

# Behavior and nest architecture of the bee *Caenohalictus alexandrei* (Hymenoptera, Halictinae)

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**Abstract.** In Colombia, few studies have focused on nest architecture, behavior or sociality of wild bees. This study provides basic information on the nests of *Caenohalictus alexandrei* and presents behavioral observations outside the nests, derived from direct field observation of 40 nests and 39 male sleeping cavities in two localities of the Savanna of Bogota, Colombia. We recognized four different behavioral activities carried out by adult females: foraging, guarding, opening and closing of the entrance of the nest, with foraging being the most frequent of them. The observed activities were carried out more frequently from 9:00 h to 13:00 h. Males were observed performing two behavioral activities in the sleeping cavities: staying at the cavity entrance and cavity departure. A third male activity, corporal grooming, was carried out outside the sleeping cavity. Our field observations indicate that more than one adult female was active simultaneously performing the different tasks within a single nest. The nests presented different levels of complexity in their architecture, depending on the age of the nest and number of adult females in them. Descriptions of the nesting sites and behavioral activities as well as illustrations of the nest architecture are provided.

**Keywords.** *Caenohalictina*; Nesting sites; High Andean species; Halictini.

## INTRODUCTION

*Caenohalictus* Cameron is a genus that comprises slender, bright green bees, rarely brassy, red, or nonmetallic black (in the latter case, the metasoma is usually blackish or partly amber), with long hairs on the eyes and minutely and closely granular punctation on the head and mesosoma (Michener, 2007; Dalmazzo *et al.*, 2014). The genus is widely distributed in the Neotropical region, being especially diverse in the Andes of South America, with a few species inhabiting Central America though it is absent in the Amazon biome (González *et al.*, 2005; Michener, 2007). In high-montane areas, *Caenohalictus* species are very common, diverse and frequently collected. The genus belongs to the Halictinae sweat bees, a subfamily that includes from solitary to eusocial species, and, within it, to the tribe Halictini, a monophyletic group with four subtribes: Thrinchostomina, *Caenohalictina*, Sphecodina and Halictina (Pesenko, 1999, 2004; Danforth *et al.*, 2004, 2008; Gonçalves & Melo, 2010; Michener, 2007; Moure *et al.*, 2007). *Caenohalictina* is the

only subtribe represented exclusively in the New World especially in the Neotropical region. Phylogenetic analyses, either molecular (Danforth *et al.*, 2004) or morphological (Gonçalves & Melo, 2010), support the monophyly of this subtribe. *Caenohalictus* is, among the *Caenohalictina*, the genus with the largest number of known species (56 spp.), more than *Agapostemon* (36 spp.) and *Pseudagapostemon* (26 spp.) (Roberts, 1972; Moure & Hurd, 1987; Cure, 1989; Moure, 2012). The remaining genera of the subtribe each have fewer species.

Many Halictinae species build their solitary, communal or social nests in the ground, while others species nesting in decaying wood (Michener, 1974, 2007). Such social behavior can be influenced by several variables (*e.g.*, parasitism, age, size) and environmental conditions, such as season of the year, food availability or even altitude and latitude (Field *et al.*, 2010; Purcell, 2011).

Plasticity in the social behavior and the diversity of nesting habitats in Halictinae are interesting biological characteristics that have made this subfamily an important model for the development

Pap. Avulsos Zool., 2023; v.63: e202363002

<https://doi.org/10.11606/1807-0205/2023.63.002>

<https://www.revistas.usp.br/paz>

<https://www.scielo.br/paz>

Edited by: Kelli dos Santos Ramos

Received: 25/04/2022

Accepted: 08/11/2022

Published: 23/01/2023

ISSN On-Line: [1807-0205](https://doi.org/1807-0205)

ISSN Printed: [0031-1049](https://doi.org/0031-1049)

ISNI: [0000-0004-0384-1825](https://orcid.org/0000-0004-0384-1825)



and evaluation of theories about the origin and evolution of social behavior in bees (Eickwort, 1969; Eickwort et al., 1996; Michener, 1974, 2007; Silveira et al., 2002; Danforth et al., 2008; Gonçalves, 2019).

*Caenohalictus* species generally construct their nests in vertical earth banks in solid and firm soil, with several individuals per nest, sometimes in dense aggregations (Roberts & Brooks, 1987; González et al., 2005; Nates-Parra et al., 2006). Some species build their nests with lateral burrows each leading to a single horizontal cell, while other species organize the nests into a cluster of more or less horizontal cells in a cavity, similar to the nests of some Augochlorini (Claude-Joseph, 1926; Michener, 2007). Although we know some aspects of the natural history of *Caenohalictus*, few studies (Nates-Parra et al., 2006; Michener et al., 1978) have focused on studying more details of the nesting behavior, social structure, and nest architecture of the Andean species belonging to this genus, as is the case of *Caenohalictus alexandrei* that was recently described by Celis et al. (2014) from the Savanna of Bogota Plateau in the Eastern Andes of Colombia.

Due to the lack of knowledge on the natural history of Halictinae in Colombia, the aim of this study was to present some aspects about the basic biology of *Caenohalictus alexandrei*, to complement the previously taxonomic descriptions and to expand the knowledge about the behavioral traits of this species.

## MATERIAL AND METHODS

### Nesting site

The nests of *Caenohalictus alexandrei* were studied at two nesting sites in the Eastern Andes of Colombia in the Cundinamarca department: Zipaquirá (Z) (San Jorge, 4.998722°N 74.021083°W, 2780 m.a.s.l.) and Cajicá (C) (Campus Universidad Militar Nueva Granada [UMNG], 4.9450°N, -74.0119°W, 2580 m.a.s.l.) (Fig. 1). External characteristics, consistency of the soil and accompanying vegetation and fauna were determined for both sites.

### Field observations

The observations were made over four months (June-September/2009) at nesting site C and eight months (August/2011 to March/2012) at nesting site Z. At the end of each observation period, some of these nests were excavated. In each nesting site, the active nests were observed every two days, from 8:00 h to 16:00 h. During each observation session, each nest was observed by the same observer continuously for 30 min, for 8 h a day, totaling 1,472 h of observation. The order in which the nests were observed was randomly assigned each day to avoid observation biases. We categorized weather as "rainy" or "sunny", at each time observation intervals (half-hourly) in both sites. The activity of the bees was recorded following the methods described by Michener et al. (1955). Timing of the bee activities was in minutes

with the mean and  $\pm$  standard error,  $n$  refers to the total number of each activity carried out during the total time observations (1,472 h). Other data refer to the average of the total number of females in which an activity (for example, foraging) was observed across the sampling period. Nests were numbered individually, and their entrances were marked with colored flags (Figs. 2B and C) and with colored oil paint (Fig. 2D). The length of activity periods, weather, departures and arrivals of the adult females and the presence of pollen loads were recorded as described by Dalmazzo & Roig-Alsina (2012).

The same methodology was used for sleeping cavities (inhabited by males) which were observed just in site Z; two of them were excavated to describe the internal structure.

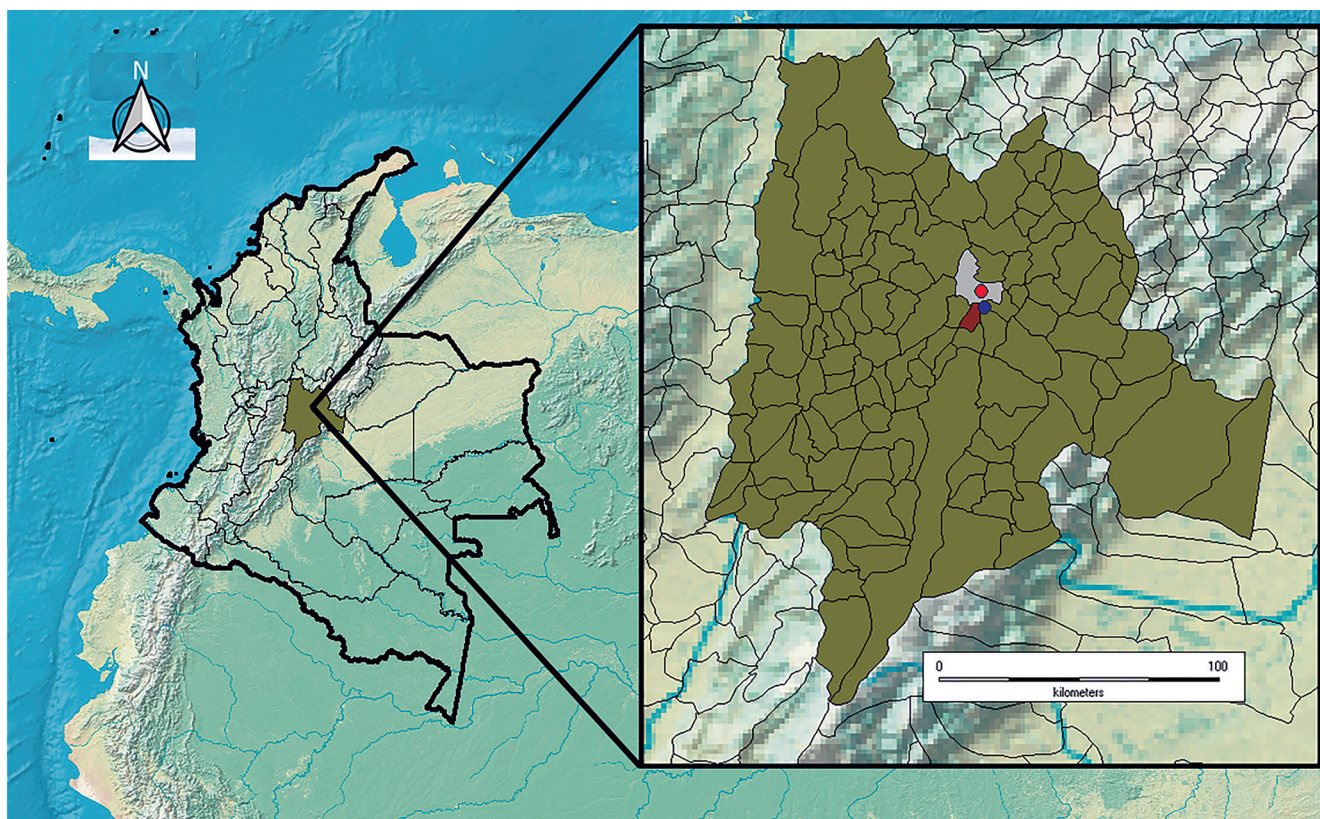
### Excavation and description of nests

Eight nests (five in nesting site Z and three in nesting site C) were chosen for excavation. They were selected based on their conspicuous activity (females constantly leaving, entering, or guarding the nests entrance). The methodology described by Sakagami & Michener (1962) and Abrams & Eickwort (1980) was followed for nest excavation, i.e., a mixture of talcum powder and water was injected through the nest entrance to assist us in following the nest structure as it was excavated. The nests were excavated using a field knife and a pointer. In order to capture all the adults from the nest, the excavation was carried out early in the morning (7:00 h), before the bees flew out of the nest to begin foraging.

