



RESEARCH ARTICLE - BEES

Adaptive and Foraging Behavior of Two Stingless Bee Species (Apidae: Meliponini) in Greenhouse Mini Watermelon Pollination

IGA BOMFIM¹, AD DE M BEZERRA¹, AC NUNES², FAS de ARAGÃO², BM FREITAS¹

1 - Universidade Federal do Ceará, Fortaleza, CE, Brazil

2 - Embrapa Agroindústria Tropical, Fortaleza, CE, Brazil

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Corresponding author

Isac Gabriel Abrahão Bomfim

Faculdade de Tecnologia Centec (FATEC)

Sertão Central, Eixo Tec. de Recursos

Naturais, Quixeramobim, CE, Brazil

E-Mail: isacbomfim@yahoo.com.br

Abstract

The growth of protected cultivation in world agriculture has driven crops traditionally cultivated in open field to greenhouse, which, in turn, prevent the access of pollinators to flowers. Therefore, it is necessary to identify suitable pollinators for closed environments. Stingless bees have been identified in Brazil as the ideal pollinators for crop pollination under greenhouse conditions. Colonies of two stingless bee species (*Melipona subnitida* Ducke) and *Scaptotrigona* sp. nov. were evaluated in a greenhouse during the flowering of seeded (diploid) and seedless (triploid) mini watermelon (*Citrullus lanatus* T.) varieties. The feasibility of using these bees in this production system was based on their adaptation and foraging behavior. *M. subnitida* did not show any interest to the crop under the experimental conditions; and this species stopped foraging trips and egg laying in a diapause-like stage. In comparison, *Scaptotrigona* sp. nov. foragers were active and collected floral resources soon after their introduction. Moreover, the foragers of *Scaptotrigona* sp. nov. showed an essential behavior for mini watermelon pollination, because they visited staminate and pistillate flowers from both seeded and seedless genotypes for nectar collection. Thus, we concluded that *M. subnitida* did not adapt to greenhouse mini watermelon cultivation under the experimental conditions and should not to be used for pollination purposes in this environment; however, *Scaptotrigona* sp. nov. adapted well as a pollinator in the greenhouse and can be used for mini watermelon pollination in protected culture.

Introduction

The growth of protected cultivation in world agriculture has led to crops traditionally cultivated in open field to be grown in protected environments, but this condition creates a physical barrier which prevents the access of natural pollinators to flowers (Guerra Sanz, 2008). Hand pollinating the flowers in a protected environment, as well as the application of hormones to induce fruit production, is laborious and increases production costs (Cruz & Campos, 2009). Thus, a more economical alternative would be the introduction of native pollinators that are able to adapt to the conditions of closed environments and to meet the pollination requirements of crops under these conditions (Banda & Paxton, 1991; Cruz & Campos, 2009).

The most promising pollinator in Brazil seems to be the stingless bees (subtribe Meliponina), since they occur

naturally throughout Brazil and have a stunted vestigial sting that makes them safe for workers carrying out daily cultural practices in this confined environment. These insects also form perennial colonies that are usually active all over the year, and they can be managed in small hives which can be easily transported to the site requiring pollination services (Heard, 1999; Slaa et al., 2006).

Several stingless bees species have been determined to provide adequate pollination services to many different crops grown under protected cultivation, such as strawberry (*Fragaria x ananassa* D.), eggplant (*Solanum melongena* L.), sweet pepper (*Capsicum annuum* L.), tomato (*Lycopersicon esculentum* Mill.) and cucumber (*Cucumis sativus* L.) (Malagodi-Braga, 2002; Cruz et al., 2005; Del Sarto et al., 2005; Santos et al., 2008; Nunes-Silva et al., 2013; Nicomedo et al., 2013). Since the stingless bees form a very diverse group, c.a. 400 species, with very different physical and behavioral characteristics, it is possible to select



species with more suitable characteristics for certain plant species or type of protected cultivation (Slaa et al., 2006). The use of stingless bees for pollination of cucurbit species inside greenhouse is restricted to cucumber (Santos et al., 2008; Nicomedo et al., 2013). However, the watermelon [*Citrullus lanatus* (Thunb.) Matsum & Nakai] is of great economic interest and its cultivation in protected environments is increasing in Brazil and the world, especially with the use of genotypes that produce small fruits weighting up to 1.5 kg. These small, mini watermelon fruits are higher in price per kilogram than conventional large fruits, both in the domestic and international markets (Bomfim et al., 2013).

This crop has high pollination requirements, because flowers require multiple bee visits to deposit pollen on the stigma in order to set fruits (Stanghelliniet al., 1997; Walters & Schultheis, 2009; Winfree et al., 2007). Regarding seedless watermelon varieties (triploid - 3n), their pollination requirements is even greater (Walters, 2005). The objective of this study was to evaluate the feasibility of using colonies of the stingless bees, *Melipona subnitida* Ducke and *Scaptotrigona* sp. nov., to pollinate seeded and seedless mini watermelon varieties under protected culture.

