingless bee, *Plebeia* aff. *flavocincta* Cockerell, 1912, popularly called 'mosquito' at Northeast of Brazil.

In previous surveys, it was found that *P*. aff. *flavocincta* is quite rare at rural areas and more abundant at urban areas (Ribeiro et al., 2009). Urban areas can be inadequate for several animals, including bees. In such fragmented habitats, studies indicated there is a tendency for reduction in the bee species composition, richness and number of floral specialists, but also an increase of cavity-nesting bees (Cane et al., 2006; Hernandez et al., 2009).

On the other hand, urban habitats may provide many ecological niches, food resources with high availability and diverse environments (Banaszak-Cibicka, 2014). Besides, it can bring some advantages, as smaller interspecific competition, decrease of predation and less interference caused by climatic variations (Torres et al., 2014). Moreover, some bee species may indeed become abundant in urban areas due to increase in the potential nesting sites and food resources (mostly provided by cultivated plants) and even by the elimination of competitors, as for example, *Apis mellifera* L. colonies (Taura & Laroca, 1991; 2001). A study with an Australian stingless bee (*Tetragonula carbonaria* Smith) showed that the foraging activity and the resource intake was indeed, improved in gardens that were rich in flowers (Kaluza et al., 2016).

Open access journal: http://periodicos.uefs.br/ojs/index.php/sociobiology
ISSN: 0361-6525

In Brazil, bees have been studied in urban areas concerning aspects as food resources (Knoll et al., 1993; Taura & Laroca, 2001; Faria et al., 2012; Aleixo et al., 2013; Antonini et al., 2013) and nesting behavior (Taura & Laroca, 1991; Alvarenga, 2008; Araújo et al., 2016). Studies on the nesting habits of bees may, in fact, be useful for predicting how bees respond to habitat fragmentation in urban areas (Cane et al., 2006). Monitoring bee populations in urban areas may be useful for identifying the community dynamics and providing information for bee conservation (Hernandez et al., 2009). Moreover, knowing vegetal species that are useful for bees as nesting sites may help in urbanization planning and increase of green spaces in cities. In this way, the objective of this study was to monitor the presence of *P.* aff. *flavocincta* nests in two urban areas in a tropical semiarid environment throughout years.

Material and Methods

Trees at urban areas of Petrolina and Juazeiro in Northeast Brazil were examined to detect the presence of nests of *Plebeia* aff. *flavocincta,* a stingless bee species with natural occurrence in the region (Ribeiro et al., 2009). Petrolina (state of Pernambuco) and Juazeiro (state of Bahia) are two cities that have about 345,000 and 220,000 inhabitants, respectively, separated by the São Francisco river and located at a semiarid region inserted in the Caatinga biome. We searched for *P.* aff. *flavocincta* nests inspecting trees randomly in public squares, sidewalks and on the river banks of the São Francisco. The trees were identified concerning their botanical species, by comparison with exsiccates of the Herbarium of the Embrapa Semiárido. In order to confirm whether they were native or not, we consulted the site of the Jardim Botânico do Rio de Janeiro (Flora do Brasil 2020). Eventually other substrates (as wall or pipe) were also checked when we were informed by the population on the presence of the bees nests.

In case a nest was found, it was geo-referenced using a GPS, and data on the height at nest entrance (HNE), tree diameter at nest entrance (DNE), and tree diameter at breast height (DBH) were obtained. Occasionally, the DBH was not obtained because the trunk was bifurcated (therefore, the sample size of this measure in table 2 is different from for HNE and DNE). In order to compare the diameter of trees with and without nests, we selected the tree species where we found the most number of bees nests and measured the DHB also from several individuals that had no nests.

The research was made in 2009 (February - in Petrolina and Juazeiro; also in March and June - Petrolina); 2015 (in August for both cities), and 2018 (February, for both cities, and still in April - Petrolina, and in May - Juazeiro). We used satellite maps to plot the points where trees were checked and nests were found, and to calculate the total area sampled. Since the same trees that had nests in 2009 were visited again in 2015 and 2018, it was possible to know whether the nests survived and/or the tree was cut down. Moreover, because some other trees in that were not checked in previous years were checked in 2018, we also discovered new nests. Because the sample of trees was different for each year and city (Table 1), a percentage for the presence of nests was calculated.

Table 1. Trees investigated for *Plebeia* aff. *flavocincta* nests in Petrolina and Juazeiro, in 2009, 2015 and 2018.

