



## RESEARCH ARTICLE - ANTS

## Studies on an Enigmatic *Blepharidatta* Wheeler Population (Hymenoptera: Formicidae) from the Brazilian Caatinga

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### Abstract

*Blepharidatta* is a rare Neotropical ant genus formed by predatory species whose small colonies nest in soil or leaf-litter. A population of *Blepharidatta* that presents affinities with *Blepharidatta conops* Kempf was found in the Caatinga biome, at the “Reserva Particular do Patrimônio Natural Serra das Almas” (RPPNSA), in Crateús (State of Ceará, Brazil). The aim of our study was to obtain data on the nest architecture, size and composition of colonies, foraging behavior, and female castes morphology for this newly found population, and to compare it with other *Blepharidatta* species, particularly with *B. conops*. The results show that *Blepharidatta* sp. and *B. conops* share key features of their biology such as their basic nest architecture, diet and foraging behavior, and the presence of a single ergatoid queen with a phragmotic head. However, marked differences were also found in head and mesosoma morphology of the queen, nest architecture, colony size, and queen location in the nest. Two alternative hypotheses are presented. The newly found *Blepharidatta* population represents a new species, possibly endemic to the Caatinga biome or it represents an extreme of the phenotypic variations observed among the populations forming *B. conops*.

### Introduction

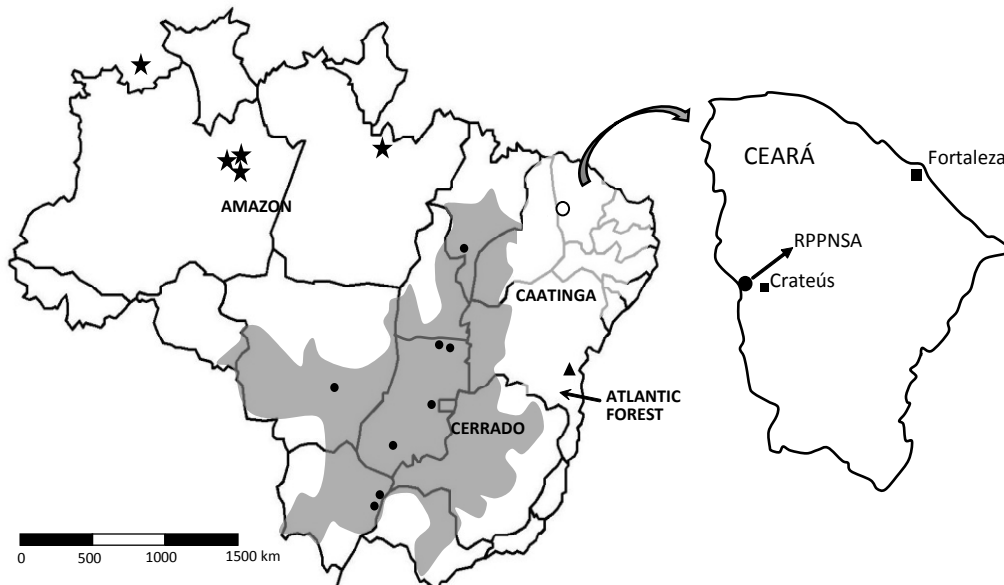
The myrmicine ant genus *Blepharidatta* is a strictly Neotropical group that was described by Wheeler (1915) from workers of *Blepharidatta brasiliensis* Wheeler collected near Belém (State of Pará, Brazil), in the Amazon Forest. Together with the genus *Wasmannia*, it forms the monophyletic tribe *Blepharidattini* (Wheeler & Wheeler, 1991; Bolton, 1995), which is considered to be a close relative of the fungus-growing ant tribe *Attini* (Schultz & Meier, 1995).

Up to seven species are currently recognized (Silva, 2007), but most of them are waiting for a formal taxonomic treatment or confirmation. Based on morphological as well as behavioral data, only three species are formally recognized: *B. brasiliensis* found in the Amazonian forest, *Blepharidatta conops* Kempf, an inhabitant of savanna-like formation from central Brazil (Cerrado), and an undescribed species (*Blepharidatta* sp-ba, hereafter) known from the Atlantic rainforest

of the State of Bahia, eastern Brazil (Rabeling et al., 2006; Brandão et al., 2008; Cassano et al., 2009) (Fig 1). All are ground-dwelling predatory species that nest in the ground or in the leaf-litter, with small monogynous or polygynous colonies (Brandão et al., 2001; Rabeling et al., 2006; Silva, 2007). Queens are ergatoid (*sensu* Peeters, 1991, i.e. permanently wingless and worker-like) and, at least in *B. conops*, it is believed that the foundation of new colonies is by fission of established colonies (Brandão et al., 2001).