Material and Methods

Site and agricultural practices

The experiment was conducted from August to October 2011 in a 160 m² arched-roof greenhouse (8 m wide x 20 m long x 3.5 m high), fitted with automated drip fertigation and temperature control systems situated at Embrapa Tropical Agroindustry, Fortaleza, Ceará, Brazil. The greenhouse was covered with a high density polyethylene film treated with anti-UV additives (material that filters ultraviolet radiation), having white colored roof and transparent light diffuser color on the sidewalls. It also had a 50% Aluminet® type screen installed 2.5 m above the ground, which function was to lower the temperature inside the greenhouse. Throughout the experiment, the plants were drip-fertigated with water and nutrients suitable for each stage of plant growth.

Seeds of two seeded mini watermelon varieties (2n) (Minipol and Polimore) and three seedless mini watermelon varieties (3n) (HA-5106, HA-5158 and HA-5161) were sown in plastic trays filled with a commercial substrate composed of dried coconut powder. Twelve days later, 408 seedlings were transplanted to 5 liter-plastic pots, previously filled with raw coconut fiber and powdered coconut fiber (1:1). The pots were spaced 0.8 m between rows and 0.4 m between plants. A 3:1 ratio between triploid and diploid varieties was used in dedicated rows (Fiacchino & Walters, 2003; Dittmar et al., 2009) for seedless watermelon cultivation. Plants were trellised to facilitate management of this crop in a protected environment greenhouse. As suggested by Mohr (1986) and Campagnol et al. (2012), only one fruit was allowed to develop on each plant, so that later developing

fruits would not influence the development of the first fruit. Thus, additional fruits that set were removed during their early days of development.

Preparation of stingless bee colonies

Before being introduced in the greenhouse, four colonies of *M. subnitida* and two of *Scaptotrigona* sp. nov. (Oliveira & Jesus, personal information) were selected in the meliponary of the Federal University of Ceará (UFC), and then, colonies of each species were standardized in respect to the quantity of brood and food stores. These colonies were introduced in the greenhouse as soon as the plants emitted their first staminate flowers.

Adaptive behavior to the protected environment

Bee flight activity, defined as foraging bees leaving their hives during the time that flowers were open (anthesis), was monitored throughout the experiment. Foraging bees were enumerated by using a manual tally counter to determine the number of individuals that flew out their hives within a 10-minute period of each hour. Therefore, it was possible to calculate the mean daily number of foragers leaving their hives within a 10-minute period of each hour. The activity performed by bee foragers after leaving their colonies and the amount of days that each species begin foraging on mini watermelon flowers after their introduction in the experimental greenhouse were also determined.

Bee colony development was monitored during the period in which they were used for pollination services. Parameters such as changes in brood area, food store and queen oviposition (considering it active or not) were collected. Additionally, data on luminosity (klux) were measured using a digital lux meter and data on temperature (°C) and relative humidity (%) were measured hourly by a data logger placed inside the greenhouse.

Foraging behavior in mini watermelon flowers

The general foraging behavior of individuals belonging to both bee species were observed to determine the kind of resources collected from mini watermelon flowers, and if they touched the anthers (in staminate flowers) or the stigma (in the case of pistillate flowers) during visitation. It was also determined how many ways these bees approached the flowers and what part of their body contacted the anthers or stigma, depending on the flower approach. Between 07:00 and 10:00 a.m. during two non-consecutive days, we recorded the frequency of bee visitation to flowers, measured by the number of visits received by a flower for five minutes. Finally, we used a chronometer to record the mean time spent by bees per flower during a single visit.

Data analysis

Data regarding the flux of foraging bees leaving their hives were submitted to regression analysis with analysis of variance of the curve by F-test using the program Table Curve 2D version 5.01. Other data were analyzed by descriptive statistics, such as means with standard deviations (SD) or standard errors.

Results and Discussion

Adaptive behavior to the protected environment

During the first day of adaptation to greenhouse, both bee species showed a similar behavior. Many foragers flew out their hives at first sunlight, flew towards the sunlight and collided against the plastic covering of the greenhouse. As the sun moved in the sky during the day, foragers collided against different parts of the greenhouse structure, as they followed the sun as a reference for navigation. Most of these foragers became disoriented, unable to return to their hives, and continued to collide against the plastic structure until their death by exhaustion. According to several researches (Free, 1993; Malagodi-Braga, 2002; Cruz et al., 2004), *Apis mellifera* L. and stingless bee foragers usually become disorientated, attempt to escape and eventually collide against the greenhouse covering in the early days after being introduced into this enclosed environment. In comparison, species belonging to the genus *Bombus* do not become disorientated and begin pollination services as soon as they are introduced into a protected greenhouse environment, rarely colliding against the greenhouse covering (Fisher & Pomeroy, 1989; Guerra Sanz, 2008).

After the death of several foragers and a few days of adaptation, there was a large decrease in the numbers that tried to escape, which resulted in less collision against greenhouse structures and disorientation behaviors. As soon as a colony is introduced in the greenhouse, the oldest and most experienced bees of the colony are those that are the first to go out foraging and fly high in the sky determining their usual flight paths. However, the artificial environment of a greenhouse has a limited size, which imposes physical barrier to the flight of foragers. Therefore, it is understandable that bees initially collide against the transparent covering of the greenhouse in an attempt to continue the foraging activity they did before being introduced into this enclosed environment (Free, 1993; Slaa, 2003).