City	Year	Family	Tree species	Native tree species	Number of bee nests/ Number of trees
Petrolina	2009	Anacardiaceae	*Mangifera indica* L.	no	0/12
		Anacardiaceae	*Spondias tuberosa* Arruda	yes	0/1
		Annonaceae	*Annona squamosa* L.	no	0/2
		Apocynaceae	*Plumeria rubra* L.	no	0/1
		Aracaceae	*Phoenix dactylifera* L.	no	0/32
		Bignoneaceae	*Tabebuia caraiba* (Mart.) Bureau	yes	0/4
		Combretaceae	*Terminalia catappa* L.	no	0/4
		Crysobalanaceae	*Licania tomentos*a (Benth.) Fritsch	yes	0/5
		Fabaceae	*Prosopis juliflora* (Sw.) DC.	no	14/172
		Fabaceae	sp. 1		0/33
		Fabaceae	sp. 2		0/9
		Fabaceae	sp. 3		0/3
		Fabaceae	*Delonix regia* (Bojer ex Hook.) Raf.	no	0/6
		Fabaceae	*Libidibia ferrea* (Mart. ex Tul.) L. P. Queiroz	yes	0/2
		Fabaceae	*Senna* sp.	yes	0/2
		Fabaceae	*Caesalpinia echinata* Lam.	yes	0/2
		Fabaceae	*Tamarindus indica* L.	no	0/2
		Meliaceae	*Azadirachta indica* A. Juss	no	0/5
		Moraceae	*Ficus benjamina* L.	no	0/36
		Myrtaceae	*Syzygium jambolanum* (Lam.) DC.	no	0/8

Table 1. Trees investigated for *Plebeia* aff. *flavocincta* nests in Petrolina and Juazeiro, in 2009, 2015 and 2018. (Continuation)

City	Year	Family	Tree species	Native tree species	Number of bee nests/ Number of trees
Petrolina	2009	Myrtaceae	*Eucalyptus* sp.	no	0/8
		Rhamnaceae	*Ziziphus joazeiro* Mart.	yes	0/6
			total		**14/355**
	2015	Fabaceae	*Prosopis juliflora* (Sw.) DC.	no	1/103
		Bignoneaceae	*Spathodea campanulata* P. Beauv.	no	1/1
			total		**2/104**
	2018	Fabaceae	*Prosopis juliflora* (Sw.) DC.	no	12/63
		Bignoneaceae	*Spathodea campanulata* P. Beauv.	no	1/1
		Combretaceae	*Terminalia catappa* L.	no	1/1
		Fabaceae	*Tamarindus indica* L.	no	0/1
			total		**14/66**
Juazeiro	2009	Anacardiaceae	*Mangifera indica* L.	no	0/53
		Anacardiaceae	*Anacardium occidentale* L.	yes	0/1
		Anacardiaceae	*Spondias tuberosa* Arruda	yes	0/4
		Annonaceae	*Annona squamosa* L.	no	0/2
		Apocynaceae	*Nerium oleander* L.	no	0/4
		Aracaceae	*Phoenix dactylifera* L.	no	0/2
		Aracaceae	*Copernicia prunifera* (Mill.) H. E. Moore	yes	0/12
		Aracaceae	*Cocos nucifera* L.	no	0/6
		Combretaceae	*Terminalia catappa* L.	no	1/16
		Crysobalanaceae	*Licania tomentosa* (Benth.) Fritsch	yes	0/8
		Fabaceae	*Hymenaea courbaril* L.	no	1/2
		Fabaceae	*Inga edulis* Mart.	yes	0/76
		Fabaceae	sp. 2		0/27
		Fabaceae	sp. 3		0/10
		Fabaceae	*Delonix regia* (Bojer ex Hook.) Raf.	no	0/19
		Fabaceae	*Senna* sp.	yes	0/1
		Fabaceae	*Bauhinia forficata* Link	yes	0/2
		Fabaceae	*Erythrina velutina* Willd.	yes	0/1
		Fabaceae	*Prosopis juliflora* (Sw.) DC.	no	1/17
		Malpighiaceae	*Malpighia emarginata* D. C.	no	0/1
		Malvaceae	*Ceiba speciosa* (A. St.-Hill.) Ravenna	yes	0/1
		Meliaceae	*Azadirachta indica* A. Juss	no	0/1
		Moraceae	*Ficus benjamina* L.	no	0/81
		Myrtaceae	*Eucalyptus* sp.	no	0/4
		Myrtaceae	*Syzygium jambolanum* (Lam.) DC.	no	0/1
		Myrtaceae	*Psidium guajava* L.	no	0/1
		Oleaceae	*Jasminum* L.	no	0/2
			total		**2/355**
	2015	Fabaceae	*Prosopis juliflora* (Sw.) DC.	no	0/17
		Fabaceae	*Hymenaea courbaril* L.	no	1/2
			total		**1/19**
	2018	Fabaceae	*Prosopis juliflora* (Sw.) DC.	no	0/2
		Fabaceae	*Hymenaea courbaril* L.	no	0/2
		Myrtaceae	*Eucalyptus* sp.	no	3/78
			total		**3/82**