Observations and measurements of the cells and their contents were conducted at the Entomology Laboratory in Campus Universidad Militar Nueva Granada (UMNG) by means of a stereomicroscope Stemi SV 11 (Zeiss). Measurements are given in millimeters. Adult specimens (females and males) found in the excavated nests were collected, pinned, and identified. These specimens were subsequently deposited in the entomological collection of the Universidad Militar Nueva Granada, Cajicá, Colombia (UMNG-ins) and Instituto de Ciencias Naturales (ICN) of the Universidad Nacional de Colombia, Bogotá, Colombia. Eggs, larvae and pupae found in the excavated nests were described and preserved in 70% ethanol, and were deposited in the UMNG-ins.

### Pollen masses

Samples of pollen obtained from cells of four nests excavated from nesting site Z (Table 1, \*) were processed following the acid acetolysis technique (Erdtman, 1960). We analyzed the pollen resource of only one cell from each one of the four nests. Thirty microscope slides were obtained from each pollen cell, totaling 120 slides. For each nest, we randomly selected five slides; thus, 20 slides were analyzed out of all the 120 slides. An average was estimated for the 20 slides, in which the pollen grains were counted and the percentage of different



**Figure 1.** Geographical location of the nesting sites. Green area represents the Cundinamarca department. Gray area corresponds to the municipality of Zipacquirá with the nesting site Z (red circle) and brown area corresponds to the municipality of Cajicá with the nesting site C (blue circle).

morphotypes estimated. The samples were analyzed by optical microscopy using an Axio Lab.A1 (Zeiss) at 40X and 100X, and a photographic record of the pollen grains was made. Identifications of the pollen grains were made by comparing the morphotypes with the pollen samples deposited in the pollen library of the UMNG (Palinoteca Grupo Biodiversidad y Ecología de Abejas Silvestres [PBEAS]), as well as by comparisons with the pollen obtained directly from the plants located near the nesting sites, which also were deposited at the PBEAS. In addition, we also used palynological atlases and descriptions by Jiménez & Rangel (1997), Sánchez & Lupo (2009), Silva et al. (2010), Giraldo et al. (2011) and Mercado-Gómez et al. (2011). Exsiccatae with the plants visited by *C. alexandrei* were made for the most precise identification taxonomic category as possible using taxonomic keys, consulting specialists and through comparison with exsiccatae previously housed in the UMNG herbarium, where the material of this study was deposited.

## RESULTS AND DISCUSSION

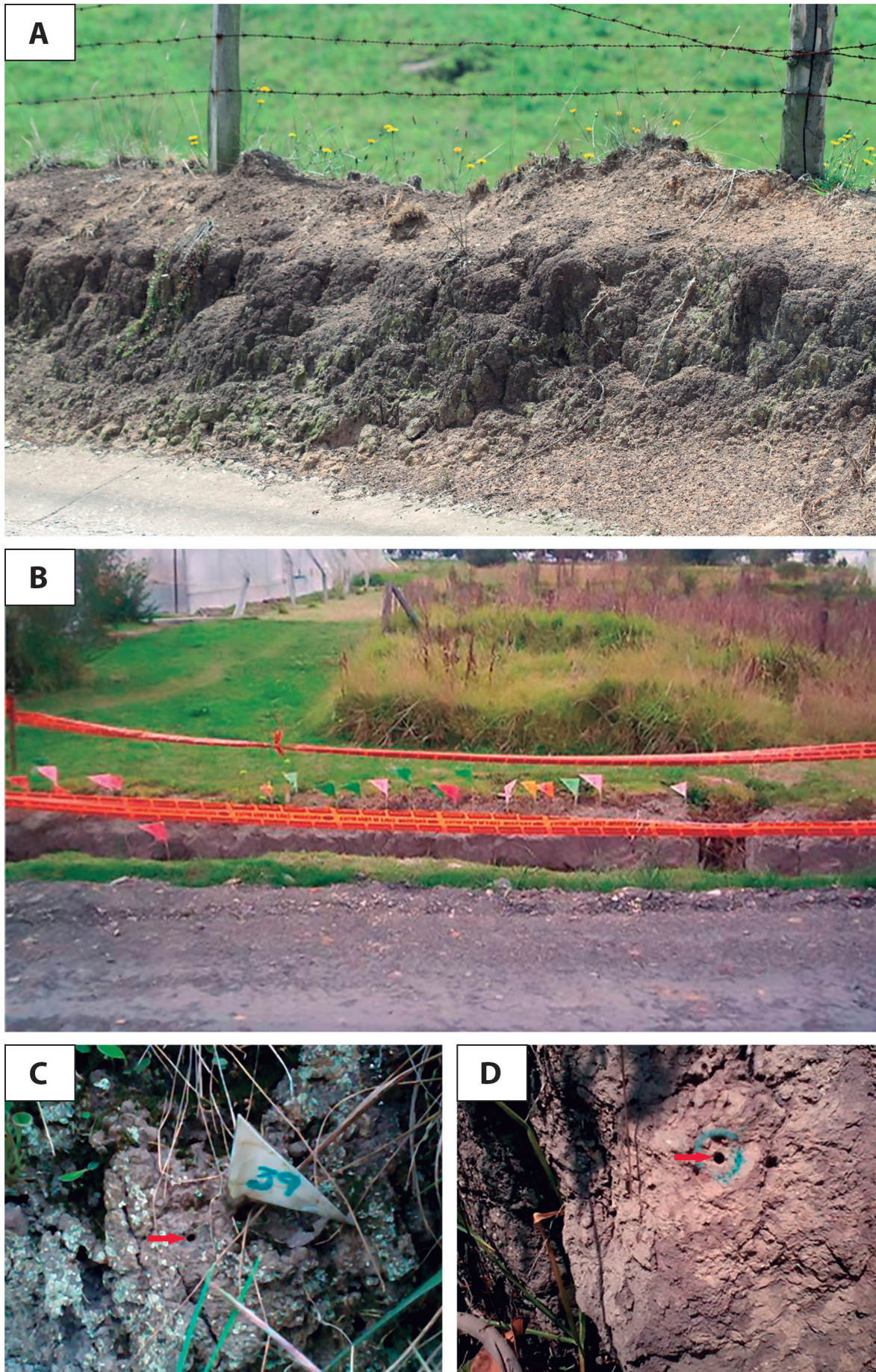
### Nesting site

*Caenohalictus alexandrei* is a soil nesting species whose females build their nests on vertical earth banks. This species can use banks along roads and streets with anthropogenic influence. Nesting site Z was an earth bank elevated 80 cm from the ground floor (Fig. 2A) and was very close to a restaurant parking lot. Nesting

site C, in turn, was located in a canal 90 cm deep into the ground (Fig. 2B) and was near greenhouses at the university campus.

The vertical earth banks in the nesting sites Z and C were exposed with uncovered surfaces or sparse associated vegetation (herbs and grasses) (Figs. 2A and B); there, the substrate was well-drained, arid and moderately loose soil with some compact regions, silty with some roots and considerable solar exposure (Fig. 2A). These characteristics also are present in nest of *Habralictus bimaculatus* Michener, 1979 and *Caenohalictus eberhardorum* Michener, 1979, both species nesting in a vertical, roadside bank of soft but firm, decomposed rock (Michener et al., 1978). *Agapostemon splendens* (Lepelletier, 1841), conversely, nests in sandy vertical bank fairly stable and sparsely covered with grass (LaBerge & Ribble, 1966). A moderate to compact soil makes nesting construction easier and provides a sufficiently stable substrate to prevent loose soil from collapsing when the bees attempt to continue building and deepening their nests (Michener et al., 1958; Sakagami & Michener, 1962; Michener & Kerfoot, 1967; Michener, 1977; Roubik, 1992). Soil composition, humidity, surface slope, degree of sun exposure and vegetation cover seem to be important factors not only for Caenohalictina, but at even broader taxonomic levels, since some Augochlorini – e.g., *Neocorynura pubescens* (Friese, 1916) (Michener, 1977) and *Pseudaugochlora* Michener, 1954 (Michener et al., 1958) – also have preference for vertical banks of bare, more or less compact soil.

*Caenohalictus alexandrei* formed aggregations at both nesting sites, which consisted of a cluster of nests



**Figure 2.** Nesting sites of *C. alexandrei* on earth banks in Colombia. (A) nesting site Z (Zipaquirá – San Jorge); (B) nesting site C (Cajicá – UMNG); (C-D) Red arrows indicate the entrance of the nest in the earth banks.

**Table 1.** Architectural characteristics and content of the excavated nests of *C. alexandrei*.

Nest №	Nesting site	Breeding cells			Empty cells and trace of old pollen	Pollen provisioning cells	Pollen storage cells	Cells with adult individuals	Total cells	№ females	№ dead females	№ males	Total individuals ♀+♂	№ entries of the nest <sup>e</sup>	№ lateral tunnels	Depth of the nest (cm)
		eggs	larvae	pupae												
1*	San Jorge	3	16	0	16	5	5	3	48	2	3	0	5	1	4	16
2*	San Jorge	6	13	1	6	1	0	6	33	8	0	0	8	1	4	16
3*	San Jorge	1	2	1	14	1	0	0	19	2	0	0	2	1	3	11
4*	San Jorge	0	1	0	67	3	3	3	77	2	5	3	10	1	8	13
5	San Jorge	0	3	0	3	0	0	1	7	2	0	0	2	1	5	6
6	Campus UMING	0	0	0	0	1	0	9	10	9	0	0	9	2	3	12.5
7	Campus UMING	0	2	0	2	1	0	4	9	5	0	0	5	1	6	8.5
8	Campus UMING	0	0	0	0	0	0	4	4	4	0	1	5	1	3	9.3