The best studied species is *B. conops* (Diniz & Brandão, 1997; Diniz et al., 1998; Brandão et al., 2001, 2008). *B. conops* colonies (up to 250 workers) live in simple nests with a single opening and a vertical tunnel with 2 cm of diameter and 20 cm deep that ends in a cone-shaped widening at the bottom. Furthermore, all mature nests have a horizontal subsidiary chamber connected to the vertical tunnel through a narrow tunnel that opens at the mid-length of the vertical tunnel. It serves as a refuge for queen and brood when





**Fig. 1.** Distribution of known *Blepharidatta* populations and study site location in the state of Ceará (Brazil) (RPPNSA: Reserva Particular do Patrimônio Natural Serra das Almas). (★) *Blepharidatta brasiliensis*; (●) *Blepharidatta conops*; (▲) *Blepharidatta* sp-ba; ○ studied *Blepharidatta* new population

ests are visited or inhabited by myrmecophiles or predators. Queens of *B. conops* have a characteristic phragmotic head that together with the anterior slope of pronotum forms a frontal disk whose shape and dimensions fit the subsidiary chamber entrance and is used by the queen to block that entrance (Brandão et al., 2001). The foragers patrol a roughly circular area around the nest opening, where they collect live and dead arthropods (mainly other ants) to feed their larvae (Diniz & Brandão, 1997). In the bottom area of the nest, prey is dismembered, chewed and fed to larvae via trophallaxis. The discarded remains are arranged in a ring around the nest opening and workers concealed in the nest entrance ambush arthropods, including other ants, attracted by the carcasses ring (Diniz et al., 1998).

The other known species of *Blepharidatta* have different nesting habits: their smaller colonies (mean number of workers varying from 112 to 132) are found within the leaf-litter, in natural cavities between leaves or in rotting branches (Rabeling et al., 2006; Silva, 2007). Contrary to *B. conops*, the queens of *B. brasiliensis* and *Blepharidatta* sp-ba do not have a phragmotic head (Silva, 2007).

Colonies of *B. conops* occur in locally dense but widely scattered populations, with large areas of Cerrado devoided of *B. conops* (Diniz & Brandão, 1997; Brandão et al., 2001). According to Brandão et al. (2001), this distribution pattern may be explained by the limited dispersal mode of ergatoid queens and by the type of nest foundation (fission of established colonies). Such nest distribution may be characteristic of all *Blepharidatta* species and probably explains why ant species of this genus are considered rare (Brandão et al., 2001).

Here we present data on nest architecture, size and com-

position of colonies, foraging behavior, morphology (female castes) of a Caatinga population of an unidentified species of *Blepharidatta* (Quinet & Tavares, 2005). We compare our findings to the data available for other species of *Blepharidatta*, particularly *B. conops* and discuss the identity of this potential new taxon

## Material and methods

### Study site

The study was conducted from November 2011 to June 2012 in a 4.5ha area of the “Reserva Particular do Patrimônio Natural Serra das Almas - RPPNSA” (5°08’S, 40°51’W), a 6146ha protected area of deciduous thorny woodland vegetation (Caatinga) in Crateús (State of Ceará, northeastern Brazil, 5°10’S 40°40’W) (Fig 1).

### Nest architecture, size and composition of colonies

Twenty nine nests of *Blepharidatta* sp. were located in the study area, using freshly killed termites as baits. Any *Blepharidatta* sp. worker that picked a termite was then followed back to its nest.

Nineteen nests were excavated to study nest architecture as well as the size and composition of colonies. Before initiating a nest excavation, the maximum diameter of the nest opening and of the carcasses ring around it was recorded (see Diniz & Brandão, 1997). The carcasses forming the ring were collected, and a 30-cm deep trench was dug in order to obtain a 20-cm side cube with the nest opening in the middle of the

upper face. Starting from one lateral side, the cube was sliced with a spatula until a nest chamber or a tunnel was found. The depth, maximum height and maximum diameter of each chamber were recorded, as well as the direction, diameter and length of each tunnel leading to a chamber.

All biological material (workers, queen(s), male(s) and brood; invertebrate and vegetal fragments; myrmecophiles) found in chambers or tunnels was collected, and, whenever possible, its exact location in the nest was recorded. Workers, queen(s), male(s), brood and myrmecophile organisms found in each nest were counted and fixed in ethanol 90%.

#### Diet and foraging behavior

In order to obtain information on *Blepharidatta* sp. diet, all invertebrate and vegetal fragments found in the carcasses ring and chambers of 10 excavated nests were analyzed. Fragments were first separated in three categories: ants, other invertebrates and seeds. Ant fragments were identified at least to genus level. Fragments of other invertebrates were identified to order level.