In order to test whether the number of nests in each year was significantly different and to make a comparison between the two cities considering tree measure variables (HNE, DNE and DBH), Kruskal-Wallis and Mann-Whitney tests were applied (PAST).

Results

Approximately an area of 16.50 ha, and more than 900 trees were checked in both cities in the three years. Figure 1 shows the points sampled in the search for the bees nests and the exact location of each bee nest found in 2009 (Fig 1a), 2015 (Fig 1b) and 2018 (Fig 1c). The observations at urban areas in both cities were made in 38 plant species (3 not identified at species level), belonging to 15 botanical families (Table 1). From these, 19 species, (54.29%) were exotic (cultivated or sub-spontaneous) and 16 species (45.71%) were native. (It was not possible to say whether the not identified species were native and, therefore, they were excluded for the calculus for the percentage, i.e., we used a total of 35).

Most of trees (between 79% and 99%) did not have a bee nest and the amount of bees nests varied along the years. In fact, nests of *P.* aff. *flavocincta* were found in a very low number of trees in the three years (Table 1). The percentages of trees containing nests in 2009 were 3.94% and 0.56%, respectively for Petrolina and Juazeiro. In 2015, the percentages were: 1.92% and 5.26%. And in 2018, they were 21.21% and 3.66%. Comparing the percentages of nests of the three years in Petrolina, first it decreased and then increased, respectively in 2015 and 2018. In Juazeiro, the number increased in 2015, but decreased in 2018.

The plant species most used by bees for nesting was *Prosopis juliflora* (Table 1). Figure 2 shows the percentage of bee nests in each tree in both cities and considering the three years. *P. juliflora* appears with high percentages in the three years in Petrolina, but in Juazeiro only in 2009. Juazeiro has fewer individuals of this plant species than Petrolina. When we consider all nests found in both cities and in all years, 84.38% of them were located in *P. juliflora* trees while 16.62% were distributed in four other vegetal species (*Spathodea campanulata*, *Terminalia catappa*, *Hymenaea courbaril*, and *Eucalyptus* sp.).

Concerning the variables measured for the trees, table 2 shows the obtained results.

The differences found for HNE were not significant among the cities and the years. The other variables measured (DNE and DBH) are dependent on the plant species, and even so they were not significantly different among cities and years in most of cases. The exception was the year of 2018: the trees of Petrolina had in average (2.14 ± 0.07 m, n = 14) significantly larger diameters than Juazeiro (1.33 ± 0.09 m, n = 3) (P = 0.003, H = 0, Table 2).

On the other side, when only *P. juliflora* is considered (taking into account both cities and the three years) the values for HNE, DNE and DBH were 1.25 ± 0.14 m (n = 28), 1.81 ± 0.10 m (n = 28), and 1.97 ± 0.07 m (n = 25), respectively. Comparing individuals of *P. juliflora* that had no nests (again taking into account both cities and the three years sampled), the value for DBH was 1.77 ± 0.07 m (n = 97). This difference was significant (P = 0.012, H = 812.5).

