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Practical meliponiculture: use of trap boxes to control Tracuá Carpenter ants (*Camponotus atriceps* Smith, 1858), an important natural enemy

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ABSTRACT. This study aimed to observe the attractiveness efficiency of trap boxes installed in collective shelters of stingless bee colonies (*Melipona flavolineata*, *Melipona fasciculata* and *Scaptotrigona* aff. *depilis*), as an alternative method for non-chemical control of tracuá carpenter ants (*Camponotus atriceps*). The study was conducted at Embrapa Amazônia Oriental, in Belém, Pará, Brazil, from March to August 2015. The results showed that the efficiency of this technique depended on the presence of bee colonies and on the bee species in the collective shelter. Overall, an efficiency of 40.6% was found in the capture of *C. atriceps* individuals, which rose to 75% considering only collective shelters of *M. fasciculata* colonies, and to 87.5% for collective shelters of *M. flavolineata*. Trap boxes installed in collective shelters of *S. depilis* did not attract any *C. atriceps* group or individuals. The use of trap boxes in collective shelters of stingless bee colonies of the genus *Melipona* (*M. flavolineata* and *M. fasciculata*) is an efficient alternative method of non-chemical control of tracuá carpenter ants (*C. atriceps*).

Keywords: stingless bees; amazon region; natural enemy; meliponary.

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Introduction

The species of indigenous stingless bees, or meliponines (Nogueira-Neto, 1997), are widespread in tropical regions and occupy virtually all of Latin America, Africa, Southeast Asia and Northern Australia. However, it is in the Americas that we can find most of the approximately 400 described types (Villas-Bôas, 2012). These bees are of paramount importance in keeping an ecological balance and floristic diversity (Camargo & Pedro, 2003, 2013) and vital to maintain biodiversity (Kevan & Philips, 2001).

Depending on the ecosystem, meliponines are estimated to be responsible for about 40 to 90% of the pollination of native trees, in addition to acting on the productivity of cultivated plants (Drucker, 2004; Slaa, Sánchez Chaves, Malagodi-Braga, & Hofstede, 2006; Westerkamp & Gottsberger, 2006; Bomfim, Bezerra, Nunes, Aragão, & Freitas, 2015; Ramírez, Ayala, & González, 2018) and on the fertility of vegetables that rely on cross-pollination (Kerr et al, 1996). However, their importance is also widely valued when it comes to their rational keeping for the exploitation of their products.

Meliponiculture, name given to the rational keeping of indigenous stingless bees (Silveira, Melo, & Almeida, 2002), is a very old activity (Palazuelos Ballivián, 2008) still in full expansion in tropical and subtropical regions of the world (Contrera, Menezes, & Venturieri, 2011; Imperatriz-Fonseca, Canhos, Alves, & Saraiva, 2012). In the Brazilian Amazon region, the species of stingless bees *'uruçu amarela'* (*M. flavolineata*, Friese), *'uruçu-cinzenta'* (*M. fasciculata*, Smith) and *'canudo'* (*S. depilis*) are among the most used in the activity, having honey as their main exploited product (Albuquerque, Gostinski, Rêgo, & Carreira, 2013; Venturieri, Leão, Rego, & Venturieri, 2018).

Initially developed by the Indians, Brazilian meliponiculture is based on family labor, promoting an increase in income, as well as the pollination and perpetuation of thousands of plants (Magalhães & Venturieri, 2010; Garibaldi et al, 2013). However, this activity is still far from reaching its maximum

productive potential and, annually, many cases of colony losses due to enemy attacks, mainly ants and phorid flies, are recorded (Oliveira, Venturieri, & Contrera, 2013).

Tracuá carpenter ants (*C. atriceps*, Smith) (Hymenoptera: Formicidae) stand out among the ants harmful to meliponiculture. They belong to a very diverse and broad genus, and, because of their powerful jaws, that can easily destroy entire colonies of meliponines, are considered as one of the main natural enemies of stingless bees (Schwarz, 1938; Fernández, 2003). This ant species is responsible for the death of colonies, mainly during the season of food scarcity in the field, when they are weaker and less capable of defending the nest. *C. atriceps*, also attacks the *Apis mellifera* social bees (Marcolino, Oliveira-Junior, & Brandeburgo, 2000), causing serious losses for beekeepers. Although there are rare records of mutualistic interactions between stingless bees and *Camponotus* ants, as observed by Laroca and Almeida (1989), carpenter ants are in general threats to stingless bee colonies. Many stingless beekeepers use the method for protecting the boxes recommended by Nogueira-Neto (1997), which consists of protecting the base where the colonies are placed with cotton tow impregnated with grease or burnt oil. However, *C. atriceps* foragers still manage to gain access to the bee colonies, causing losses for producers. Another possible technique is the use of trap boxes to attract *C. atriceps* individuals, making it easier to control them. There are records of this technique being employed by stingless beekeepers in the state of Pará, Brazil (personal communication), but they have not yet been measured or assessed experimentally.