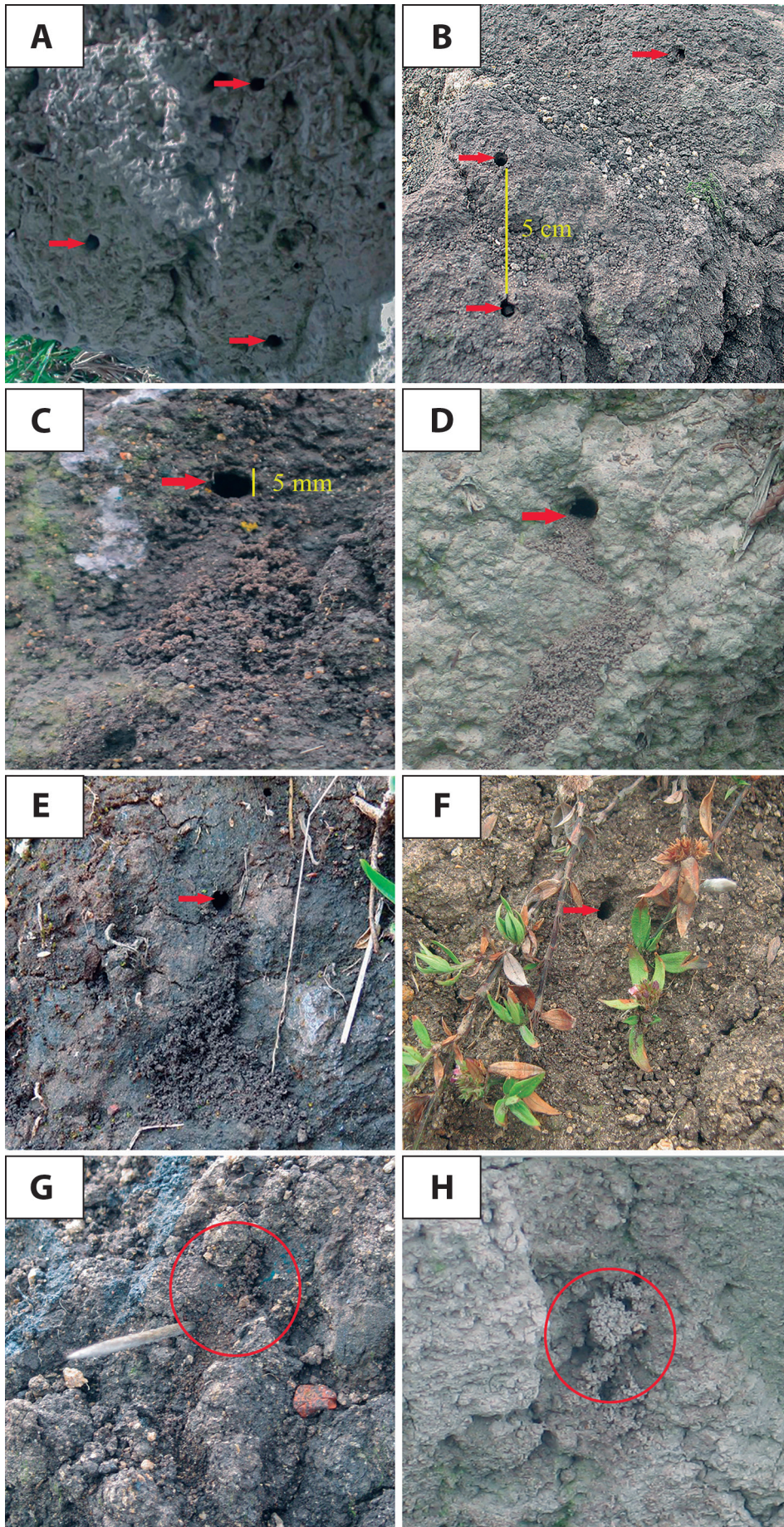
<sup>e</sup> № entries of the nest. Refers to the number of entrances to the same nest and that were used by adult females to enter and exit it.

\* Correspond to nests where pollen resource was used for the Acetolysis technique.

separated from each other by a few centimeters (5 to 7 cm) (Figs. 3A and B). A first aggregation of 13 nests was found occupying an area of 18.24 m<sup>2</sup> at nesting site Z (Fig. 2A). A second aggregation of 27 nests was found occupying an area of 3.44 m<sup>2</sup> at nesting site C (Fig. 2B), leading to a total of 40 nests. Sleeping cavities were found only at nesting site Z for a total number of 40. Roberts & Brooks (1987) and Nates-Parra *et al.* (2006) also reported an aggregative tendency for the genus *Caenohalictus*, and Nates-Parra *et al.* (2006) recorded for an unidentified species of this genus a total of 56 nests in 8 m<sup>2</sup> and a separation among them of 8 cm in average. In *C. eberhardorum* Michener, 1979, thousands of nests occupied an area about 8 m long and 1 m high and in *H. bimaculatus* Michener, 1979 the nests were separated by 2 to 4 cm in a bank of 2 m high and 2.5 m long (Michener *et al.*, 1978). There have been suggestions that such aggregations are formed, at least in some species, to increase defenses against natural enemies (Michener *et al.*, 1958; Sakagami & Michener, 1962; Michener, 1974). Such aggregations also result from a tendency for young adults to return to the vicinity of their natal nests (Albert & Packer, 2013; Michener, 1966). Michener *et al.* (1958) have confirmed that groups appear as a result of innate aggregative factors of the bees in response to preferences of habitat requirements. According to this idea, bees that nest in groups proceed in this way because the environment is optimal for the construction of nests in localized areas (Michener *et al.*, 1958).

Individual nests of *C. alexandrei* were located within the aggregations by their entrances or tumuli. The entrances consisted of a circular aperture in the soil of several sizes between 5-7 mm in diameter (Figs. 3A-F), and the tumuli consisted of an accumulation of compact soil around the entrance of the nest that contrasted with the straight surface of the soil and vegetation. Some nests did not present tumuli in their principal entrance, because these structures are very prone to be lost by heavy rains or wind that frequently erode them. Nates-Parra *et al.* (2006) reported similar characteristics from nests of *Caenohalictus* sp., with an entrance of 4 mm in diameter and a tumulus present in the entrance. LaBerge & Ribble (1966) reported for *A. splendens* an entrance of 8 mm in diameter. We infer that such a difference in size in the diameter of the circular aperture of the nests may be associated with the body size of females within Caenohalictina.

Sakagami & Moure (1967) reported nests of *Caenohalictus curticeps* (Vachal, 1903) from Brazil also in vertical earth banks on roadsides. Other species such as *Agapostemon nasutus* Smith, 1953, *A. splendens*, and *Caenohalictus sabanaensis* (pers. obs. by the authors) are selective in the choice of various degrees of soil inclination for nesting sites and are found exclusively in vertical earth banks (Sakagami & Michener, 1962; Michener & Kerfoot, 1967; Eickwort & Eickwort, 1969). Such characteristic does not seem to be exclusive to the Caenohalictini, since in Augochlorini it is also common to find reports of species that nest only in vertical banks, as reported by Michener & Kerfoot (1967) for nests of *Pseudaugochlora graminea* (Fabricius, 1804) and Michener (1977) for nests



**Figure 3.** Nests *C. alexandrei* in the earth banks. (A-B) aggregations of nests: (A) nesting site C; (B) nesting site Z: (C-F) Open nests entrances with traces of soil falling down the bank: (G-H) Close nests entrances with soil. Red arrows indicate the open entrances of the nest. Red circles indicate the close entrance of the nest.

of *N. pubescens*, two species that generally nest in vertical soil earth banks close to houses, near empty plots of land and in suburbs.

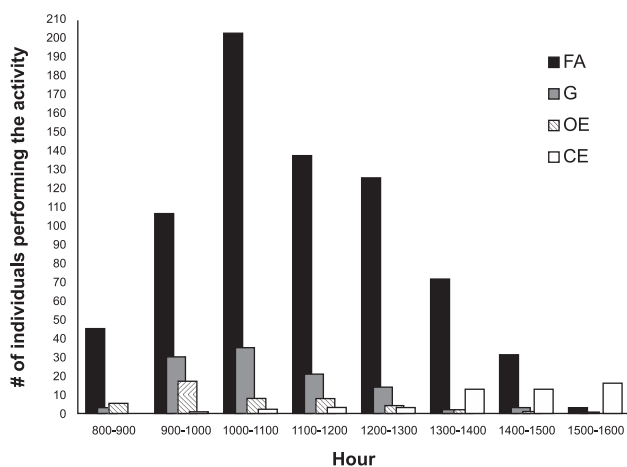
Nests of other bee species, including *Protandrena* sp., Colletidae sp. and *Lasioglossum* sp. were also observed by us near aggregations of *C. alexandrei*. Bee species of the cleptoparasitic genus *Sphecodes* Latreille, 1804 were frequently observed flying throughout the day around the nests of *C. alexandrei*. Michener et al. (1978) reported a similar associative behavior among bee species, where the *H. bimaculatus* nests were located near to *C. eberhardorum* nests, both species sharing the same nesting site. Those same authors also found the cleptoparasitic species *Microsphcodes truncaticaudus* Michener, 1979 in this *H. bimaculatus* + *C. eberhardorum* association, where males and females of this cleptoparasitic bee were commonly observed flying around the nesting areas or females inside the cells of *H. bimaculatus*.

### Field and behavioral observations

#### Behavioral observations of females

Through a total of 925 observations of females in the field obtained during a total of 1,472 h and in 40 nests of *C. alexandrei*, four external activities were determined: foraging activity (FA), guarding of the principal entrance (G) and opening (OE) and closure of the nests entrance (CE). 77.84% of the individuals presented FA during the total observation time, followed by G in 11.78% of the nests, CE in 5.51% of the nests and OE in 4.86%. All the activities occurred from 8:00 h to 16:00 h with the highest frequency peaks from 9:00 h to 14:00 h, except for CE, which was frequently observed during the afternoon from 13:00 h to 16:00 h and at sunset (Fig. 4).

These same four behavioral activities were reported in the Caenohalictini species *A. nasutus* (Eickwort & Eickwort, 1969; Roberts, 1969) and *Agapostemon*



**Figure 4.** Number of active females on nests where the various behavioral activities were registered at different times of the day and present in the nesting site that were under observation. (FA) = foraging activity, (G) = guarding of the principal entrance, (OE) = opening of the entrance and (CE) = closure of the entrance of the nest.