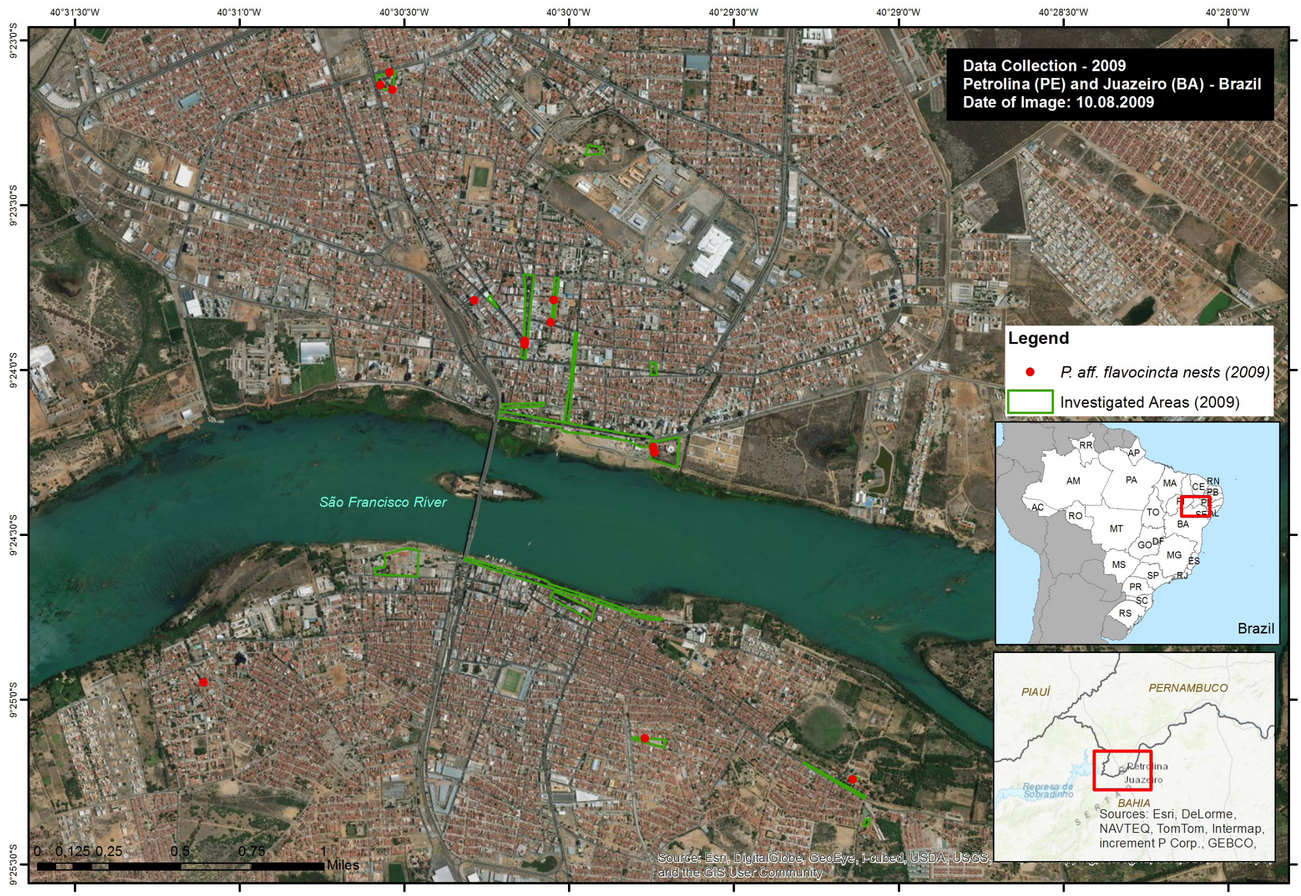
As already mentioned, besides hollow trees, stingless bees may use other substrates. In Juazeiro, two nests were also found inside a wall (in 2009 up to 2018) and another one in a pipe (2018). The HNE found for these nests were respectively: 0.30 m, 0.15 m, and 0.38 m.

The survival of bees nests varied, but in general was not so high. In Petrolina, from the 14 nests found in 2009, only one survived in 2015, and this one was dead in 2018. Some trees were cut down, and some others survived, but not the nests. From the two nests found in 2015, only one survived up to 2018. In Juazeiro, from the two nests of 2009, only one was alive in 2015, but was dead in 2018. The three ones found in 2018 were not searched in previous years, so they could be older. In fact, at one square of Petrolina in five months of observation (December 2017 to April 2018) performed for another study (unpublished information), three new nests appeared. The observations at nest entrance showed that workers were leaving the nests carrying cerumen what suggested that some nests were swarming.