The diel foraging activity pattern was investigated by monitoring three colonies for a 24h period: the first from 12/16/2011 (10 a.m.) to 12/17/2011 (9 a.m.), the two others from 06/16/2012 (9 a.m.) to 06/17/2012 (8 a.m.). The nest opening of each colony was observed for 10min every hour and all ants that left or entered the nest were counted. In total, four hours of foraging activity were monitored per nest. Soil surface temperature was recorded each time ant activity was measured.

The density and foraging area size of *Blepharidatta* sp. nests were assessed using a 144 m<sup>2</sup> area (12 x 12 m) with a grid of 1m<sup>2</sup> quadrats (Fig 6). Each quadrat was baited with sardine and checked 40 min later. Baiting was repeated three times in three successive days. Each quadrat whose bait was visited by *Blepharidatta* sp. workers at least one time was then baited four times, on three consecutive days, with dead workers (or soldiers) of *Nasutitermes* sp. All workers carrying termites back to the nest were followed and their path marked with pieces of plastic straw.

#### Body measurements

The maximum transverse diameter of the frontal disk (phragmotic head plus anterior slope of the pronotum) of 10 *Blepharidatta* sp. queens (one per nest) and the maximum head width of 54 workers (from 18 nests) were measured under a stereomicroscope with an ocular micrometer. A Petri dish filled with fine white sand was used to correctly position the ants under a microscope.

Total body length of 10 queens (from the foremost part of the frontal disk to the tip of gaster, in individuals with outstretched body) and 20 workers (from the middle part of the clypeus to the tip of the gaster, in individuals with outstretched body) were also measured.

Voucher specimens of *Blepharidatta* sp. are deposited at the Myrmecological Collection of the Laboratório de Entomologia, Universidade Estadual do Ceará, in Fortaleza, CE, Brazil, at the Myrmecological Collection of the Museu de Zoologia of the Universidade de São Paulo [MZSP] in São Paulo, SP, Brazil, and at the Myrmecological Collection of the Centro de Pesquisas do Cacau [CPDC], CEPLAC, in Itabuna, BA, Brazil.

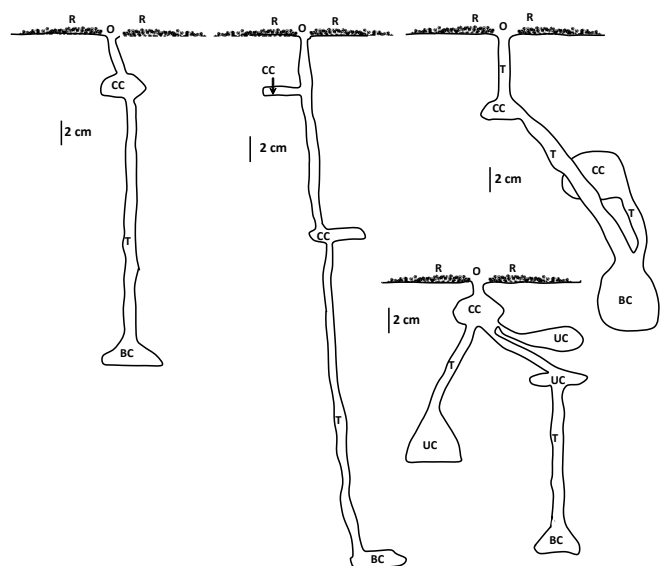
## Results

#### Nest architecture, size and composition of colonies

All excavated nests (n=19) had a carcasses ring that surrounded the nest opening, with invertebrate fragments sometimes partially obstructing it. In three nests, however, the carcasses ring was inconspicuous, with very few invertebrate or vegetal fragments. Carcasses rings were circular or elliptic, with maximum diameter varying from 5 to 20cm. Mean fragment density in the carcasses ring was 8 ( $\pm$  6.3) fragments/cm<sup>2</sup> (n=10; range = 1.7-19.1 fragments/cm<sup>2</sup>).

All nests had only one circular (sometimes elliptic) nest opening whose mean diameter was 0.73 ( $\pm$  0.17) cm (n=14; range: 0.5-1 cm). Each nest entrance was connected to a tunnel with approximately the same mean diameter (0.74  $\pm$  0.23 cm; n=15; range = 0.5–1.3 cm), that led to a first chamber (carcasses chamber), and ended in a bottom chamber (Fig 2). In six nests, the tunnel was a straight and vertical structure connecting the nest opening to the carcasses and the bottom chambers (Fig 2). In other seven nests, the tunnel had sloped parts, with, sometimes, a branching pattern (Fig 2).