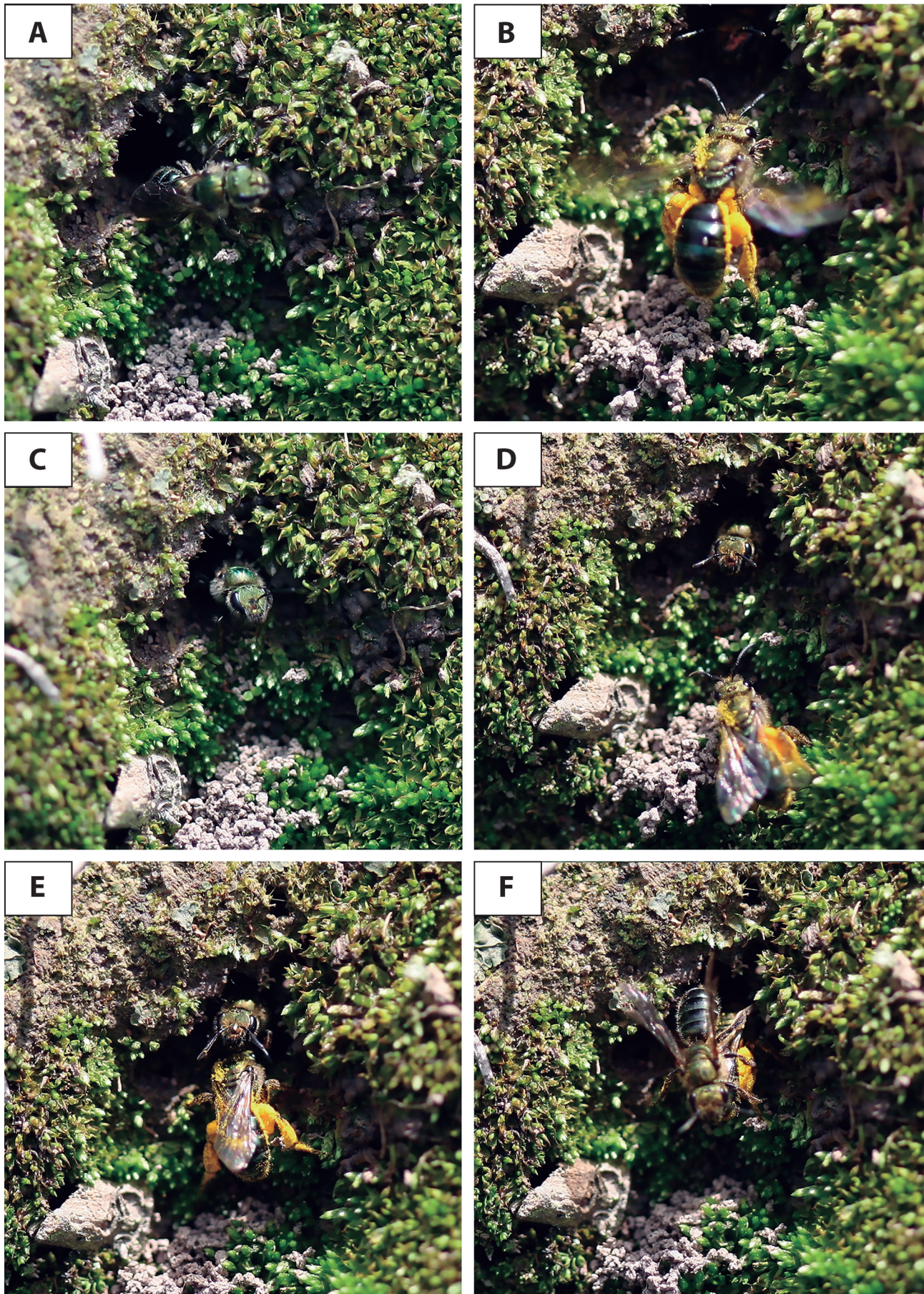
*virescens* (Fabricius, 1775) (Abrams & Eickwort, 1980); in the Halictini *Halictus farinosus* Smith, 1853 (Nye, 1980); and in the Augochlorini *Pseudaugochlora* sp. (Michener & Kerfoot, 1967) and *Augochlora daphnis* Smith, 1853 (Dalmazzo & Roig-alsina, 2012). FA was the most frequently observed activity, because the females must actively collect pollen and nectar to construct a pollen ball (mass provisioning) where all of the larval food is supplied before the egg is laid (Abrams & Eickwort, 1981).

**Opening of the entrance of the nest (OE):** This activity is strongly related to the foraging and guarding activities, because it determines, in most cases, the beginning of foraging. This activity was most frequently performed from 8:00 h to 13:00 h, with the highest activity peaks being from 9:00 h to 12:00 h, a time period in which between 8 and 17 females presented this activity; in this time range, FA was also higher (Fig. 4). In the development of OE, females of *C. alexandrei* were observed removing the soil that sealed the entrance of the nest; this soil formed tumuli around the entrance or fell down the earth bank, leaving the principal entrance free and well-defined (Figs. 3C-F). A minority of nests open in the afternoon from 12:00 h to 14:00 h, even the same nest can open and close at least twice on the same day due to rain (Fig. 4).

**Closure of the entrance of the nest (CE):** Females were observed sealing the entrance of the nest with soil (Figs. 3G and H). This activity was more frequently carried out in the afternoon and at sunset, with the highest frequency peak from 13:00 h to 16:00 h, a time at which the entrances of approximately 13-16 nests were closed each hour (Fig. 4). Once the entrance was sealed, all other activities ceased. This CE activity was also carried out in the morning from 9:00 h to 13:00 h, albeit less frequently (Fig. 4) and mostly associated with rain.

This tendency of opening and closing the entrance of the nests was reported by LaBerge & Ribble (1966), Eickwort & Eickwort (1969), Roberts (1969), and Abrams & Eickwort (1980) for the genus *Agapostemon*. According to Eickwort et al. (1986), nests of *Dufourea novaeangliae* (Robertson, 1897) remain open while the bees are foraging, but they are permanently closed with soil at the end of daily activities (from 15:30 h to 16:00 h); after the time of closing, the bees are unlikely to be seen on flowers. Zobel & Paxton (2007) suggested possible cleptoparasitism of the nests as a factor in the closing of the entrance, because many cleptoparasitic bees inspect nests for oviposition. According to Roubik (1992), CE is also influenced by weather factors and especially by heavy rains since humidity and floods must be avoided inside the nests. Thus, this activity is important to ensure the survival of both the adult and the immature bees.

**Foraging activity (FA):** Females were observed leaving and entering the nests (Fig. 5). This activity began soon after the sunlight hit the earth banks (8:00 h) and finished in the afternoon (16:00 h) (Fig. 4). We observed females foraging for pollen on flowers near the nests,



**Figure 5.** Nests of *C. alexandrei* in the earth banks. (A) Female leaving the nest to perform foraging activity; (B, D-E) Female arrival and entry to the nest with a load of pollen in her hind femoral, tibial and ventral scopa and contact with the guardian female; (C) Female performing guarding activity in the entrance of the nest and waiting for the returns of another nest female's; (F) Exit of the guardian female to forage just after the return of another adult female from the nest.



where the males were also observed flying. After foraging, most females returned to their nest, carrying pollen loads in their hind femoral and metasomal scopa (Figs. 5B, D and E). When returning to the nest, females exhibited erratic flight and inspected cracks and small holes in the earth bank, before entering their respective nests. FA was performed more frequently from 9:00 h to 14:00 h, with the highest activity peaks being from 9:00 h to 12:00 h, during which time 106-200 females were observed foraging (Fig. 4).

Michener & Wille (1961) observed the first flights of *Lasioglossum inconspicuum* (Smith, 1853) from nests in the morning from 6:40 h to 9:00 h, and they reported that these first flights can be delayed on cloudy days. In the afternoon, a late female of this species was seen leaving a nest as late as 16:55 h. For *Caenohalictus* sp., Nates-Parra et al. (2006) reported FA occurring from 9:00 h to 16:00 h in sunny weather. For *A. nasutus*, Eickwort & Eickwort (1969) reported FA carried out from 8:00 h to 13:00 h, with the return of these bees with pollen to the nest occurring most frequently at 10:45 h. Roberts (1969, 1973) reported females and males of *Agapostemon* obtaining nectar or pollen from the flowers during morning hours, with the best foraging time being around 9:00 h, when the weather is warm and sunny, and a rapid decrease in females on flowers occurring in the afternoon.

*Caenohalictus alexandrei* was seen foraging and resting on flowers adjacent to or near the edges of roads where the nesting sites were located. These flowers belong to plant species of the families Poaceae (*Pennisetum clandestinum*), Asteraceae (*Taraxacum officinale*, *Bidens pilosa*, *Hypochaeris radicata*, and *Senecio madagascariensis*) and Fabaceae (*Trifolium repens* and *Trifolium pretense*). Nates-Parra et al. (2006) reported that species of *Caenohalictus* visited the following ornamental plants: *Crososmia aurea* (Iridaceae), *Jasminum* sp. (Oleaceae) and *Antirrhinum majus* (Scrophulariaceae).

**Guarding of the entrance (G):** During FA, the nests of *C. alexandrei* were constantly guarded by adult females that remained at the entrances of nests, blocking them with their heads. The head of a guarding female was visible at such an entrance until the return of another female (Figs. 5C and D); when other adult females came back to the nest with pollen loads (Figs. 5D and E), the guarding female departed to forage (Fig. 5F). Usually, as soon as a female left the nest, another one showed its head at the entrance (as long as the nest was made up of more than two adult females). The first guardians appeared at the entrances of the nests from 8:00 h, and the nests were guarded throughout FA. However, G decreased in the afternoon from 13:00 h to 16:00 h, at which time FA also decreased considerably and closure of nest entrances began. G and FA are very related activities, so when the FA ends, G also ceases. As observed, both activities ended after 16:00 h (Fig. 4).

Similar guarding activity was reported by Eickwort & Eickwort (1969) in *A. nasutus* nests in that when the

pollen-foraging activity ceased, the guard bees were absent. Roberts (1969), Abrams & Eickwort (1980, 1981) and Dalmazzo & Roig-Alsina (2012) also reported G activity in *Agapostemon* and *Augochlora*, in these genera, an adult female with its head peeking out of the nest entrance to protect it during foraging was common. According to Abrams & Eickwort (1980) and Dalmazzo & Roig-Alsina (2012), female guards of *A. virescens* and *A. daphnis* that leave a nest to forage are immediately replaced by other females of the nest.

In our observations, *Sphecodes* sp. females constantly flew around the aggregations, and one female was seen attempting to enter a nest of *C. alexandrei*. In the presence of this *Sphecodes* female, the guard female attacked it with open mandibles, ramming and pushing the *Sphecodes* female toward the outside of the nest, and as the attack continued, the guard female turned around, plugging the hole with her metasomal terga. Thus, guarding behavior functions to protect the nest against these cleptoparasitic bees. Sometimes the slightest movement was enough to cause a guard's disappearance into the nest for some minutes; similar behavior by the males in the cavities was also seen. During nest excavation, male pupa belonging to *Sphecodes* genus were obtained, suggesting that some nests had been parasitized by those bees. Roberts (1969) and Michener (2007) reported that bees from the genus *Sphecodes* are usual cleptoparasites in various nests of Halictinae. Therefore, Abrams & Eickwort (1980) reported the presence of *Sphecodes davisii* Robertson, 1897 males emerging from the nests of *Agapostemon virescens* (Fabricius, 1775) and Eickwort (1985) reported *Sphecodes arvensiformis* Cockerell, 1904 as the cleptoparasite most frequently present in nests of *Halictus farinosus* Smith, 1853.

The time of G activity of females of *C. alexandrei* was not measured, but could be similar to the time that a female of the same nests spends exiting and entering the nest during foraging activity, and the replacement of the guardian female. Abrams & Eickwort (1981) reported that *A. virescens* females remain at their posts, guarding the nests for at least 1 h.

### Behavioral observations of males

The males were observed in sleeping cavities and presented the following activities:

**Staying at the cavity entrance (S):** Males remained with their heads at the entrance of the cavities before they left, since they only inspected the exterior previously to go out and fly around the earth banks and the flowers for resources and females. In one first cavity, a male was observed at the entrance from 10:30 h to 12:10 h before finally departing; in a second cavity, a male was observed from 10:57 h to 11:03 h; and in a third and fourth cavity, males were observed for only 2 and 3 min, respectively, indicating that the time of S was variable.