After initial attempts to escape, *M. subnitida* foragers showed little movement at the hive entrances throughout the entire period they were in the greenhouse (Fig 1A and C). Furthermore, their activities were primarily restricted to brief flights of short distances to remove litter from inside their hives (pieces of dead larvae, pupae and adult bees). Since foragers of this species did not visit the crop flowers, colony inspections showed a progressive reduction in food stores, brood area and queen laying.

After 14 days inside the greenhouse, the colonies of *M. subnitida* had to be removed, because their populations

were considerably reduced and colony internal development conditions were greatly reduced, resulting from the lack of food storage, high brood mortality, and disruption of egg laying by the queen (Fig 2A, B, C and D). Furthermore, some colonies had already completely sealed the entrance to the nest with resin to prevent them from being attacked by other colonies that were less affected.

Although stingless bees will generally adapt well in protected environments, some species will not adapt to the conditions imposed by certain greenhouse structures or to the crop being grown. Malagodi-Braga (2002) reported that the foragers of *Schwarziana quadripunctata* (Lepeletier) and *Scaptotrigona bipunctata* (Lepeletier) stingless bees did not visit the flowers of the strawberry grown under protected environment due to the lack of interest in the strawberry floral resources or they were not able to adapt to the conditions imposed by protected greenhouse structure.

Unlike the present study, Cruz et al. (2005) were successful in using colonies of *M. subnitida* for pollination in protected cultivation. However, the crop was sweet pepper, and the greenhouse had different features, such as a glass roof and insect-proof screens covering the lateral sides and did not have an automated temperature control system. This non-adaptation of *M. subnitida* may have been caused by the coating material treated with anti-UV additives, and may not be related to the attractiveness of the mini watermelon crop for these bees.

According to Guerra Sanz (2008) and Bartelli et al. (2014), a preventive measure against insect pests in greenhouses is using coverings that have the ability to block or reduce the entry of ultraviolet (UV) light. These materials are able to transmit the band of light that the plant needs for photosynthesis, but diminish or block the entrance of UV light, which is the most important part of color spectrum visible to insects. Although this type of plastic covering reduces the use of pesticides, which is desirable to consumers and also to the bees introduced for pollination purposes, it can also alter the activity of pollinators and may even prevent them from accomplishing what they were intended to do inside the greenhouse.

Another possible reason of the non-adaptation of *M. subnitida* colonies to the experimental greenhouse was the interaction between temperature (maximum average of 33.4 °C) and high humidity (maximum average of 97.4%) imposed by the experimental environment (Fig 3). These of environmental conditions are not found in the Caatinga (xeric shrub land and thorn forest), the region where the colonies used in this experiment were native.

Although the foragers of *Scaptotrigona* sp. nov. during the first day in the protected environment showed a similar behavior to *M. subnitida* foragers, including high mortality by incessant collisions against the covering structures of the greenhouse, they began foraging in mini watermelon flowers in the second day after their introduction. This initial contact occurred about 10:00 a.m. of the second day, when a few foragers returned to their hives after visiting some flowers. Then,