The bottom chamber was located at a mean depth of 26.5 ( $\pm$  6.3) cm (n=19; range = 15-40 cm). It housed the



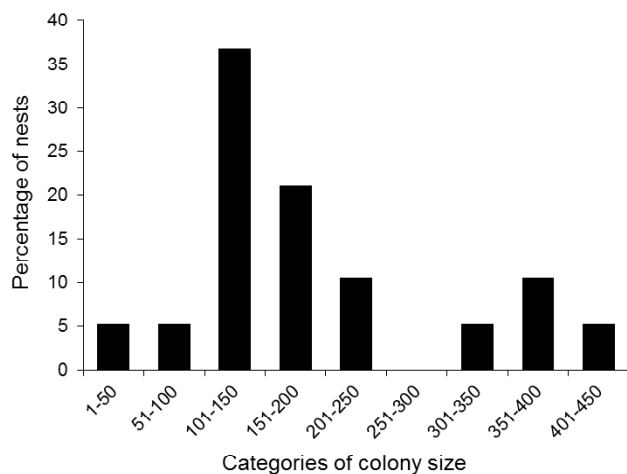
**Fig 2.** Schematic drawing of four *Blepharidatta* sp. nests excavated from 11/2011 to 06/2012, at the “Reserva Particular do Patrimônio Natural Serra das Almas”, CE, Brazil. R: carcasses ring; O: nest opening; T: tunnel; CC: carcasses chamber; BC: bottom chamber; UC: undefined chamber.

queen and brood in most nests ( $n=8$ ) where a queen was found ( $n=12$ ). Its mean maximum height and diameter were  $2.3 (\pm 1.54)$  cm ( $n=12$ ; range: 0.7-2.5 cm) and  $3.3 (\pm 1.07)$  cm ( $n=12$ ; range: 2-4 cm) respectively.

A carcasses chamber, so called because it was full of carcasses, was found in most (14 out 19) nests. It was located very close to the nest opening (Fig 2), at a mean depth of 4.9 ( $\pm 2.4$ ) cm ( $n=14$ , range: 1-11 cm), and its mean maximum height and diameter were  $1.9 (\pm 0.95)$  cm ( $n=12$ ; range: 0.5-2.5 cm) and  $2.8 (\pm 0.8)$  cm ( $n=12$ ; range: 2-4 cm), respectively. In six nests, one to two additional carcasses chambers were found at a depth varying from 10 to 25 cm (Fig 2). In one nest, up to five chambers were found (a carcasses chamber, a bottom chamber and three extra chambers), in addition to branching tunnels (Fig 2).

A possible subsidiary chamber, as defined by Brandão et al. (2001) for *B. conops*, was found in one nest. This chamber was located at the mid-length of the vertical tunnel leading to the bottom chamber, at a depth of 13 cm, and contained a queen, brood and 15 males. Queens were missing in seven nests, possibly because they escaped during the excavation process. All other nests ( $n=12$ ) had a single queen, except one that had two queens. Most queens (8 out 13) were found in the bottom chamber together with brood. In three remaining nests, the queen was found in the carcasses chamber ( $n=2$ ) or in the subsidiary chamber (*sensu* Brandão et al., 2001) ( $n=1$ ). Two queens could not be assigned to a specific nest location. A total of twenty five males were found, in chambers of four nests, together with the queen and brood. Fifteen of them were found in the subsidiary chamber of one nest; in the three other nests, from one to five males were found in the bottom chamber.

Three myrmecophiles were frequently found in *Blepharidatta* sp. nests, almost always in the bottom chamber: two species of the crustacean genus *Trichorhina* Budde-Lund (Oniscidea, Platyarthridae) and one cockroach species (Corydiidae); we also noticed the frequent occurrence of the pseudoscorpion *Petterchernes brasiliensis* Heurtault (Cher-



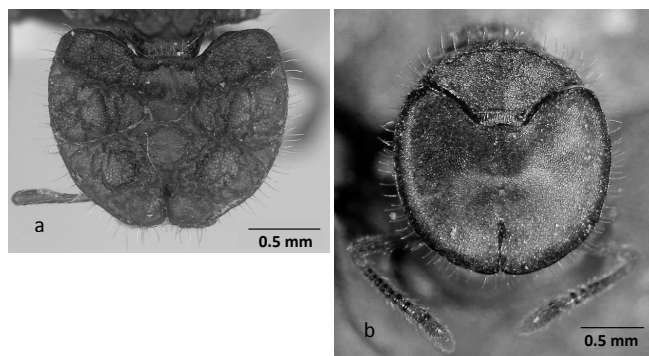
**Fig 3.** Distribution (%) of colony size categories for 19 nests of *Blepharidatta* sp. excavated from 11/2011 to 06/2012, at the “Reserva Particular do Patrimônio Natural Serra das Almas”, CE, Brazil.

netidae), up to now recorded only from burrows of small mammals in the state of Pernambuco, Brazil (Heurtault, 1986).

Average colony size (number of workers) was  $193 (\pm 107.4)$  ( $n=19$ ; range: 30-437). Most colonies (13 out 19) ranged from 110 and 220 workers; a few ( $n=4$ ) reached more than 300 workers (one of them had more than 400 workers) (Fig 3).