**Cavity departure (D):** Males were observed leaving cavities they inhabited to fly rapidly towards the flowers near

the nesting site, where they either perched immobile or stopped occasionally in the flowers, leaves or branches of *T. officinale* and *B. pilosa*, or on the ground of the same earth bank. Later, they returned to the cavities exhibiting erratic flight and inspecting cracks and small holes in the bank. In some cases, the males did not determine cavities and were seen entering cavities different from the ones they had exited. D was the most frequent activity that males performed.

**Corporal grooming (C):** The males cleaned their bodies, antennae and wings with their anterior and hind legs. This activity was usually performed on flowers or earth banks. C activity usually took place during the immobile time and before they returned to the cavities.

Roberts (1969) and Abrams & Eickwort (1980) reported similar behavior for males in sleeping cavities of several species of *Agapostemon*, where the males were seen leaving from cavities and flying to the flowers to obtain nectar or pollen; this activity was generally performed in the morning (9:00 h) and into afternoon hours. D was the most frequently performed activity, because males, like females, must go out in search of food. Eickwort & Eickwort (1969) reported *A. nasutus* males were seen flying near *Lantana camara* flowers. Abrams & Eickwort (1980) reported S activity in *A. virescens*, where the males were found with their heads in the entrance of the sleeping cavity for roughly 20 min, after they were observed leaving to the cavity in search of food. Some *C. alexandrei* males were found in females' nests, and according to Dalmazzo & Roig-Alsina (2015), *Augochlora phoemonoe* (Schrottky, 1909) emerged males stayed within the nest for 12-20 h, and did not participate in any activity. C was not performed by females, possibly to avoid pollen removal before they entered in the nest.

### Nest internal architecture

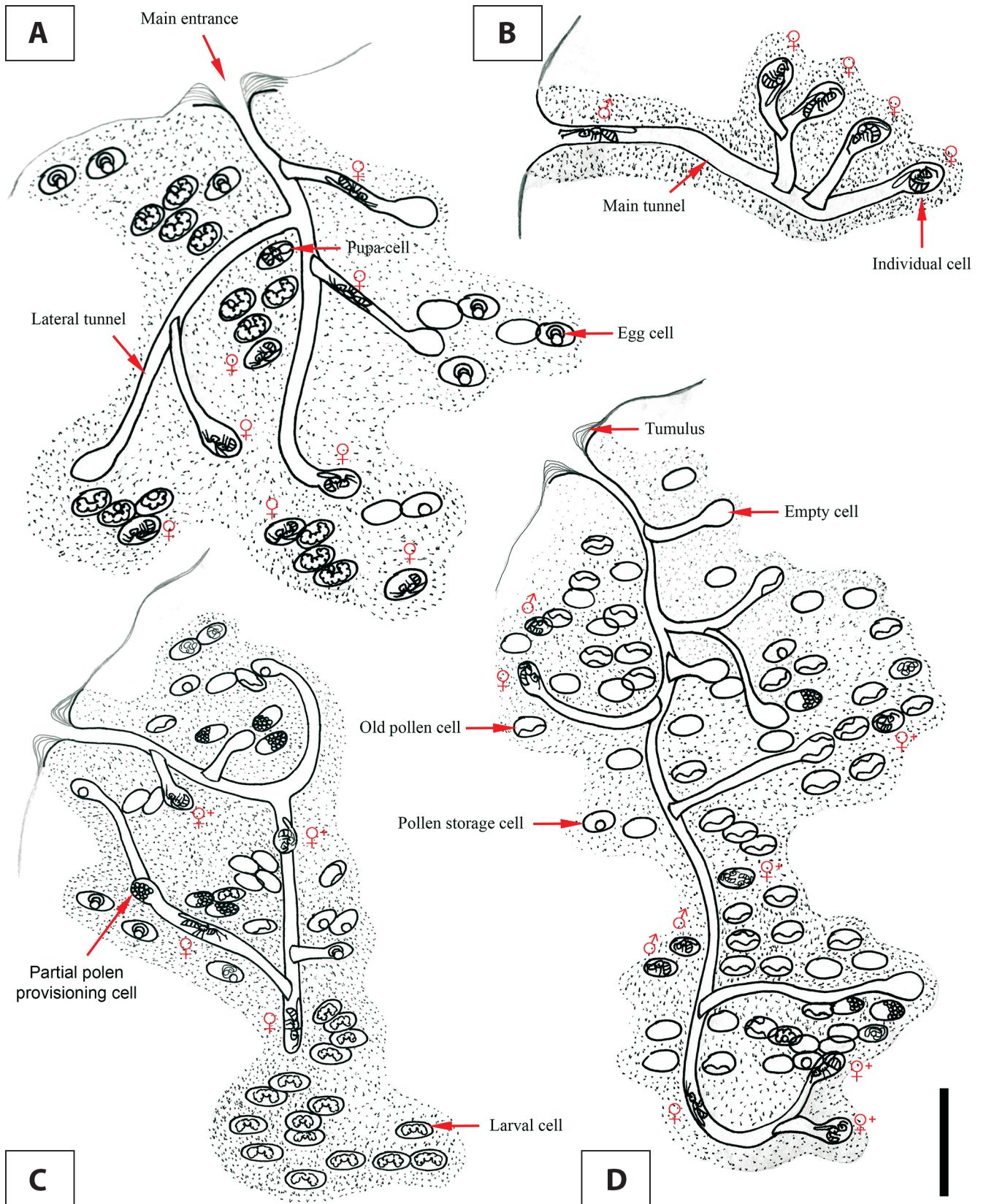
The nests of *C. alexandrei* differed considerably in their internal architecture, mainly in the number of cells (4 to 77 cells ( $25.87 \pm 9.03$ ,  $n = 8$ )) and the number of lateral tunnels (3 to 8 for a nest ( $4.5 \pm 0.62$ ,  $n = 8$ )). Michener *et al.* (1978) reported the maximum number of cells associated with a single nest of *C. eberhardorum* was eight. The level of complexity of the nest architecture depended on the number of adult females inhabiting them. All nests were inhabited by more than one adult female (2 to 9 females) ( $4.25 \pm 1.01$ ,  $n = 8$ ) (Table 1).

*Caenohalictus alexandrei* nests were composed of an entrance leading to a main tunnel, which penetrated vertically downwards to the interior of the earth bank; a minimum and maximum depth of 6 and 16 cm ( $12.40 \pm 3.19$ ,  $n = 8$ ), respectively, was observed. Attached to main tunnel were laterals extending from them sub-horizontally or horizontally into the ground. Usually empty cells were found at the end of each lateral, except in the nest 4, where two cells with a trace of old pollen were found (Fig. 6). Michener *et al.* (1978)

reported *C. eberhardorum* nests with a vertical entrance 7 to 20 cm deep (2.5 mm in diameter) and the laterals from 4 to 42 mm (2 mm in diameter), each leading to a single cell. Roberts (1973) reported *Agapostemon* sp. nests with a vertical entrance 20 to 150 cm deep and the laterals from 5 to 20 cm long. LaBerge & Ribble (1966) reported *A. splendens* nest 100 cm deep and with three laterals, one extending irregularly and somewhat downward for 30 cm, two with single cells containing pollen and with at least one of these cells terminal on the lateral. In comparison to those *Caenohalictini*, nests of *C. alexandrei* were not deep. Abrams & Eickwort (1980) remarked from *A. virescens* that both laterals and cells vary considerably by nest, depending on the number of females and the age of the nest. *Agapostemon* sp., *Caenohalictus curticeps* (Vachal, 1903) and *H. farinosus* nests exhibited a similar pattern with *C. alexandrei* in relation with the nest structure, such as a vertical main burrow and short horizontal laterals, each subtending a single horizontal cell (Sakagami & Moure, 1967; Eickwort & Eickwort, 1969; Roberts, 1969; Roberts & Brooks, 1987; Nye, 1980). Michener *et al.* (1958) mentioned that the function of this spatial arrangement of the cells around the main tunnel in the ground seems to be the increase in the number of cells within a limited space.

Nests 1, 2, 3, 4 and 6, with greater depth (11-16 cm) and number of cells (10-77), were considered as old nests due to their complex architecture (Table 1). During the nest observation period, these nests were very active. Conversely, nest 8 had fewer cells with respect to the other nests excavated and presented simple architecture, leading to it being considered as a newer nest. As other evidence for considering it as a new nest was due to during its excavation, four newly emerging adult females were found, each in their respective natal cell, and a male was also found together with them. This male did not have a cell of its own inside the nest and presumably it tried to mate with the females (Fig. 6B), although no mating was observed near the nesting sites. Similarly, no castes were established among adult females obtained from the nests, and no externally distinguishable queen was present.

Most *C. alexandrei* cells were occupied by immatures at different stages of development (egg, larva and pupa) (Table 1). Immature cells were found sealed with soil and were not directly connected to the laterals (Fig. 6), similar to what was reported by Michener *et al.* (1978) for *C. eberhardorum*. The cells of the nests were in a horizontal, subhorizontal, or slightly downward position. They were never in an upright position. The cells were an average of 7 to 10 mm in length ( $8.69 \pm 0.95$ ,  $n = 13$ ) and 5 to 7 mm in width ( $5.77 \pm 0.73$ ,  $n = 13$ ) and were constructed with compacted particles of soil. The external surface of the cells was irregular (Fig. 7A), and their internal surface was smooth and shiny (Figs. 7B and D). Both the cells and the laterals in nests were covered in a shiny coating similar to wax. The cells were ovoid, with the lower surface slightly flattened (Fig. 7A, B and D). Within *Caenohalictini* cell's size seems to be a variable feature. Michener *et al.*



**Figure 6.** Internal architecture of nests of *C. alexandrei*. (A) Nest 2; (B) Nest 8; (C) Nest 1; (D) Nest 4. ♀ and ♂ indicating on sex of adult bees, female and male respectively, ♀+ indicate dead females. Scale: 2 cm.