Thus, this research aimed to observe the attractiveness efficiency of trap boxes installed in collective shelters of stingless bee colonies (*M. flavolineata*, *M. fasciculata* and *S. depilis*), as an alternative method for non-chemical control of tracuá carpenter ants (*C. atriceps*).

Material and methods

Study region and site characterization

The study was conducted at the Iratama Demonstrative Meliponary, linked to the Botany Laboratory of Embrapa Amazônia Oriental (1°26'11.52"S, 48°26'35.50"W) in Belém, Pará, Brazil (Figure 1), from March to August 2015. The climate in the region is of the *Af* type, characterized as a humid or super humid tropical climate (relative humidity ranging from 84% to 86%,) and by medium/high temperatures, with relatively abundant rainfall throughout the year, with a minimum of 60 mm in the driest months, in accordance with the Köppen Geiger classification (Peel, Finlayson, & Mcmahon, 2007).



Figure 1. Spatial image of part of the city of Belém, with highlight to the Iratama Demonstrative Meliponary at Embrapa Amazônia Oriental, and estimate flight range of bee workers (2.5 km radius), indicated by black circle

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The Embrapa area is considered an environmental conservation area due to its occupancy and environmental characteristics, such as: fragments of primary forest, springs, groves and agroecosystems (Watrin, Santos, & Valente, 2011). Secondary vegetation areas can also be found in various succession stages with variable density and structure (Pires & Salomão, 2000). The vegetation cover is of the dense rainforest type, with areas of dry land, lowland and blackwater-flooded forests (Japiassú & Góes Filho, 1974). Thus, the described area provides a wide and diversified source of floral resources to stingless bee colonies raised in the Iratama meliponary, and within the maximum foraging distance of each species (Araújo, Costa, Chaud-Netto, & Fowler, 2004) (Figure 1).

Evaluation of the attractiveness efficiency of the trap boxes

To evaluate the efficiency of a trap box in attracting tracuá carpenter ants (*C. atriceps*), four collective shelters (Figure 2) were monitored at two different moments: three of them occupied with rational hives (INPA type) populated by colonies of stingless bees (one collective shelter for each bee species), and another occupied with empty trap boxes only. The treatments used in the experiments were: Treatment 1 – Collective shelter composed of 33 colonies of *M. flavolineata* and four trap boxes; Treatment 2 – Collective shelter composed of 27 colonies of *M. flavolineata* and four trap boxes; Treatment 3 – Collective shelter composed of 19 colonies of *S. depilis* and four trap boxes; Treatment 4 (Control) – Collective shelter without stingless bee colonies, composed of four trap boxes only. Thus, 16 trap boxes (four for each treatment) were used at each moment.



Figure 2. Collective-shelter model for the stingless bee colonies in the Iratama Demonstrative Meliponary.

The trap box consisted of an empty stingless beehive (INPA type), without upper nest and supers, installed on the shelves of each collective shelter, among the hives inhabited by the stingless bee colonies (Figure 3a and 3b). In each collective shelter, two new (virgin) boxes and two used boxes (which had previously been occupied by a stingless bee colony), also empty, were installed, so that the ants' preference for the type of trap box was known.

Over one-month period, weekly inspections were carried out to indicate the presence or absence of *C. atriceps* in the trap boxes. Once found in the trap boxes, they were kept until the end of the four weeks. The experiment was repeated after an interval of 30 days from the closure of the previous one, totaling 32 trap boxes (16 trap boxes x 2 moments). After each experimental period, the ants present in the traps were exterminated with ethyl acetate. Subsequently, the individuals were separated by caste and counted, which provided an average number for how many individuals from each caste occupied the trap boxes.



Figure 3. (a) Trap box for ants installed in one of the collective experimental shelters of Embrapa Amazônia Oriental; (b) tracuá carpenter ants (*C. atriceps*) inside the trap box, Belém, Pará, Brazil.

Data analysis

For data analysis, we used the Statistica 7 program (StatSoft 2005). Because the data are non-parametric and binomial – with the ants attracted to the trap being equivalent to 1, and those not attracted to 0 (zero), they were compared using the Cochran's Q test, at a 5% level of significance.

Results and discussion

Only the trap boxes installed in the collective shelters that contained the species *M. fasciculata* and *M. flavolineata* attracted the *C. atriceps* ants. The occupancy rate of the trap boxes for these two bee species stood at 87.5 and 75%, respectively, not differing statistically from each other (p > 0.05) (Table 1). However, the occupancy rate of ants in the trap boxes installed in the collective shelters of these two species of the genus *Melipona* differed significantly from that referring to the trap boxes installed in collective shelters of *S. depilis* (0.0%) (p < 0.05) and from those installed in collective shelters without bee colonies (Control treatment) (0.0%) (p < 0.05).

The groups of *C. atriceps* (n=13) inside the trap boxes did not have queens or males, being made up of workers (\bar{x} =381.8 ± 266.5), soldiers (\bar{x} =169.8 ± 132.2) and immatures (\bar{x} =45.6 ± 21.3) (Figure 3b).