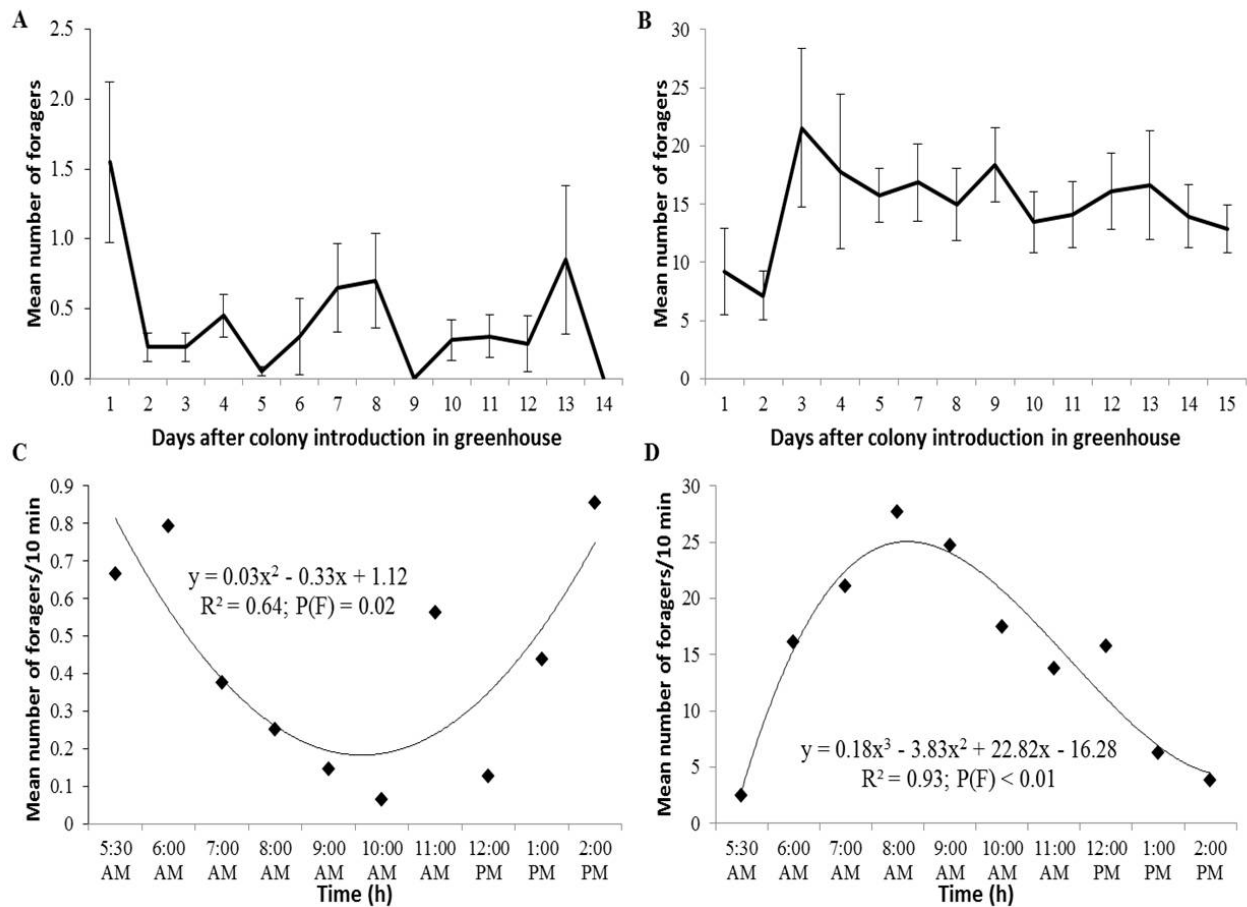


Fig 1. Outgoing flux of foragers during the anthesis of mini watermelon (*Citrullus lanatus*) flowers under a greenhouse. A - Mean daily number with standard errors of *Melipona subnitida* workers leaving their hives within a 10-minute period of each hour. B - Mean daily number with standard errors of *Scaptotrigona* sp. nov. workers leaving their hives within a 10-minute period of each hour. C - Mean hourly number of *Melipona subnitida* workers leaving their hives within a 10-minute period of each hour. D - Mean hourly number of *Scaptotrigona* sp. nov. workers leaving their hives within a 10-minute period of each hour.

suddenly, there was a great increase in the flight activity, with individuals of *Scaptotrigona* sp. nov. flying directly to the flowers and gradually expanding their range within the greenhouse. From the third day until the end of the experiment, these bees visited mini watermelon flowers throughout their anthesis all over the entire area of the greenhouse.

Older bees usually have the task of collecting floral resources, and already have certain habits and experiences established with the previous environment, therefore, they have a greater difficulty in adapting to the protected greenhouse environment, which also limits their flight range (Free, 1993). This explains the high mortality of foragers of both species on the first day after introduction. On the other hand, according to Free (1993), Cauch et al. (2004) and Cruz et al. (2004), the sudden change in behavior on the second day can be explained by the young foraging bees, who did not perform foraging activities before the introduction of the colonies in the protected environment. For that reason, they had not yet established their flight paths, and consequently, did not have much trouble in adapting to the new conditions imposed by protected cultivation. After adapting to the greenhouse, which was when bees start of foraging on flowers, only a very small fraction of foragers continued to

collide against the greenhouse covering and focused on removing debris from their colonies until the experiment terminated. From the third day after introduction of colonies into the protected environment, the vast majority of *Scaptotrigona* sp. nov. foragers that left their hives flew directly to the flowers in search of food resources, and continued that behavior until the end of experiment (Fig 1B).

The period of adaptation for stingless bees to a greenhouse conditions can vary greatly between species as well as between colonies of the same species. Some species, such as, *Nannotrigona perilampoides* (Cresson) require anywhere from five days to eight weeks to be consistent in their visitation to tomato flowers grown in greenhouse (Macias et al, 2001; Cauch et al., 2004). Cruz et al. (2004) reported that *M. subnitida* foragers started collecting floral resources and providing pollination services on sweet pepper cultivation seven days after being introduced into greenhouse. Malagodi-Braga (2002) indicated other species of stingless bees that were consistent in their adaptation to pollinate strawberry grown in a greenhouse. For *Tetragonisca angustula* (Lepeletier) colonies, the adaptation period varied from one day to three weeks, while the adaptation period for *Nannotrigona testaceicornis* (Lepeletier) was about