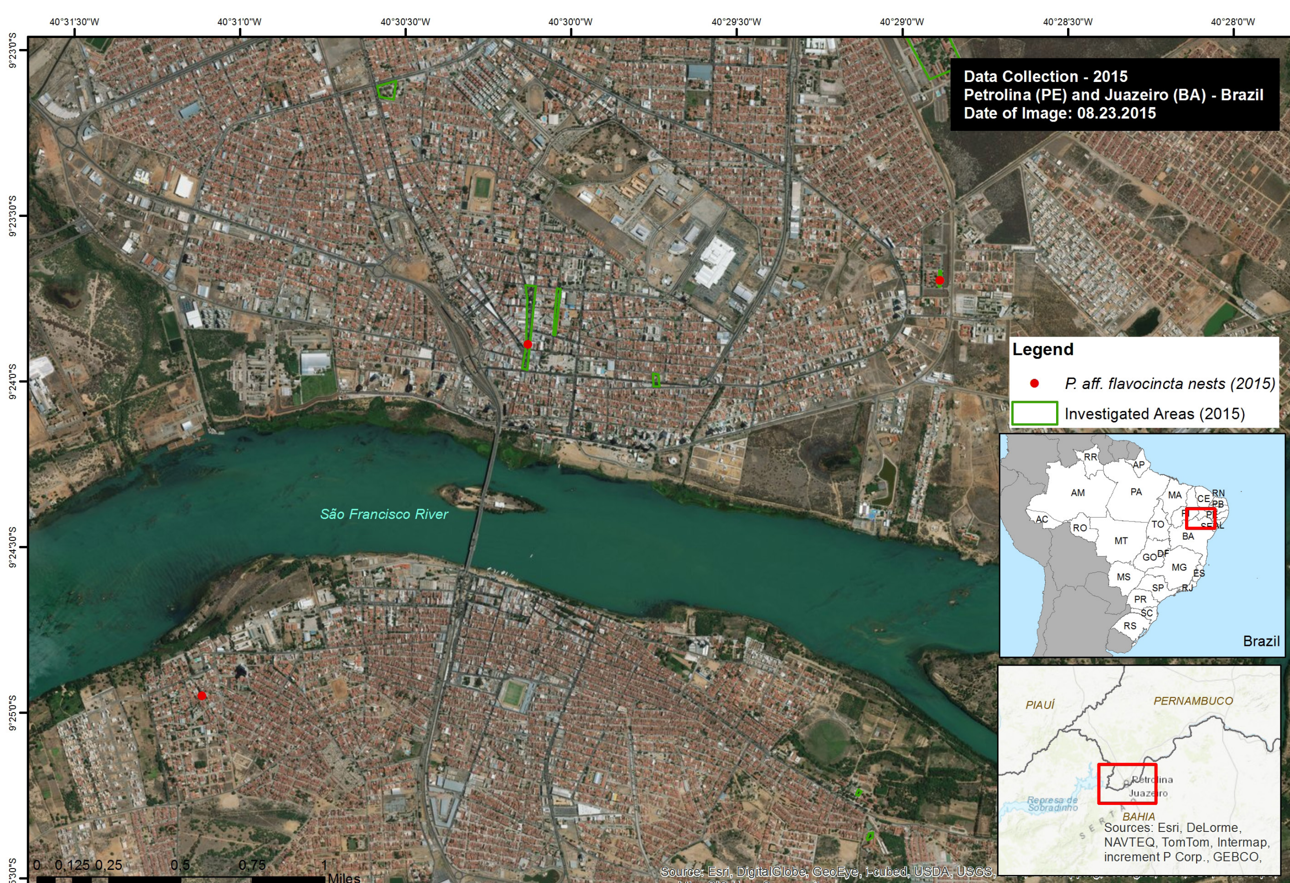
Table 2. Means (and standard errors) of measures obtained for nests of *Plebeia* aff. *flavocincta* in Petrolina and Juazeiro, in 2009, 2015 and 2018. Legend: n: number of nests.

City	Year	Height at nest entrance (m)	Diameter at nest entrance (m)	Diameter at breast height (m)
Petrolina	2009	1.01 ± 0.16 (n=14)	1.69 ± 0.15 (n=14)	1.84 ± 0.10 (n=12)
	2015	2.86 ± 1.41 (n=2)	1.15 ± 0.46 (n=2)	1.98 ± 0.37 (n=2)
	2018	1.79 ± 0.29 (n=14)	1. 95 ± 0.15 (n=14)	2.14 ± 0.07 (n=14)
Juazeiro	2009	1.45 ± 0.85 (n=2)	1.78 ± 0.38 (n=2)	4.30 (n=1)
	2015	2.29 (n=1)	2.15 (n=1)	4.30 (n=1)
	2018	1.02 ± 0.39 (n=3)	1.44 ± 0.05 (n=3)	1.33 ± 0.09 (n=3)