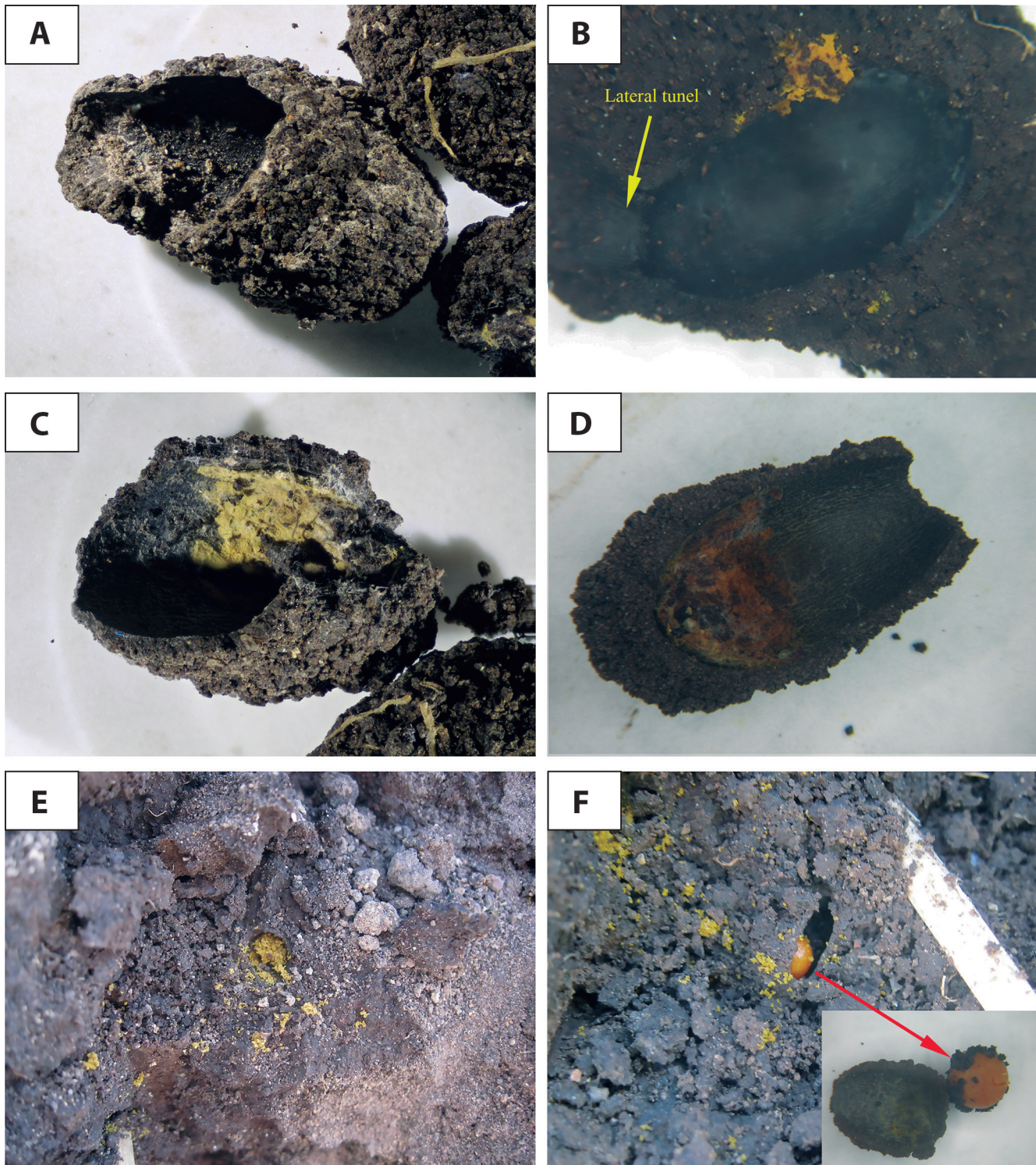
(1978) reported the measurements of cells of *C. eberhardorum* between 6 and 7 mm long and 3 mm in diameter, and Abrams & Eickwort (1980) reported the cells of *A. virescens* with sizes from 11 to 12 mm in length and from 6.5 to 7.0 mm in width. Therefore, the cells size of

*C. alexandrei* seems to be an intermediate range between these two species. The differences in the cells' measurements may be due to *C. alexandrei* is comparatively smaller (♀: 7.8-8.4 mm) (Celis et al., 2014) than *A. virescens* (♀: 11 mm) (Abrams & Eickwort, 1980), but

larger than *C. eberhardorum* (♀: 4.5-5.5 mm) (Michener, 1979). Covering cells and walls with the aforementioned waxy material according to Michener *et al.* (1958) is a very important task in controlling the humidity of the cells and in the control of mold growth, as it separates the pollen mass from the substrate, which tends to become quickly contaminated when it is in direct contact with the soil. Abrams & Eickwort (1980) reported these characteristics and attributes of shape and coating of

cells in *Pseudagapostemon*, *Ruizantheda*, *Caenohalictus* and *Habralictus* genera as well.

Many cells of *C. alexandrei* were uncovered in the course of excavation, and the contents of 207 of them were as follows: 108 empty cells (Figs. 7A and B) or cells with traces of old pollen (Fig. 7C and D), 12 partial pollen provisioning cells (Fig. 7E), 8 pollen storage cells (Fig. 7F), 49 breeding cells (Fig. 8) and 30 cells with individuals (with adult females or males) (Table 1).



**Figure 7.** Cells of nests of *C. alexandrei*. (A-B) Empty cells: (A) External appearance of cells; (B) Internal appearance of cells with a lateral tunnel; (C-D) Cells with traces of old pollen, possibly previously inhabited; (E) Cells with partial pollen provisioning, which are being provisioned for future use; (F) Pollen storage cells, ready for oviposition and its eventual use.

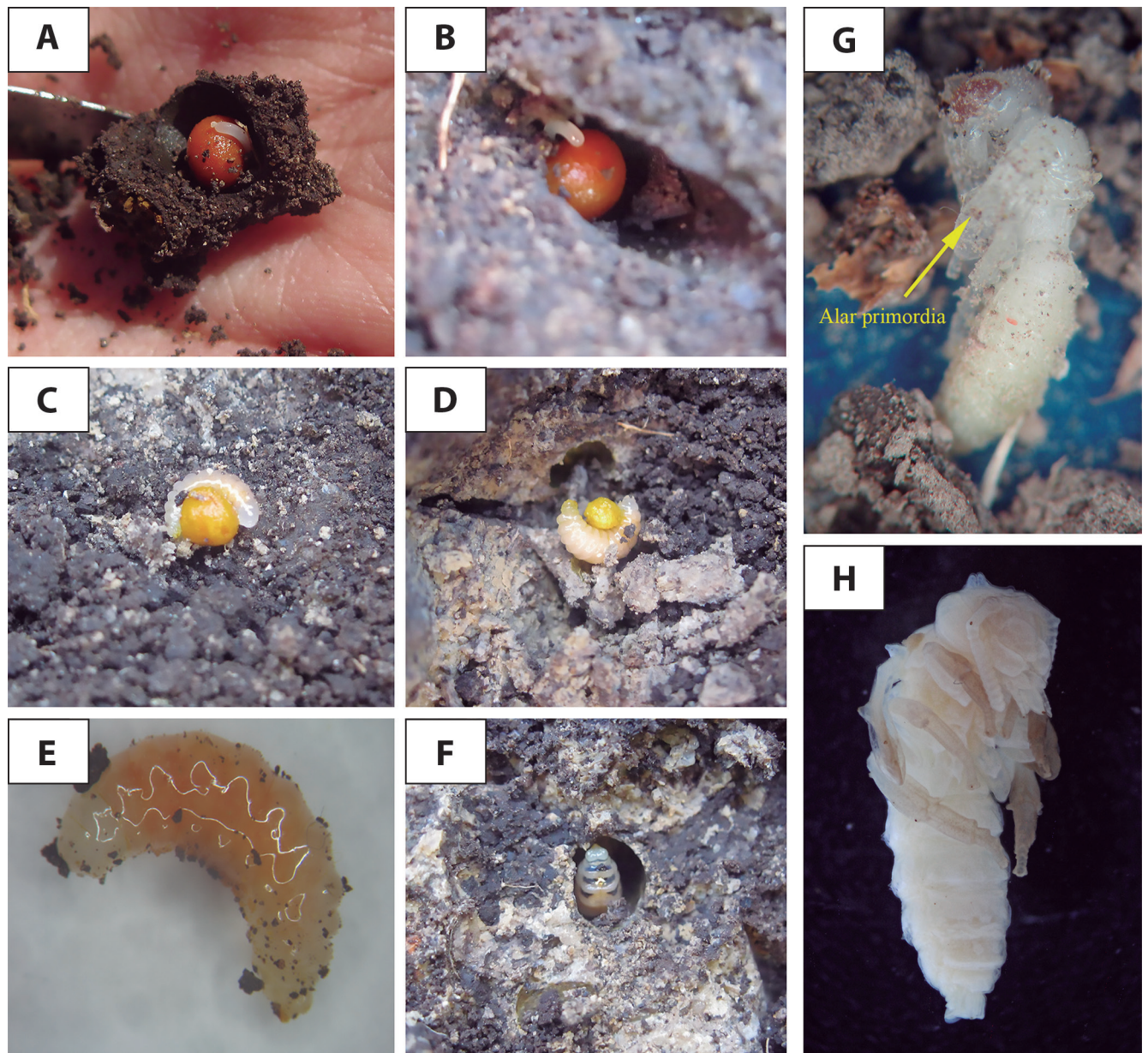
**Empty cells:** Correspond to new cells completely built, but without pollen content, and that would later be used to house the immature. The empty cells had narrow side openings that were approximately 3 mm wide (Fig. 7B). This union with the laterals permits a direct connection of the empty cells with the main tunnel and with the outside, facilitating their provisioning (Fig. 6).

**Cells with traces of old pollen:** Cells with dry pollen on the walls, corresponding to larval feces. Therefore, these cells belonged to emerged offspring (Figs. 7C and D).

**Partial pollen provisioning cells:** Cells with fresh and dry pollen content in the form of a loose mass on the floor of the cell and a direct connection with the main tunnel and outside. According to Abrams & Eickwort (1980), in nests of *A. virescens*, this pollen is the result of the first two or three trips that an adult female makes outside the nest

in search of the resources (pollen) for feeding the immature (Fig. 7E). These cells are connected to the laterals and main tunnel to facilitate the work of the adult females, which can enter and leave the cells quicker and easily with the pollen load. The presence of empty or partially provisioned cells attached to the laterals was also reported by Eickwort & Eickwort (1969) in *A. nasutus* nests.