#### Queen and worker morphology

*Blepharidatta* sp. queens are ergatoid and have an enlarged phragmotic head that, together with the anterior slope of the pronotum, form a nearly circular disk (Fig 4) whose mean maximum transverse diameter is  $1.68 (\pm 0.04)$  mm ( $n=10$ ; range: 1.60-1.76 mm) (Table 1). Frontal disk cuticle is discretely reticulate-punctate, nearly smooth; head margin is strongly curved upwards, and stiff hairs protrude laterally from the perimeter of the disk (Fig 4). Total mean body length of queens is  $4.96 (\pm 0.12)$  mm ( $n=10$ ; range: 4.8-5.20 mm). *Blepharidatta* sp. workers are monomorphic (mean body length:  $3.79 \pm 0.22$  mm;  $n=20$ ; range: 3.50-4.48 mm), and their mean maximum head width is  $0.96 (\pm 0.05)$  mm ( $n=54$ ; range: 0.84-1.04 mm) (Table 1).



**Fig 4.** Phragmotic head of *Blepharidatta conops* (a) (photo by April Nobile – [www.antweb.org](http://www.antweb.org)) and *Blepharidatta* sp. (b) queen.

**Table 1.** Mean maximum transverse diameter (mm) of queen (Q) frontal disk and mean workers (W) maximum head width (mm) in different *Blepharidatta conops* populations (SR, BA, SEL, CH-G) and in the *Blepharidatta* population found at the “Reserva Particular do Patrimônio Natural da Serra das Almas” (RPPNSA), in Crateús (State of Ceará). SR: Serranópolis (State of Goiás); BA: Balsas (State of Maranhão); SEL: Selvíria (State of Mato Grosso do Sul); CH-G: Chapada dos Guimarães (State of Mato Grosso). Data within parenthesis refer to standard errors of the means. Data source for *B. conops*: Brandão et al. (2001).

	<i>Blepharidatta conops</i>				<i>Blepharidatta</i> sp.
	SR	BA	SEL	CH-G	RPPNSA
Q	1.55 ( $\pm 0.03$ ) $n=12$	1.2 ( $\pm 0.06$ ) $n=5$	1.45 $n=1$	1.51 $n=1$	1.68 ( $\pm 0.04$ ) $n=10$
W	0.87 ( $\pm 0.04$ ) $n=5$	0.75 ( $\pm 0.05$ ) $n=5$	0.78 $n=1$	---	0.96 ( $\pm 0.05$ ) $n=54$



### Diet and foraging behavior

In total, 13,576 fragments were found in the carcasses ring and chambers of 10 *Blepharidatta* sp. nests. Of these, 8,385 (62%) were from ants, 4,660 (34%) from others invertebrates, and 522 (4%) were whole seeds or pieces of seeds. Most invertebrate and seed items were found in the carcasses ring (77% and 85% respectively); the rest was found in nest chambers, mostly in carcasses chambers. Most ant fragments (93%) could be identified; 61% of the other invertebrate fragments and none of the seeds could be identified.

Ant carcasses included 41 species from 18 (or 19) genera belonging to seven subfamilies (Table 2). Three species accounted for 50% of all ant carcasses. The most common species was *Ectatomma muticum* Mayr (Ectatomminae) (23%), followed by *Camponotus crassus* Mayr (Formicinae) (16%) and *Labidus* sp. nr. *coecus* (Ecitoninae) (11%).

Other eight species (*Acromyrmex rugosus* (Smith), *Gnamptogenys striatula* Mayr, *Camponotus* sp., *Cephalotes pusillus* (Klug), *Eciton* sp., *Crematogaster* sp., *Pheidole* sp., *Odontomachus bauri* Emery) were frequent, accounting each for 3 to 7% of all ant carcasses. It is also worth mentioning the Ecitoninae group since 18% of all ant carcasses were from *Eciton* (3 species), *Labidus* or *Neivamyrmex* (4 species), *Nomamyrmex* (1 species) and an unidentified genus (1 species).

Other invertebrate carcasses were from eight insect groups (Blattodea [including termites], Coleoptera, Dermaptera, Hemiptera, Hymenoptera, Lepidoptera, Neuroptera, Orthoptera), and two arachnid groups (Araneae, Scorpiones). However, the vast majority (92%) of carcasses were from four groups: Hemiptera (37.5%), Coleoptera (22.4%), Araneae (19.4%) and termites (13%) (all termite fragments were heads of *Nasutitermes* sp. soldiers).