Treatments	New trap box*			Used trap box*			n	Total	% Total
	n	Occupied Quantity	% Occupancy	n	Occupied Quantity	% Occupancy	Total	Occupied Quantity	Occupancy
M. flavolineata ('uruçu amarela')	4	3	75.0	4	4	100.0	8	7	87.5a
M. fasciculata ('uruçu-cinzenta')	4	4	100.0	4	2	50.0	8	6	75.0a
S. depilis ('canudo')	4	0	0.0	4	0	0.0	8	0	0.0b
Control treatment	4	0	0.0	4	0	0.0	8	0	0.0b
Total	16	7	43.7	16	6	37.5	32	13	40.6

 Table 1. Occupancy rate of tracuá carpenter ants (C. atriceps) in trap boxes (new and used) inside collective shelters of different species of stingless bees. Embrapa Amazônia Oriental, Belém, Pará, Brazil.

n = number of trap boxes available. Values followed by different letters in the same column differ significantly between them (p < 0.05). * Values in the same line did not differ significantly between them (p > 0.05).

No occupancy was found for the trap boxes (n=8) in the collective shelter without stingless bee colonies (Control treatment), showing that tracuá carpenter ants are attracted by the presence of bees and/or their colony resources. Likewise, when in collective shelter of *S. depilis* bees, the ants are not attracted to the traps (Table 1). In this case, the defensive behavior of *S. depilis*, or even chemical information perceived by the ants, may have discouraged the occupancy of these trap boxes (Lehmberg, Dworschak, & Blüthgen, 2008).

In general, species of bees from the Meliponini tribe that do not belong to the genus *Melipona* have more strategies to defend their nest inside and outside (Couvillon, Wenseleers, Imperatriz-Fonseca, Nogueira-

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Neto, & Ratnieks, 2008; Shackleton et al., 2014). Lehmberg et al. (2008) and Wang, Wittwer, Heard, Goodger, and Elgar (2018) describe, as a form of defense for this group, aggressive bites, use of plant resins and propolis to bond the enemy, or even the action of chemical components released by the material used for preparing the entrance to their nests. And, in the case of *S. depilis*, the very entrance to the nest in the form of a 'straw' makes it even more difficult for ants to attack.

Sometimes, ants colonize an area near a food source (Dreisig, 2000; Feldhaar et al., 2007), but from what we could observe, not only the presence of food, but also the ease of obtaining it is determining for the formation of a satellite nest (secondary nest) of *C. atriceps*. This is a totally opportunistic behavior on the part of the ants, which preferred to occupy the trap boxes of the collective shelters that had bees of the genus *Melipona*.

Many cases of attacks by carpenter ants are observed in colonies of bees of the genus *Melipona* (Nogueira-Neto, 1997; Pereira, Souza, & Lopes, 2010; Contrera & Venturieri, 2010). These bees are virtually helpless against these ants, which, with their sharp jaws, occupy hives, kill workers and the queen, and take the offspring, which will serve as food (Bueno & Campos-Farinha, 1998; Dreisig, 2000). Thus, they are responsible for the extermination of many colonies of stingless bees in the Amazon region, causing significant damages and losses, both for the bees and the keepers.

The period of stay in the collective shelters of *M. flavolineata* and *M. fasciculata* had no influence, for two days after the trap boxes were installed, there already were ants inside. The fact that the trap box had been previously occupied by stingless bee colonies did not increase the efficiency of this method for enemy control (p > 0.05) (Table 1), showing that the odors left by the bee colonies do not influence occupancy or are attractive to ants.

Carpenter ants work to build nests that can house their eggs, known as primary nests. Satellite nests are built when the primary nest is established and starts maturing (Schultner & Pulliainen, 2020). All individuals attracted by the trap boxes included workers, soldiers and immatures. Due to the absence of queens, the captured nests are presumed to be satellite nests, which were attracted by the smell of the food source (bee colonies and their products) near the site. Therefore, this control technique can not only eliminate satellite nests, which, instead of invading the bee colonies, will inhabit the traps, but also help find the primary nest, which can thus be exterminated more easily. Additionally, the use of trap boxes is even more interesting as it does not require chemical compounds that could contaminate the colonies, as well as their products.

This technique studied in the present research, however, should not be understood as the solution to the problems of attacks by tracuá carpenter ants in meliponaries, but rather as another tool that should be used in combination with other techniques that contribute to the defense of bee colonies (e.g., cleaning the area, the use of PET bottles, greases and tow with burnt oil). Moreover, because ants are attracted to the colonies by the smell of food, it is important for stingless beekeepers to handle the hives carefully in order to avoid the exposure of pollen and honey pots and, this way, to avoid attacks by these enemies (Villas Bôas, 2012).

Conclusion

The use of trap boxes in collective shelters of stingless bee colonies of the genus *Melipona* (*M. flavolineata* and *M. fasciculata*) is an efficient alternative method of non-chemical control of tracuá carpenter ants (*C. atriceps*).

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