**Pollen storage cells:** Cells with pollen in the form of a wet uniform mass. The pollen contained in the partial pollen provisioning cells was mixed with nectar and transformed into a bright yellow spherical mass (Fig. 7F). A single mass of pollen was found per cell. *Caenohalictus alexandrei* is a mass provisioner species and this spherical pollen mass is the complete supply to feed the offspring (Figs. 8A and B). These cells were found to be sealed with soil and isolated from the outside, likely to protect the brood after oviposition from environmental factors, fungi growth or the



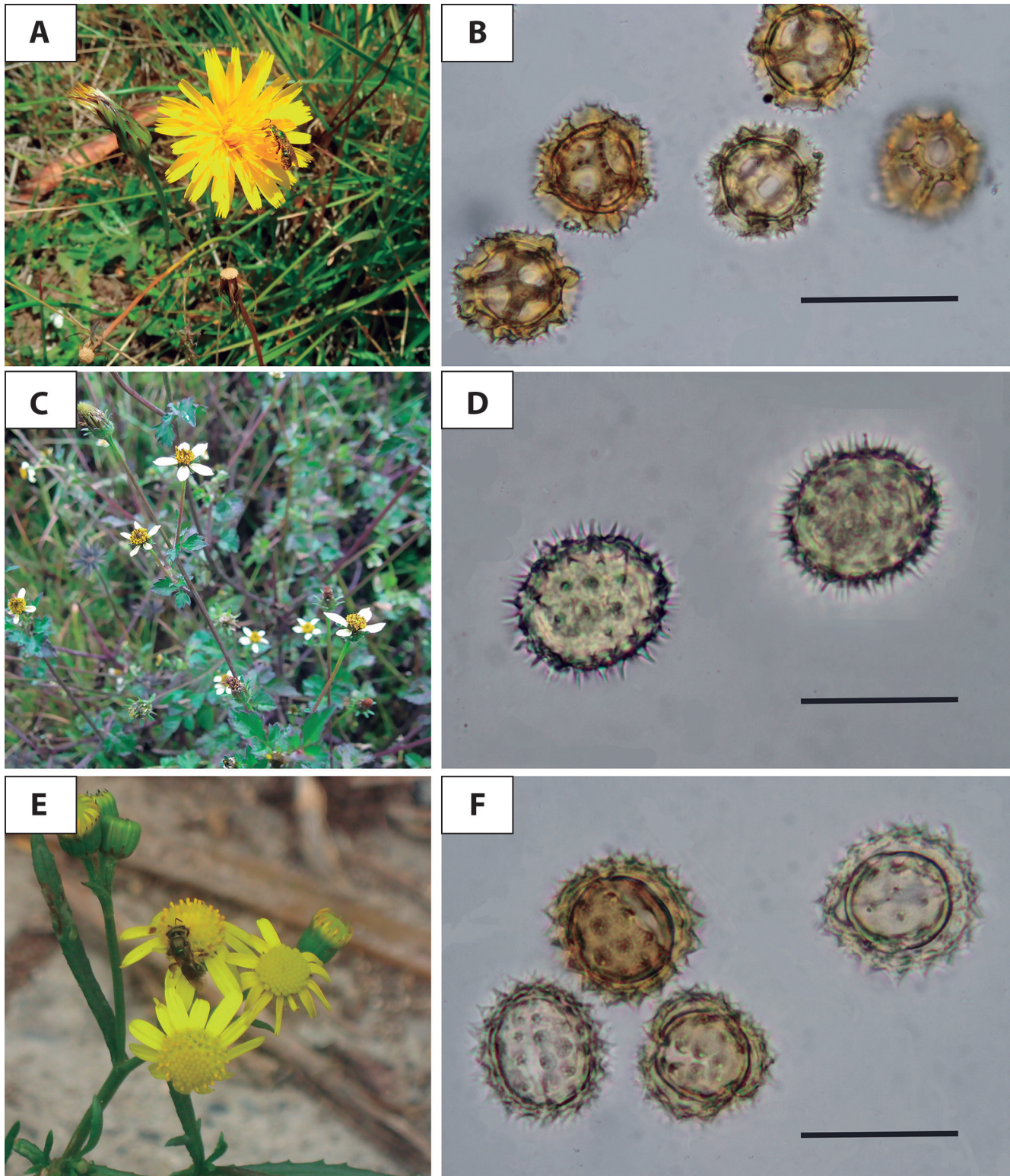
**Figure 8.** Breeding cells of *C. alexandrei*. (A-B) Eggs on pollen mass in the cell; (C-F) Larvae: (C) Young larva; (D-E) Larvae in intermediate stage with pollen in their digestive tract; (F) Mature larva; (G-H) Pupae: (H) Young pupa; (G) Mature pupa.

attack of cleptoparasites or predators. Sealing also allows the brood to develop under favorable conditions in terms of humidity and temperature to ensure success in the emergence of the adults (Roubik, 1992).

**Breeding cells (eggs):** Eggs of *C. alexandrei* are curved and white, with only one egg deposited on a single mass of pollen per cell. Each egg was placed on top of the

spherical pollen mass so that both ends of the egg were in contact with the mass. Each egg was observed with its axis parallel to the long axis of the cell (Fig. 8A and B).

**Breeding cells (larvae):** Larvae of *C. alexandrei* are white and C-shaped. The ventral parts of the young larvae surrounded the pollen mass (Fig. 8C). Larvae in an intermediate state of maturity swelled visibly; at this stage,



**Figure 9.** Pollinic resource used by *C. alexandrei*. (A-B) *Taraxacum officinale*: (A) Flower with adult female; (B) Pollen grains; (C-D) *Bidens pilosa*: (C) Flowers; (D) Pollen grains; (E-F) *Senecio madagascariensis*, (E) Flower with adult female; (F) Pollen grains. Scale: 100  $\mu$ m.

the larvae became yellow due to the presence of pollen in their digestive tract (Figs. 8D and E). The prepupae has white coloration with a dark stain on the dorsal part and with well-pronounced tubercles forming thoracic ridges (Fig. 8F), as also reported for the prepupae of *Agapostemon splendens* (Lepelletier, 1841) and *Agapostemon radiatus* (Say, 1837) (LaBerge & Ribble, 1966).

**Breeding cells (pupae):** Pupae are shaped like an adult. The young pupae are completely white and wingless, and present movement of the apical abdominal segments 3 to 5 (Fig. 8H). The mature pupae have reddish eyes, and the alar primordia began to form (Fig. 8G). Later, the pupae become dark acquiring the characteristic colors of the adults (metallic olive-green). At this late stage of development, the pupae present movements in the apical abdominal segments and the hind legs.

**Cells with adults:** Cells occupied by emerged and dead adult bees. We could not establish the cause of death.

### Sleeping cavities

The sleeping cavities of the males were simple. Males were seen using a cavity as a place of refuge and rest; this cavity was excavated and consisted of a single entrance that led to a single main tunnel extending 2.5 cm deep into the ground, ending in a single cell. Except for those present in nests, males are solitary; no more than one male was observed inhabiting a sleeping cavity, and no cooperation was observed in building or preparing these cavities.

### Pollen masses

Pollen resource obtained from *C. alexandrei* nests corresponds to exotic plants belonging to the Asteraceae family, suggesting that *C. alexandrei* is an oligolectic species. The percentage of each pollinic types found in the 20 samples was as follows: *T. officinale* (93.83%), *B. pilosa* (2.17%), *H. radicata* (2.10%) and *S. madagascariensis* (1.9%) (Fig. 9). Abrams & Eickwort (1980) and Michener (1977) also reported *A. virescens* and *N. pubescens* bees foraging on Asteraceae, mainly on *B. pilosa* flowers.

The Asteraceae pollen values found in nests of *C. alexandrei* were high due to possible factors such as: season-round presence, high local abundance, frequent presence in areas with anthropic disturbance and proximity to nesting sites. Roberts (1969) reported that bees of the genus *Agapostemon* do not go beyond a few feet away from their nests to find pollen and nectar. Solomon (1970) mentioned that Asteraceae flowers are very attractive to many bees due to their yellow coloration and for the high production of both pollen and nectar resources, in comparison with other plant families that lack one of these two resources, or where the pollen and nectar are present in small quantities or the nectar is quite diluted.

**AUTHORS' CONTRIBUTIONS:** CJC: Methodology, Visualization, Data Curation, Formal Analysis, Investigation, Writing – original draft; CJC, MLA: Conceptualization; MLA, JRC: Supervision; CJC, MLA, JRC: Writing – review & editing. All authors actively participated in the discussion of the results, they reviewed and approved the final version of the paper.

**CONFLICT OF INTEREST:** Authors declare there are no conflict of interest.

**FUNDING INFORMATION:** The authors are grateful to Vicerrectoría de Investigaciones UMNG for financial support (CIAS 1063).

**ACKNOWLEDGMENTS:** We thank the colleagues at the Bee Biodiversity and Ecology research group (BEAS) from UMNG for their assistance and support in the field trips. Alexander Escobar for his assistance in obtaining the photographs in the field. Marcos Fianco, Mario Cupello and Jose Ricardo Assmann for the comments on the manuscript.

### REFERENCES

- Abrams, J. & Eickwort, C.G. 1980. Biology of the communal sweat bees *Agapostemon virescens* (Hymenoptera: Halictidae) in New York state. *Search Agriculture*, 1: 1-20.
- Abrams, J. & Eickwort, C.G. 1981. Nest switching and guarding by the communal sweat bees *Agapostemon virescens* (Hymenoptera: Halictidae). *Insectes Sociaux*, 28: 105-116.
- Albert, J.R. & Packer, L. 2013. Nesting biology and phenology of a population of *Halictus farinosus* Smith (Hymenoptera, Halictidae) in northern Utah. *Journal of Hymenoptera Research*, 32: 55-73. <https://doi.org/10.3897/jhr.32.4646>.
- Celis, C.J.; Cure, J.R. & Aguilar-Benavides, M.L. 2014. Two new species of *Caenohalictus* Cameron, 1903 (Hymenoptera: Halictidae) from Colombia. *Zootaxa*, 3786(5): 574-586. <https://doi.org/10.11646/zootaxa.3786.5.5>.
- Claude-Joseph, E.C. 1926. Recherches biologiques sur les hyménoptères du Chili (Mellifères). *Annales des Sciences Naturelles, Zoologie*, 10(9): 113-268.
- Cure, J.R. 1989. Revisão de *Pseudagapostemon* Schrottky e descrição de *Oragapostemon*, gen. n. (Hymenoptera, Halictidae). *Revista Brasileira de Entomologia*, 33(2): 229-335.
- Dalmazzo, M. & Roig-Alsina, A. 2012. Nest structure and notes on the social behavior of *Augochlora amphitrite* (Schrottky) (Hymenoptera, Halictidae). *Journal of Hymenoptera Research*, 26: 17-29. <https://doi.org/10.3897/jhr.26.2440>.
- Dalmazzo, M. & Roig-Alsina, A. 2015. Social biology of *Augochlora* (*Augochlora*) *phoemone* (Hymenoptera, Halictidae) reared in laboratory nests. *Insectes Sociaux*, 62: 315-323. <https://doi.org/10.1007/s00040-015-0412-8>.
- Dalmazzo, M.; González-Vaquero, R.A.; Roig-Alsina, A. & Debandi, G. 2014. Halictidae. In: Roig-Juñent, S.; Claps, L.E. & Morrone, J.J. (Eds.). *Biodiversidad de Artrópodos Argentinos*, vol. 4. San Miguel de Tucumán Universidad Nacional de Tucumán. p. 203-215.
- Danforth, B.N.; Brady, S.G.; Sipes, S.D. & Pearson, A. 2004. Single-Copy Nuclear Genes Recover Cretaceous-Age Divergences in Bees. *Systematic Biology*, 53(2): 309-326. <https://doi.org/10.1080/10635150490423737>.
- Danforth, B.N.; Eardley, C.; Packer, L.; Walker, K.; Pauly, A. & Randrianambinintsoa, F.J. 2008. Phylogeny of Halictidae with an emphasis on endemic African Halictinae. *Apidologie*, 39: 86-101. <https://doi.org/10.1051/apido:2008002>.
- Eickwort, G.C. 1969. A Comparative Morphological Study and Generic Revision of the Augochlorine Bees (Hymenoptera: Halictidae). *The University of Kansas Science Bulletin*, 48: 1-524.
- Eickwort, G.C. 1985. The Nesting Biology of the Sweat Bee *Halictus farinosus* in California, with Notes on *H. ligatus* (Hymenoptera: Halictidae). *Pan-Pacific Entomologist*, 61(2): 122-137.
- Eickwort, G.C. & Eickwort, K.R. 1969. Aspects of the biology of Costa Rican halictine bees, I. *Agapostemon nasutus* (Hymenoptera: Halictidae). *Journal of the Kansas Entomological Society*, 42(4): 421-452.