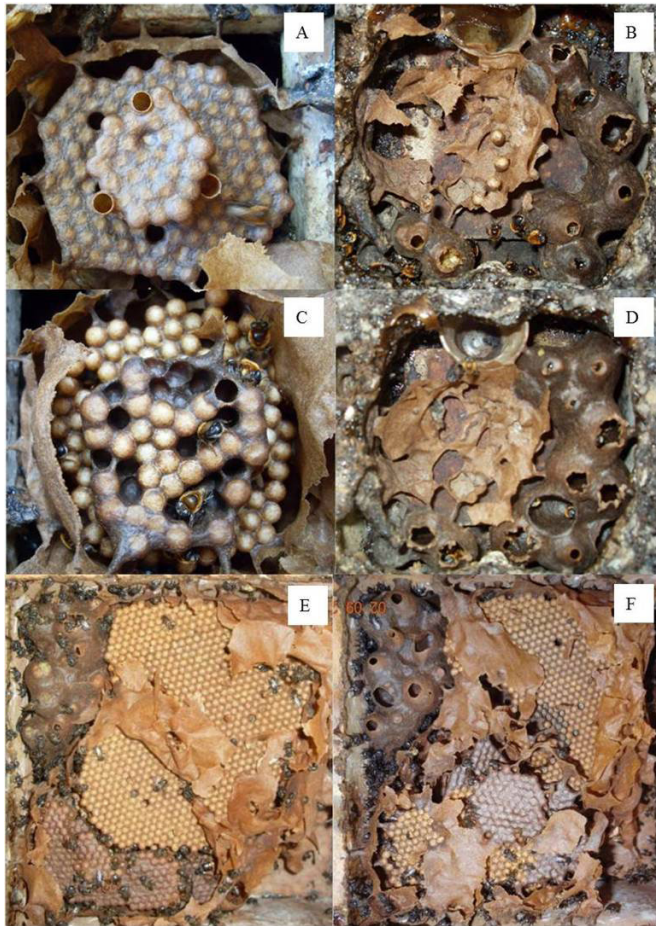


Fig 2. Development stages of stingless bees colonies used for pollination of mini watermelon (*Citrullus lanatus*) under a greenhouse. A - Brood combs in a *Melipona subnitida* colony at the beginning of the pollination services. B - Food stores in a *Melipona subnitida* colony at the beginning of the pollination services. C - Brood combs in a *Melipona subnitida* colony at the end of the pollination services. D - Food stores in a *Melipona subnitida* colony at the end of the pollination services. E - Brood combs and food stores in a *Scaptotrigona* sp. nov. colony at the beginning of the pollination services. F - Brood combs and food stores in a *Scaptotrigona* sp. nov. colony at the end of the pollination services.

three days. Also in the same experiment, a wild nest of *Trigona spinipes* (Fabricius) was found to take only four hours to have its foragers consistently visiting the strawberry flowers.

In this study, the foragers of *Scaptotrigona* sp. nov. took only c.a. 30 hours to start foraging in mini watermelon flowers. Moreover, the internal colony conditions and bee population levels were not considerably affected throughout the period of confinement under protected culture conditions (Fig 2E and F), although they were faced with adverse environmental conditions, such as high temperatures, high humidity and lack of ultraviolet light (UV). Also, after returning to the meliponary these colonies recovered to levels similar to those prior to their introduction in the greenhouse in less than 30 days. These results suggest that *Scaptotrigona* sp. nov. has potential for commercial uses as a pollinator in greenhouse because it quickly adapts to the protected environment, initiates flower visitation in a few hours after introduction, internal colony development

and bee population are not affected by the confinement, and populations recover fast after leaving the greenhouse.

Foraging behavior in mini watermelon flowers

The foraging activity of *Scaptotrigona* sp. nov. foragers usually began at 5:30 a.m. (Temperature = 24.5°C; Humidity = 97% and Luminosity = 1.79 klux; Fig 3), which was shortly after the first sunlight appeared in the greenhouse and the beginning of the anthesis of mini watermelon flowers. Although only 2.46 ± 3.49 bees flew out of their hives at this time, numbers increased rapidly with a maximum peak occurring at 08:00 a.m. (27.71 ± 15.36 bees; Fig 1D). After this time, the number of foragers flying out of their nests decreased slowly until they almost ceased by 2:00 p.m., when there was a mean of 3.82 ± 5.10 foragers (Fig 1D). It appears that these bees remained active outside their nests only while the flowers were open, since flowers were almost completely closed by 2:00 p.m. Also, the foragers of *Scaptotrigona* sp. nov. had their maximum visitation peak within the optimal period of stigma receptivity and pollen viability for many cucurbit crops, which, according to Free (1993), Nepi and Pacini (1993) and Kwon et al. (2005), take place up to five to six hours after flower opening.

Unlike *A. mellifera* colonies that direct a certain amount of foragers to collect pollen from watermelon flowers early in the morning (Azo'oela et al., 2010), *Scaptotrigona* sp. nov. foragers did not appear to be collecting this resource.

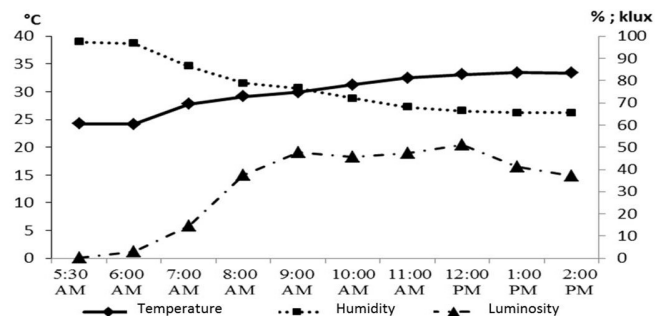


Fig 3. Temperature (°C), relative air humidity (%) and luminosity (klux) inside the greenhouse where the mini watermelon (*Citrullus lanatus*) was cultivated and pollination services by stingless bees were required.

These bees primarily seek nectar in both pistillate and staminate flowers, although during their visits to staminate flowers, they had their bodies dusted with a great amount of pollen grains (Fig 4A). A portion of these pollen grains was transferred to the pollen basket and later taken to the hives (Fig 4B), but a certain amount of pollen grains remained on their bodies, mainly in the head, legs, and thorax and abdomen area.