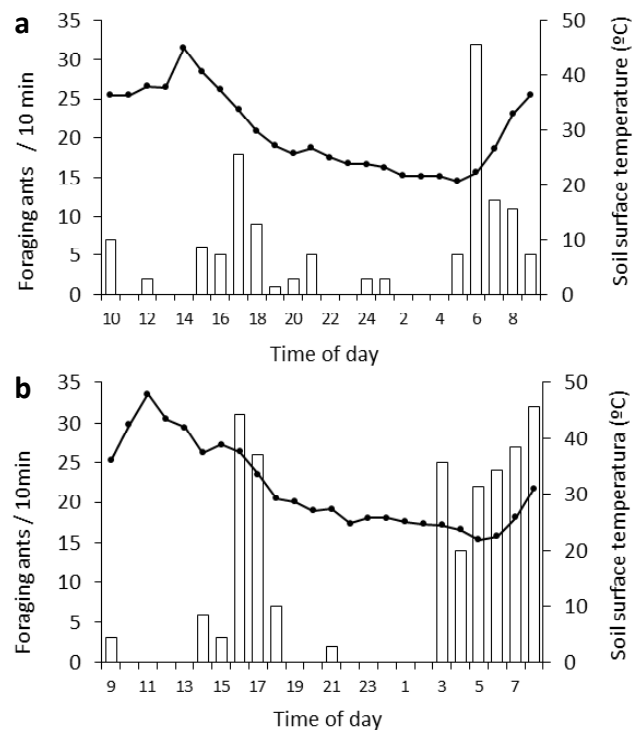
**Table 2.** List of subfamilies and genera identified in ant carcasses found in the carcasses ring and nest chambers of 10 *Blepharidatta* sp. nests, with number of species found in each genus.

Subfamily	Genus	Species number
Dolichoderinae	<i>Dolichoderus</i>	1
	<i>Dorymyrmex</i>	1
Ectatomminae	<i>Ectatomma</i>	1
	<i>Gnamptogenys</i>	1
Ecitoninae	<i>Eciton</i>	3
	<i>Labidus/Neivamyrmex</i>	4
	<i>Nomamyrmex</i>	1
	unidentified genus	1
Formicinae	<i>Brachymyrmex</i>	1
	<i>Camponotus</i>	5
Myrmicinae	<i>Acromyrmex</i>	1
	<i>Cephalotes</i>	1
	<i>Crematogaster</i>	4
	<i>Cyphomyrmex</i>	1
	<i>Pheidole</i>	10
	<i>Solenopsis</i>	2
	<i>Strumigenys</i>	1
Ponerinae	<i>Odontomachus</i>	1
Pseudomyrmecinae	<i>Pseudomyrmex</i>	1

Foraging activities of *Blepharidatta* sp. are predominantly crepuscular, with two peaks of activity, the first one corresponding to the night/day transition period (5 a.m.-9 a.m.), the second one to the day/night transition period (4 p.m.-7 p.m.) (Fig 5). At other periods, especially during the warmest period of the day ( $\pm$  10 a.m.-3 p.m.), foraging activity stops almost entirely, at least at the nest opening (Fig 5). However, at night, from 7 p.m. to 2 a.m., intense cleaning activity was observed in all nests, with workers carrying carcasses or sand particles out of the nest and depositing them on the carcasses ring, while others removed carcasses obstructing the nest opening and organized them on the carcasses ring.

During the period when nests foraging activity was monitored (up to 12 hours of nest opening observation for the three nests), it was observed that the carcasses ring was frequently visited by invertebrates, including by ants that robbed carcasses and sometimes inspected *Blepharidatta* sp. nest opening. However, none of these visitors was ambushed by *Blepharidatta* sp. workers, contrary to what was observed in *B. conops*, whose workers, concealed inside the nest entrance, frequently ambush visitors (Diniz et al., 1998).

Collective transport of large prey items was observed in *Blepharidatta* sp. in one occasion. More than 12 workers



**Figure 5.** Daily cycle of activity of two *Blepharidatta* sp. colonies (a, b) at the “Reserva Particular do Patrimônio Natural Serra das Almas”, CE, Brazil. All ants exiting/entering the nest were counted during 10 minutes, during a 24-hour observation period (a: 16.12.2011; b: 16.06.2012). Bars: foraging ants/10 min; line with dots: soil surface temperature. Sunrise: 5.30 h (onset of daylight: 6.00 h); sunset: 17.30 h (beginning of darkness: 18.00 h). The daily cycle of activity of the third monitored nest (16.06-17.06.2012), not shown here, had similar pattern.

were observed grasping and pulling the appendages of two still alive and fighting *Ectatomma muticum* workers, trying to transport them to the nest.

Only two nests were found in the 144 m<sup>2</sup> area mapped (three if a nest close to the area border is included) (Fig 6), giving a nest density of 0.020 nest/m<sup>2</sup>. Although some foragers were observed foraging at a distance of seven meters from the nest, the most common foraging distances observed were two to three meters (Fig 6). *Blepharidatta* sp. nest foraging area can therefore be described as a nearly circular area with a radius of  $\pm 2.5$ m around the nest opening.