- Eickwort, G.C.; Eickwort, J.M.; Gordon, J. & Eickwort, M.A. 1996. Solitary behavior in a high-altitude population of the social sweat bee *Halictus rubicundus* (Hymenoptera: Halictidae). *Behavioral Ecology and Sociobiology*, 38(4): 227-233. <https://doi.org/10.1007/s002650050236>.
- Eickwort, G.C.; Kukuk, P.F. & Wesley, F.R. 1986. The nesting biology of *Dufourea novaeangliae* (Hymenoptera: Halictidae) and the systematic position of the dufoureae based on behavior and development. *Journal of the Kansas Entomological Society*, 59(1): 103-120.
- Erdtman, G. 1960. The acetolysis method: a revised description. *Svensk Botanisk Tidskrift*, 54: 561-564.
- Field, J.; Paxton, R.J.; Soro, A. & Bridge, C. 2010. Cryptic Plasticity Underlies a Major Evolutionary Transition. *Current Biology*, 20: 2028-2031. <https://doi.org/10.1016/j.cub.2010.10.020>.
- Giraldo, C.; Rodríguez, A.; Chamorro, F.J.; Obregón, D.; Montoya, P.; Ramírez, N.; Solarte, V. & Nates-Parra, N. 2011. *Guía ilustrada de polen y plantas nativas visitadas por abejas – Cundinamarca, Boyacá, Santander, Sucre, Atlántico y Sierra Nevada de Santa Marta-Colombia*. Bogotá, Universidad Nacional de Colombia. 215p.
- Gonçalves, R.B. 2019. Phylogeny of the *Augochlora* clade with the description of four new species (Hymenoptera, Apoidea). *Revista Brasileira de Entomologia*, 63(1): 91-100. <https://doi.org/10.1016/j.rbe.2018.12.003>.
- Gonçalves, R.B. & Melo, G.A.R. 2010. Phylogeny of the bees subtribe *Caenohalictina* Michener (Hymenoptera, Apidae s.l., Halictinae s.l.). *Zoologica Scripta*, 39(2): 187-197. <https://doi.org/10.1111/j.1463-6409.2009.00414.x>.
- González, V.H.; Ospina, M. & Bennett, D.J. 2005. *Abejas Altoandinas de Colombia: guía de campo*. Bogotá, Instituto de Investigación de Recursos Biológicos Alexander von Humboldt. 57p.
- Jiménez-B., L.C. & Rangel-C.H., J.O. 1997. Atlas Palinológico de la Amazonia Colombiana II. Familia Asteraceae. *Caldasia*, 19(1-2): 29-39.
- LaBerge, W.E. & Ribble, D.W. 1966. The nests and larvae of two species of *Agapostemon* (Hymenoptera: Halictidae). *Journal of the Kansas Entomological Society*, 39(3): 467-472.
- Mercado-Gómez, J.D.; Jiménez-Bulla, L.C. & Sánchez-Montaña, L.R. 2011. Pollen of the Magnoliopsida in el volcán (Pamplona, Colombia) I: families Apiaceae, Asteraceae, Cunoniaceae, Ericaceae, Fabaceae and Gentianaceae. *Caldasia*, 33(2): 619-635.
- Michener, C.D. 1966. Interaction among workers from different colonies of sweat bees (Hymenoptera, Halictidae). *Animal Behaviour*, 14(1): 126-129. [https://doi.org/10.1016/S0003-3472\(66\)80020-9](https://doi.org/10.1016/S0003-3472(66)80020-9).
- Michener, C.D. 1974. *The social behavior of the bees: a comparative study*. Cambridge, Harvard University Press. 404p.
- Michener, C.D. 1977. Nest and seasonal cycle of *Neocorynura pubescens* in Colombia (Hymenoptera: Halictidae). *Revista de Biología Tropical*, 25(1): 39-42.
- Michener, C.D. 1979. New and little-known halictine bees from Colombia (Hymenoptera: Halictidae). *Journal of the Kansas Entomological Society*, 52(1): 180-208.
- Michener, C.D. 2007. *The Bees of the World*. Baltimore, The Johns Hopkins University Press. p. 363-364.
- Michener, C.D. & Kerfoot, W.B. 1967. Nests and social behavior of three species of *Pseudaugochloropsis* (Hymenoptera: Halictidae). *Journal of Kansas Entomological Society*, 40(2): 214-232.
- Michener, C.D. & Wille, A. 1961. The Bionomics of a Primitively Social Bee, *Lasioglossum inconspicuus*. *University Kansas Science Bulletin*, 42(11): 1123-1202.
- Michener, C.D.; Breed, M.D. & Bell, W.J. 1978. Seasonal cycles, nests, and social behavior of some Colombian halictine bees (Hymenoptera; Apoidea). *Revista de Biología Tropical*, 27(1): 13-34.
- Michener, C.D.; Cross, E.A.; Daly, H.V.; Rettenmeyer, C.W. & Wille, A. 1955. Additional Techniques for Studying the Behavior of Wild Bees. *Insectes sociaux*, 2(3): 237-246.
- Michener, C.D.; Lange, R.B.; Bigarella, J.J. & Salamuni, R. 1958. Fatores determinantes da distribuição de ninhos de abelhas em barrancos terrosos. *Dusenía*, 8(1): 1-24.
- Moure, J.S. 2012. *Augochlorini* Beebe, 1925. In: Moure, J.S.; Urban, D. & Melo, G.A.R. (Eds.). *Catalogue of Bees (Hymenoptera, Apoidea) in the Neotropical Region – online version*. <http://moure.cria.org.br/catalogue?id=88139>. Access: 13/03/2021.
- Moure, J.S. & Hurd, P.D. 1987. *An Annotated catalog of the Halictid bees of the Western Hemisphere (Hymenoptera: Halictidae)*. Washington, Smithsonian Institution. 405p.
- Moure, J.S.; Urban, D. & Melo, G.A.R. 2007. *Catalogue of Bees (Hymenoptera, Apoidea) in the Neotropical Region*. Curitiba, Sociedade Brasileira de Entomologia. 1058p.
- Nates-Parra, G.; Parra, A.; Rodríguez, A.; Baquero, P. & Vélez, D. 2006. Abejas silvestres (Hymenoptera: Apoidea) en ecosistemas urbanos: estudio en la ciudad de Bogotá y sus alrededores. *Revista Colombiana de Entomología*, 32(1): 77-84.
- Nye, W.P. 1980. Notes on the Biology of *Halictus (Halictus) farinosus* Smith (Hymenoptera: Halictidae). *US Department of Agriculture Science and Education Administration Agricultural Research Results*, 11: 1-29.
- Pesenko, Y.A. 1999. Phylogeny and classification of the family Halictidae Revised (Hymenoptera: Apoidea). *Journal of the Kansas Entomological Society*, 72(1): 104-123.
- Pesenko, Y.A. 2004. The phylogeny and classification of the tribe Halictini, with special reference to the *Halictus* genus-group (Hymenoptera: Halictidae). *Zoosystematica Rossica*, 13(1): 83-113.
- Purcell, J. 2011. Geographic patterns in the distribution of social systems in terrestrial arthropods. *Biological Reviews*, 86(2): 475-491. <https://doi.org/10.1111/j.1469-185X.2010.00156.x>.
- Roberts, R.B. 1969. Biology of the Bee genus *Agapostemon* (Hymenoptera: Halictidae). *The University of Kansas Science Bulletin*, 48(16): 689-719.
- Roberts, R.B. 1972. Revision of the Bee genus *Agapostemon* (Hymenoptera, Halictidae). *The University of Kansas Science Bulletin*, 49(9): 437-590.
- Roberts, R.B. 1973. Bees of Northwestern America: *Agapostemon* (Hymenoptera: Halictidae). *Technical Bulletin Agricultural Experiment Station Oregon State University*, 125: 1-23.
- Roberts, R.B. & Brooks, R.W. 1987. *Agapostemonine Bees of Mesoamerica* (Hymenoptera: Halictidae). *The University of Kansas Science Bulletin*, 53(7): 357-392.
- Roubik, D.W. 1992. *Ecology and natural history of tropical Bees*. Cambridge, Cambridge University Press. 526p.
- Sakagami, S.F. & Michener, C.D. 1962. *The nest architecture of the sweat bees (Halictinae): a comparative study of behavior*. Lawrence, University of Kansas Press. 135p.
- Sakagami, S.F. & Moure, J.S. 1967. Additional observations on the nesting habits of some Brazilian halictine bees (Hymenoptera, Apoidea). *Mushi*, 40(10): 119-138.
- Sánchez, A.C. & Lupo, L.C. 2009. Asteraceae de interés en la Melisopalínología. Bosque Montano de las Yungas (Jujuy – Argentina). *Boletín de la Sociedad Argentina de Botánica*, 44(1-2): 57-64.
- Silva, C.I.; Ballesteros, P.L.O.; Palmero, M.A.; Bauermann, S.G.; Evaldi, A.C.P. & Oliveira, P.E. 2010. *Catálogo Polínico: palinología aplicada em estudos de conservação de abelhas do gênero Xylocopa no Triângulo Mineiro*. Uberlândia, Universidade Federal de Uberlândia. 154p.
- Silveira, F.A.; Melo, G.A.R. & Almeida, E.A.B. 2002. *Abelhas brasileiras: sistemática e identificação*. Belo Horizonte, Authors' edition. 253p.
- Solomon, M. 1970. Asteraceae. In: Free, J.B. (Ed.). *Insect pollination of crops* Experimental Agriculture. London, Academic Press. p. 367-368.
- Zobel, M.U. & Paxon, R.J. 2007. Is big the best? Queen size, usurpation and nest closure in a primitively eusocial sweat bee (*Lasioglossum malachurum*). *Behavioral Ecology and Sociobiology*, 61(3): 435-447. <https://doi.org/10.1007/s00265-006-0271-4>.