When approaching the flower, foragers used the petals of both floral types (staminate or pistillate) as a landing platform and then proceeded to walk in the direction of the nectary at the bottom of corolla; unintentionally, the reproductive parts of the flower come in contact with the head and dorsal portion of the thorax (Fig 4C). Another way the flowers were approached by bees was to directly land on the reproductive

parts of flowers (anthers and stigma in staminate pistillate flowers, respectively), where they proceeded towards the nectary (Fig 4D and E). In this type of approach, bees usually contacted the reproductive parts of the flowers with practically the entire ventral portion of their body (thorax, abdomen and legs).

Regardless of the way that bees approached flowers, pollen was transferred to the body of the bees. However, a larger portion of the body of these individuals came in contact with the reproductive parts of the flowers as they approached the flowers and landed directly in their reproductive parts. Some foragers were also observed performing both behaviors in the same flower during the same visit. In addition, some foragers of *Scaptotrigona* sp. nov. walked on the petals and on the stigmatic surface of pistillate flowers (Fig 4F) in order to reach the other side of the nectary, thus touching and transferring pollen grains in a single visit to more than one of the stigmatic lobes. According to Mann (1943) and Delaplane and Mayer (2000), watermelon flowers require that pollen grains be distributed to all the three lobes of the stigma in order to develop fully formed fruits.

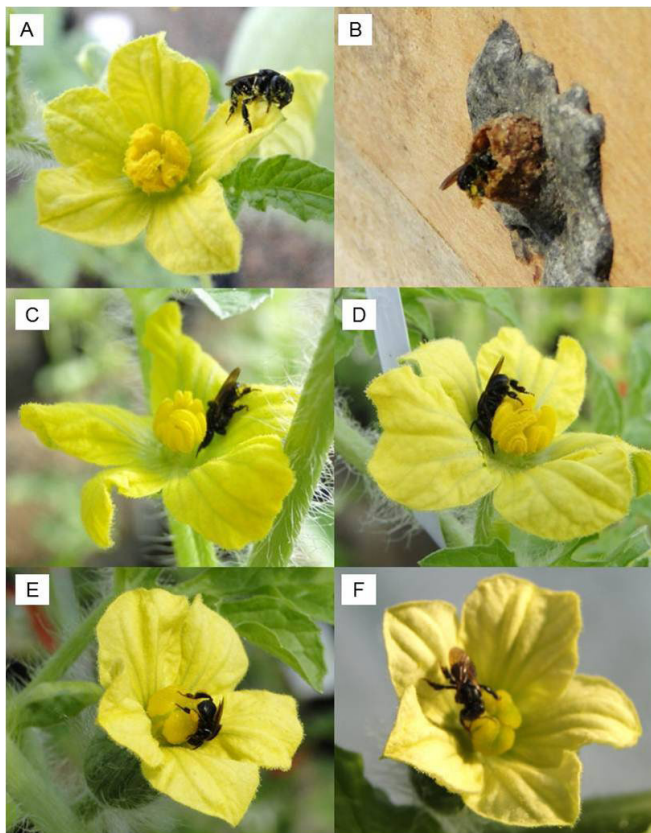


Fig 4. Foraging behavior of *Scaptotrigona* sp. nov. workers in mini watermelon (*Citrullus lanatus*) flowers grown in a greenhouse. A - Worker covered with pollen grains and stopping on a petal of a staminate flower while brushing herself before taking off. B - Forager returning to the nest with pollen on her cobiculae. C - Worker collecting nectar while on a staminate flower and touching the anthers with her head and dorsal part of the thorax. D - Forager collects nectar while sitting on the anthers of a staminate flower, acquiring pollen on her ventral parts of the thorax and abdomen. E - Worker with pollen on her corbiculae, while moving over the stigma of a pistillate flower in a search for nectar. F - Forager moving from the petal to the stigma of a pistillate flower.

The length of each time for nectar collection visit by a *Scaptotrigona* sp. nov. forager varied between 2.27 and 43.95 seconds, with an average of 13.10 ± 8.86 s ($n = 68$). *Scaptotrigona* sp. nov. foragers remained in flowers much longer than that reported by Njoroge et al. (2004) and Azo'oEla et al. (2010) and Araújo et al. (2014) for *A. mellifera* foragers visiting watermelon flowers in search of nectar, with means varying from 0.56 to 8.7 seconds. Therefore, in addition to the forager behavior of often landing and walking on the reproductive parts of the flower, the long time that the *Scaptotrigona* sp. nov. foragers spend visiting a flower seems to maximize the distribution of pollen grains on the three lobes of the stigma, which is essential for the development of fruit without deformation (Mann, 1943). In contrast to this behavior, the foragers of *A. mellifera* rarely change their position after landing on the watermelon flower, typically restricting their contact to only one of the lobes of the stigma per visit (Adlerz, 1966). Therefore, the distribution of pollen grains by *A. mellifera* in the watermelon stigma is more dependent on multiple visits rather than the time each forager spends on a flower, or even the movement performed by a forager in the flower after landing. Besides moving around in the flower, *Scaptotrigona* sp. nov. foragers made multiple visits to the mini watermelon flower. Each flower received an average of 6.0 ± 3.69 visits ($n = 24$) over a period of five minutes.

After leaving a flower, *Scaptotrigona* sp. nov. foragers, tended to move to the nearest flower, which were generally in the same plant or in the same row. However, foragers also moved between rows as well as only visiting one flower per plant before moving to another. Probably, these variations of movements favored the transfer of pollen between diploid plants and, more importantly, from diploid to triploid plants. Such behavior is essential for producing seedless watermelon fruits, because flowers from seedless varieties ($3n$) require pollen grains of seed varieties ($2n$) for setting fruits (Walters, 2005).