(a)


40°31'30"W
40°31'0"W
40°30'30"W
40°30'0"W
40°29'30"W
40°29'0"W
40°28'30"W
40°28'0"W
9°23'0"S
9°23'30"S
9°24'0"S
9°24'30"S
9°25'0"S
9°25'30"S
Data Collection - 2009
Petrolina (PE) and Juazeiro (BA) - Brazil
Date of Image: 10.08.2009
Legend
P. aff. flavocincta nests (2009)
Investigated Areas (2009)
São Francisco River
Brazil
PIAUÍ
PERNAMBUCO
BAHIA
Sources: Esri, DeLorme, NAVTEQ, TomTom, Intermap, increment P Corp., GEBCO,
0 0.125 0.25 0.5 0.75 1
Miles
Source: Esri, DigitalGlobe, GeoEye, i-cubed, USDA, USGS, and the GIS User Community

(b)


40°31'30"W
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9°24'0"S
9°24'30"S
9°25'0"S
9°25'30"S
Data Collection - 2015
Petrolina (PE) and Juazeiro (BA) - Brazil
Date of Image: 08.23.2015
Legend
P. aff. flavocincta nests (2015)
Investigated Areas (2015)
São Francisco River
Brazil
PIAUÍ
PERNAMBUCO
BAHIA
Sources: Esri, DeLorme, NAVTEQ, TomTom, Intermap, increment P Corp., GEBCO,
0 0.125 0.25 0.5 0.75 1
Miles
Source: Esri, DigitalGlobe, GeoEye, i-cubed, USDA, USGS, and the GIS User Community

(c)

Fig 1. Images of satellite that show GPS points where nests of *Plebeia* aff. *flavocincta* nests were sampled in Petrolina and Juazeiro, in 2009 (a), 2015 (b) and 2018 (c). Images were obtained from Google Earth.

Discussion

The number of bee nests was quite low in all years sampled in both cities. However, in Petrolina the increase of nests observed in 2018 was very high (from less than 4% to more than 20% of the investigated trees). This indicates that this species is well adapted to urban environment and/or the available food resources may have improved in this period. Although we did not studied the available floral resources along the entire period of this work in 2018 another study was done at the mentioned square in Petrolina, and we found 22 species in its trophic niche. Still, the number of trees did not increase in the city, but the trees already present could have enlarged in diameter, facilitating the nesting of bees. These facts may have contributed for the increase of nests observed. It seems that nesting sites as well floral resources may restrict nest density, besides richness and diversity of bees in communities (Silva et al., 2013).

Hollow trees are species-specific (Nogueira et al., 2006) and are formed due to branches breakage caused by wind or lightning, lianas, aging, fire and cavities caused by vertebrates (as birds) or insects (as termites, beatle larvae). These agents allow the attacks of fungi and bacteria to heartwood (Harper et al., 2005). Larger trees tend to have more irregular boles and hollow trunks (Nogueira et al., 2006). In case of *Eucalyptus* spp., in fact, the probability of a tree being hollow-bearing is strongly associated with its diameter. It is a slow process determined by the rate of decay of internal heartwood, taking more than 150 years (Harper et al., 2005). In 2018, Petrolina presented trees with significantly larger diameters than Juazeiro (Table 3) and this could explain (at least in part) the larger amount of nests of *P.* aff. *flavocincta* found.

Table 3. Values of probability (P) obtained for the comparisons made between cities (considering the three sampled years; Kruskal-Wallis test) and among the years (considering both cities, Mann-Whitney test), for the tree variables that contained *Plebeia* aff. *flavocincta* nests.