## Discussion

There is no doubt that *B. conops* and *Blepharidatta* sp. belong to a group of closely related taxa since they share key features of their behavior, ecology and morphology, while at the same time both show fundamental differences with the other *Blepharidatta* species living in tropical humid environments.

*B. conops* and *Blepharidatta* sp. are soil-dwelling species with nest opening surrounded by a carcasses ring (Diniz et al., 1998), while *B. brasiliensis* and *Blepharidatta* sp-ba nest in leaf-litter, and do not form carcasses ring (Rabeling et al., 2006; Silva, 2007). *B. conops* and *Blepharidatta* sp. are monogynous species whose ergatoid queens have a phragmotic head (Brandão et al., 2001), while *B. brasiliensis* and *Blepharidatta* sp-ba have polygynous colonies, and queens do not have a phragmotic head (Rabeling et al., 2006; Silva, 2007). It is worth mentioning, however, that polygyny or mo-

nogyny of *Blepharidatta* sp-ba remains a controversial question (Silva, 2007).

Frontal disk diameter in queens, head width in workers and body length in queens and workers are almost identical in both species (Table 1) (Diniz et al., 1998; Silva, 2007). On the other hand, body length in queens and workers of *B. conops* and *Blepharidatta* sp. ( $\pm 5$  mm for queens;  $\pm 3$  mm for workers) is almost three times that observed in *B. brasiliensis* and *Blepharidatta* sp-ba ( $\pm 2$ mm and  $\pm 1$ mm for queens and workers, respectively) (Silva, 2007).

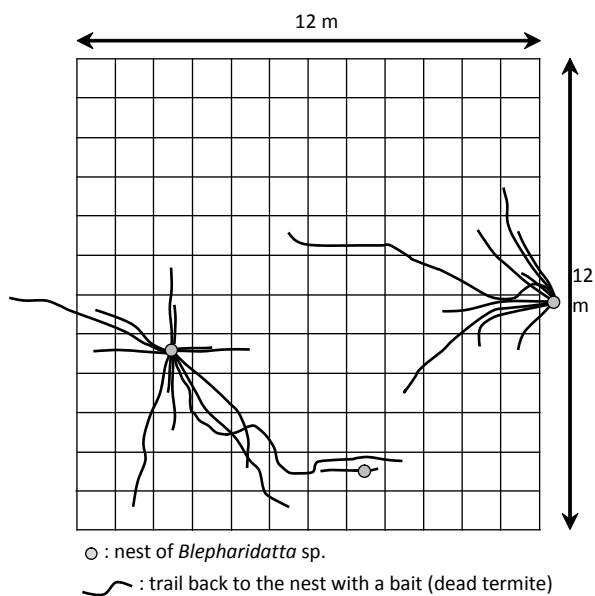
*Blepharidatta* sp. and *B. conops* have similar diet and foraging activity patterns, with two daily and mostly diurnal peaks of foraging activity separated by periods of inactivity, and a protein diet predominantly made of other ant species (Diniz et al., 1998; Brandão et al., 2008). In *B. brasiliensis*, foraging activity is predominantly nocturnal (Rabeling et al., 2006).

Finally, the two groups of populations are found in completely different environments: rainforests (Amazon and Atlantic Rainforest) for *B. brasiliensis* and *Blepharidatta* sp-ba (Rabeling et al., 2006; Silva, 2007); drier and savanna-like environments (Cerrado and Caatinga) for *B. conops* and *Blepharidatta* sp. (Diniz et al., 1998; Brandão et al., 2001).

According to Silva (2007), *B. conops* possesses more derived character states (e.g. phragmotic head of queens) when compared to other *Blepharidatta* species. Considering the number of key behavioral and morphological features shared by *B. conops* and *Blepharidatta* sp., it can be stated that *Blepharidatta* sp. shares with *B. conops* this apomorphic condition.

However, our study also shows the existence of many differences between the populations living in the Cerrado biome (*B. conops*) and the population found in the Caatinga biome (*Blepharidatta* sp.). The most noticeable are those related to queen morphology. In *Blepharidatta* sp., frontal disk cuticle is discretely reticulate-punctate, nearly smooth, while it is rugose with a strong relief composed of closely packed polygonal units delimited by ridges in *B. conops* (Brandão et al., 2001) (Fig 4). Furthermore, other morphological differences, not detailed above, can be seen in the mesosoma sculpture, much stronger in the *B. conops* queen than in *Blepharidatta* sp., in the humeral angle of the pronotum which is sharply angulated in *Blepharidatta* sp. but rounded in *B. conops*. Finally, the pilosity on the first gastral tergite of the queen is erect and short in *Blepharidatta* sp. while it is subdecumbent and a little longer in *B. conops*.