Although indirect measurements are less informative than direct measurements (Gross, 2005), they are an alternative way to measure pollinator effectiveness. The more types of information collected, the greater the likelihood that a visitor can be accredited as a pollinator.

Therefore, these results suggest that the introduction of colonies of *Scaptotrigona* sp. nov. could be an alternative to pollinate seeded and seedless mini watermelon grown inside greenhouses, since these bees adapted quickly to confinement, maintained high population levels, and exhibited foraging behaviors compatible to provide pollination services required by this crop. However, in contrast, *M. subnitida* species did not adapt to the greenhouse mini watermelon cultivation under the experimental greenhouse conditions and should not be used for pollination purposes in such situation. Further studies are needed to determine exactly what are the factors that affect the adaptation and visitation of *M. subnitida* species to flowers of crops grown inside greenhouses.

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References

- Adlerz, W.C. (1966). Honey bee visit numbers and watermelon pollination. *J. Econ. Entomol.*, 59: 28-30.
- Araújo, D., Siqueira, K., Duarte & P. Silva, N. (2014). Comportamento de forrageamento de *Apis mellifera* na melanciaira (*Citrullus lanatus*) no município de Juazeiro, BA. *Rvads*, 9: 59-67.
- Azo'o Ela, M., Messi, J., Fohouo, F.N.T, Tamesse, J.L., Kekeunou, S. & Pando, J.B. (2010). Foraging behaviour of *Apis mellifera adansonii* and its impact on pollination, fruit and seed yields of *Citrullus lanatus* at Nkolbisson (Yaoundé, Cameroon). *Cam. J. Exp. Biol.*, 6: 41-48.
- Banda, H.J. & Paxton, R.J (1991). Pollination of greenhouse tomatoes by bees. *Acta Hort.*, 288: 194-198.
- Bartelli, B.F., Santos, A.O.R. & Nogueira-Ferreira, F.H. (2014). Colony performance of *Melipona quadrifasciata* (Hymenoptera, Meliponina) in a greenhouse of *Lycopersicon esculentum* (Solanaceae). *Sociobiology*, 61: 60-67. DOI: 10.13102/sociobiology.v61i1.60-67.
- Bomfim, I.G.A., Cruz, D.O., Freitas, B.M. & Aragão, F.A.S. (2013). Polinização em melancia com e sem semente. *Embrapa Agroindústria Tropical, Documentos*, 168: 1-54.
- Campagnol, R., Mello, S.C. & Barbosa, J.C. (2012). Vertical growth of mini watermelon according to the training height and plant density. *Hort. Bras.*, 30: 726-732. DOI: 10.1590/S0102-05362012000400027.
- Cauich, O., Quezada-Euán, J.J.G., Macias-Macias, J.O., Reyes-Oregel, V., Medina-Peralta, S. & Parra-Tabla, V. (2004). Behavior and pollination efficiency of *Nannotrigona perilampoides* (Hymenoptera: Meliponini) on greenhouse tomatoes (*Lycopersicon esculentum*) in Subtropical México. *Hortic. Entomol.*, 97: 475 - 481.
- Cruz, D.O. & Campos, L.A.O. (2009). Polinização por abelhas em cultivos protegidos. *Rev. Bras. Agroci.*, 15:5-10.
- Cruz, D.O., Freitas, B.M., Silva, L.A., Silva, E.M.S. & Bomfim, I.G.A. (2004). Adaptação e comportamento de pastejo da abelha jandaíra (*Melipona subnitida* Ducke) em ambiente protegido. *Acta Sci. Anim. Sci.*, 26: 293-298.
- Cruz, D.O., Freitas, B.M., Silva, L.A., Silva, E.M.S. & Bomfim, I.G.A. (2005). Pollination efficiency of the stingless bee *Melipona subnitida* on greenhouse sweet pepper. *Pesq. Agropec. Bras.*, 40: 1197-1201. DOI: 10.1590/S0100-204X2005001200006.
- Delaplane, K.S. & Mayer, D.F. (2000). *Crop pollination by bees*. Cambridge: CABI, 344 p.
- Del Sarto, M.C.L., Peruquetti, R.C. & Campos, L.A.O. (2005). Evaluation of the Neotropical stingless bee *Melipona quadrifasciata* (Hymenoptera: Apidae) as pollinator of greenhouse tomatoes. *J. Econ. Entomol.*, 98: 260-266. DOI: 10.1603/0022-0493-98.2.260.
- Dittmar, P.J., Monks, D.W. & Schultheis, J.R. (2009). Maximum potential vegetative and floral production and fruit characteristics of watermelon pollenizers. *Hort Science*, 44: 59-63.
- Fiacchino, D.C. & Walters, S.A. (2003). Influence of diploid pollenizer frequencies on triploid watermelon quality and yields. *HortTechnology*, 13 (1) 58-61.
- Fisher, R.M. & Pomeroy, N. (1989). Pollination of greenhouse muskmelons by bumble bees (Hymenoptera: Apidae). *Entomol. Soc. Am.*, 82 (4): 1061-1066.
- Free, J.B. (1993). *Insect pollination of crops*. 2^a ed. London: Academic Press, 684 p.
- Gross, C.L. (2005). Pollination efficiency and pollinator effectiveness. In A., Dafni, P.G.; Kevan & B.C, Husband (Eds.), *Practical pollination biology* (pp. 354–363). Cambridge: Enviroquest .
- Guerra Sanz, J.M. (2008). Crop pollination in greenhouses. In R.R James & T.L. Pitts-Singer (Eds.), *Bee pollination in agricultural ecosystems* (pp. 27-47). New York: Oxford University Press, Inc.
- Heard, R.A. (1999). The role of stingless bees in crop pollination. *Annu. Rev. Entomol.*, 44: 183-206. DOI: 10.1146/annurev.ento.44.1.183.
- Kwon, S.W., Jaskani, M.J., Ko, B.R. & Cho, J.L. (2005). Collection, germination and storage of watermelon (*Citrullus lanatus* Thunb.) pollen for pollination under temperature conditions. *Asian J. Plant. Sci.*, 4: 44-49. DOI: 10.3923/ajps.2005.44.49.
- Macias, M.J.O., Quezada-Euan, J.J.G & Parra-Tabla, V. (2001). Comportamiento y eficiencia de polinización de las abejas sin aguijón (*Nannotrigona perilampoides*) en el cultivo del tomate (*Lycopersicon esculentum* M.) bajo condiciones de invernadero em Yucatán, Mexico. In J.J.G. Quezada-Euán, L. Medina-Medina & H. Moo-Valle (Eds.), *II Seminario Mexicano Sobre Abejas sin aguijón, Memorias* (pp. 119-124). Mérida: Universidad Autónoma de Yucatán – Facultad de Medicina Veterinaria y Zootecnia.
- Malagodi-Braga, K.S. (2002). Estudo de agentes polinizadores em cultura de morango (*Fragaria x ananassa* Duch. – Rosaceae). Tese (Doutorado em Ecologia), Universidade de São Paulo, São Paulo, 102 p.
- Mann, L.K. (1943). Fruit shape of watermelons as affected by placement of pollen on the stigma. *Bot. Gaz.*, 105: 257-262.