Cities (2009, 2015, 2018)	Height at nest entrance	Diameter at nest entrance	Diameter at breast height
Petrolina	P = 0.05	P= 0.14	P = 0.14
Juazeiro	P = 0.40	P= 0.40	P = 0.21
Years (Petrolina x Juazeiro)			
2009	-	-	-
2015	-	-	-
2018	P = 0.15	P = 0.19	**P = 0.003**

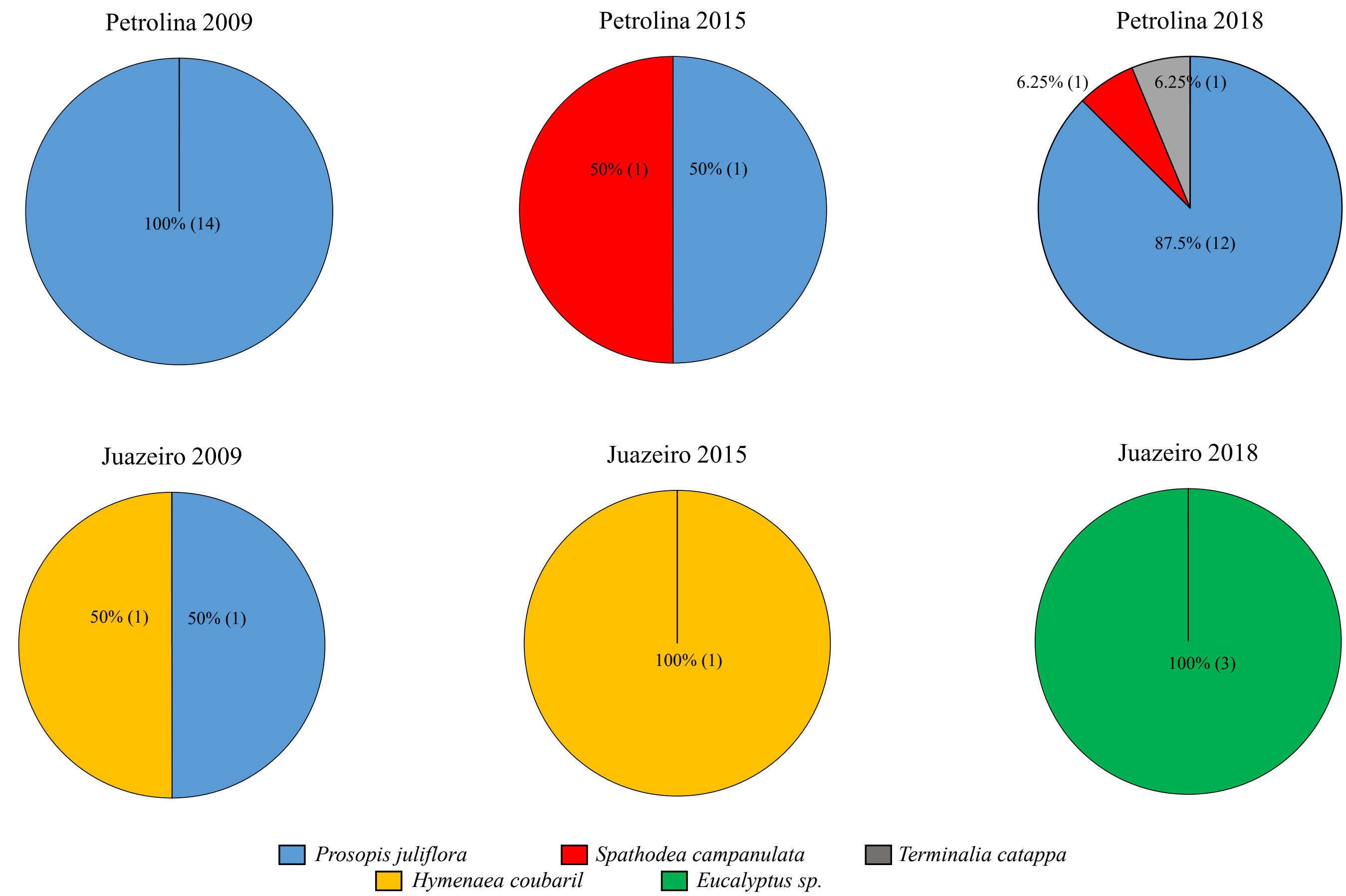

Fig 2. Percentage (%) of nests of *Plebeia* aff. *flavocincta* in trees investigated in Petrolina and Juazeiro, in 2009, 2015 and 2018. (Numbers between parentheses are absolute).

On the other hand, there are exceptions and even trees with very high diameters have no cavities (Fernandes et al., 2009) or are not chosen by the bees. For example, in this study, some trees of *P. juliflora* investigated (at the Batalhão do Exército, in Petrolina) that had no bee' nests where significantly wider in diameter (2.13 ± 0.10 m, n = 44) than the ones (of the same species) that had nests (1.97 ± 0.07 m, n = 25) (P = 0.002, H = 1,423). Thus, a larger *P. juliflora* tree does not necessarily have a larger cavity inside. This suggests that other factors must be important for the formation of the hole inside the tree (such as the kind of soil, nutritional conditions, diseases, etc.) that causes the death of internal tissues, besides its age or growing and the other factors mentioned above.