- Mohr H.C. (1986). Watermelon breeding. In M.J. Bassett (Ed.), *Breeding Vegetable Crops* (pp. 37-66). Connecticut: AVI Publishing Co.
- Nepi, M. & Pacini, E. (1993). Pollination, pollen viability and pistil receptivity in *Cucurbita pepo*. *Ann. Bot.*, 72: 527-536.
- Nicomedo, D., Malheiros, E.B., De Jong, D. & Nogueira-Couto, R.H. (2013). Enhanced production of parthenocarpic cucumbers pollinated with stingless bees and africanized honey bees in greenhouses. *Semin. Cienc. Agrar.*, 34: 3625-3634. DOI:10.5433/1679-0359.2013v34n6Supl1p3625.
- Njoroge, G.N., Gemmill, B., Bussmann, R., Newton L.E. & Ngumi, V.W. (2004). Pollination ecology of *Citrullus lanatus* at Yatta, Kenya. *Int. J. Trop. Insect Sci.*, 24: 73-77. DOI:10.1079/IJT20042.
- Nunes-Silva, P., Hrcir, M., Silva, C.I., Roldão, Y.S. & Imperatriz-Fonseca, V.L. (2013). Stingless bees, *Melipona fasciculata*, as efficient pollinators of eggplant (*Solanum melongena*) in greenhouses. *Apidologie*, 44: 537-546. DOI: 10.1007/s13592-013-0204-y.
- Santos, S.A.B, Roselino, A.C. & Bego, L.R. (2008). Pollination of cucumber, *Cucumis sativus* L. (Cucurbitales: Cucurbitaceae), by the stingless bees *Scaptotrigona* aff. *depilis* Moure and *Nannotrigona testaceicornis* Lepeletier (Hymenoptera: Meliponini) in greenhouses. *Neotrop. Entomol.*, 37: 506-512. DOI: 10.1590/S1519-566X2008000500002.
- Slaa, E.J. (2003). Foraging ecology of stingless bees: from individual behaviour to community ecology. Ph.D. thesis, Department of Behavioural Biology, Utrecht University, Utrecht, 181p.
- Slaa, E.J., Sánchez Chaves, L.A.S., Malagodi-Braga, K.S. & Hofstede, F.E. (2006). Stingless bees in applied pollination: practice and perspectives. *Apidologie*, 37: 293-315. DOI: 10.1051/apido:2006022.
- Stanghellini, M.S., Ambrose, J.T. & Schultheis, J.R. (1997). The effects of honey bee and bumble bee pollination on fruit set and abortion of cucumber and watermelon. *Am. Bee J.*, 137: 386-391.
- Walters, S.A. (2005). Honeybee pollination requirements for triploid watermelon. *HortScience*, 40: 1268-1270.
- Walters, S.A. & Schultheis, J.R. (2009). Directionality of pollinator movements in watermelon plantings. *HortScience*, 44: 49-52.
- Winfree, R., Williams, N. M., Dushoff, J. & Kremen, C. (2007). Native bees provide insurance against ongoing honey bee losses. *Ecol. Lett.*, 10: 1105-1113. DOI: 10.1111/j.1461-0248.2007.01110.x.