Different bee species need a minimum diameter in the cavity that allows its nesting. This is probably related to the comb size, thermoregulation, the kind of brood cells arrangement (Cortopassi-Laurino et al., 2009) and, obviously, and bee body size. *P.* aff. *flacvocincta* is quite small (less than 4 mm in length) and, although it has a very populous nest (M. F. Ribeiro, unpublished information) probably needs relatively small cavities to nidify. The diameter at nest entrance may indicate the internal volume. In general, the values for DNE found in this work were smaller than the DBH (Table 3). However, the nest entrance may be far from the place where the nest itself is located inside the hollow tree.

Nest entrance height may be related to the facility to find these nests (Cortopassi-Laurino et al., 2009). These authors found that *Plebeia* sp. nests were found at 2.0-4.0m of height. For *P.* aff. *flavocincta* in this work (in the year of 2018, in Petrolina), we found the highest nest entrance at 4.26m. Indeed, 42.86% of the nests were found around 2.0 m or more of height. Other nests were found at smaller heights: 35.71% around 1.50-1.60 m, and 21.43% around 0.50 m or less. Nests located very high would be protected of predators (specially the humans).

The mortality of bees nests was caused by the cutting of trees (in several cases) and/or other factors not investigated here. One of these factors might be the human habits of depositing garbage (such as plastic bags or cups) at the folds of the tree trunks. When this is deposited on the bees nests entrances may cause their death. Other reasons could be the extreme drought of last years that may have reduced the floral resources for the bees. On the other hand, other nests survived by a long period (nine years: from 2009 to 2018), as the nests found at the wall in Juazeiro.

Anyway, it is remarkable that this stingless bee is able to survive in urban conditions, despite high levels of stress (such as noise, pollution, and human interferences). Cortopassi-Laurino et al. (2009) suggest that the main reasons for the success of *Tetragonisca angustula* Latreille in occupying urban areas are its habit of visiting many plants as food sources and the use of other substrates (as walls, others constructions, etc.) for nesting. In fact, *P*. aff. *flavocinta* uses other substrates as walls and pipes, as other bees do utilize roofs, ceilings, etc. (Zanette et al., 2005).

Moreover, we found that *P*. aff. *flavocinta* is capable of swarm at urban areas. The same behavior of carrying cerumen was also observed for another stingless bee (*Nannotrigona testaceicornis* (Lepeletier)) also found at urban environment (A. B. Dias, unpublished information).

Stingless bees of small body size, as *Plebeia*, are known to fly maximum short distances as 950 m (Araújo et al., 2004). This fact could serve as incentive for urbanization plans that could include trees that provide cavities for bees nesting and plants with flowers as food sources. This would contribute for their conservation and permanent presence at urban environments. As suggested by Banaszak-Cibicka (2013) biodiversity may be manipulated in urban areas by the management of green spaces. Urban gardens may attract a diverse community of bees, which can contribute with their pollination services (Matteson et al., 2008). Simultaneously, including bee friendly spaces within the urban matrix (as botanic and home gardens, patches of remnant habitat, public parks, etc.) can provide bees habitats and promote their conservation (Hernandez et al., 2009). Moreover, the conservation of hollow-bearing trees may ensure the maintenance of ecosystem processes and biodiversity at urban environments, since cavities are important not only for bees but also for other animals (Harper et al., 2005).

Because the Caatinga biome is known having a smaller number of bee species than other biomes in the South America (Zanella, 2000), this makes its native bees quite important for the maintenance of the ecosystems. Thus, any strategy that may contribute for the restoring or conservation of areas of Northeast Brazil is urgent in order to conserve these important pollinators. These strategies could include the maintenance of urban areas as friendly environments to bees.

Acknowledgments

We are grateful to Francimária Rodrigues and Nayanny de Souza Fernandes for helping with data collection in 2009; Francisco Pereira Nonato, for help at field work in 2015 and 2018; Lúcia H. P. Kiill for the identification of plants, anonymous referees that contributed with valuable criticisms, and Embrapa, for financial support (MP1 01.15.02.003.07.00 Conservação de Recursos Genéticos de Insetos Polinizadores).

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