*Blepharidatta* sp. and *B. conops* also differ in relation to nest architecture, colony size, location of queens and brood in nests and foraging behavior. *Blepharidatta* sp. nests are deeper ( $\pm 26.5$  cm; up to 40 cm in some nests) than in *B. conops* (up to 20 cm in mature nests) (Diniz et al., 1998). Conversely, the mean diameter of tunnels and nest opening is much greater in *B. conops* (2cm versus 0.7cm in *Blepharidatta* sp.) (Brandão et al., 2001). In most excavated nests of *Blepharidatta* sp.,



**Fig 6.** Mapping of *Blepharidatta* sp. nests and of the trails used by foragers to go back to the nest with a bait (dead termite) in a 144 m<sup>2</sup> (12 x 12 m) area with a grid of 1 m<sup>2</sup> quadrats, at the “Reserva Particular do Patrimônio Natural Serra das Almas”, CE, Brazil, in 01/2012.

there was one, or more, chambers located close to the nest opening and full of carcasses. The function of those carcasses chambers in *Blepharidatta* sp. is probably to store prey carcasses before discarding them on the carcasses ring. No such chamber was found in *B. conops* nests. One of the most important differences in nest architecture between *B. conops* and *Blepharidatta* sp. is the so-called subsidiary chamber. In *B. conops*, all mature nests have a subsidiary chambers used as refuge by the queen and the brood (Diniz et al., 1998; Brandão et al., 2001). Only one *Blepharidatta* sp. nest had a chamber full with brood, queen and males and whose features suggest a subsidiary chamber.

More generally, *Blepharidatta* sp. nests are more variable in structure than *B. conops* nests. In *B. conops* nests, there is only one tunnel, always vertical (Diniz et al., 1998). In *Blepharidatta* sp., the main tunnel varies from vertical to inclined. Furthermore, the main tunnel can have branchings, the result being nests three-dimensionally more complex than those of *B. conops*. It cannot be excluded that the characteristics of the soil where the nests of *Blepharidatta* sp. were built have a strong influence on nest architecture. However, such characteristics were not analyzed in the present study, nor in studies with *B. conops* (Diniz et al., 1998).

As in other *Blepharidatta* ants, *Blepharidatta* sp. colonies are small. However, the average number of workers in *Blepharidatta* sp. colonies ( $193 \pm 107$ ,  $n=19$ ) is higher than in *B. conops* ( $142 \pm 57$ ,  $n=19$ ) (Diniz et al., 1998), *B. brasiliensis* ( $132 \pm 96$ ,  $n=13$ ) (Rabeling et al., 2006) and *Blepharidatta* sp. ba ( $112 \pm 13$ ,  $n=2$ ) (Silva, 2007). One of the *Blepharidatta* sp. nests had up to 437 workers. In *B. conops*, the largest known colony size is 248 workers (Diniz et al., 1998). Interestingly, two queens were found in a single *Blepharidatta* sp. nest. This observation supports the hypothesis that in *B. conops*, the foundation of new colonies is through fission of established colonies (Brandão et al., 2001). If this hypothesis is correct, there must be a time when two queens are present in the nest: the old resident queen and a young virgin queen.

The location of queen and brood in nests is another difference between *B. conops* and *Blepharidatta* sp. In most nests of *Blepharidatta* sp., the queen and brood were found in the bottom chamber. In *B. conops*, they are generally found in the subsidiary chamber, but never in the bottom chamber of the nest, which is used as a place for prey dismembering and to temporarily store carcasses, before they are taken out of the nest (Diniz et al., 1998; Brandão et al., 2001).

Foraging behavior of *Blepharidatta* sp. and *B. conops* is very similar. However, some differences were observed. According to Diniz et al. (1998), the ants and other arthropods visiting the carcasses ring of nests “are frequently ambushed by *B. conops* workers concealed immediately inside the nest entrance”. Such behavior was never observed in *Blepharidatta* sp. Another significant difference is the size of foraging area. In *Blepharidatta* sp. the mean radius of foraging area (2.5 m) is nearly twice that observed in *B. conops* (1.5 m), while nest

density seems to be much lower in *Blepharidatta* sp. ( $0.02$  nests/m<sup>2</sup> against  $0.2$  nests/m<sup>2</sup> in *B. conops*), at least in the investigated area (Diniz et al., 1998). However, the influence of local factors on nest density, such as prey density or competition with other ant species that use similar resources can not be excluded.

Although it cannot be excluded that *Blepharidatta* sp. is a species different from *B. conops*, it is obvious that both taxa are closely related and constitute a single evolutionary unit, with a branch that spread into the Cerrado biome (savanna) and another into the Caatinga biome (semi-arid). The traits that strongly suggest the occurrence of two distinct species are seen in the queen morphology, mainly on the frontal disk and mesosoma. On the other hand, it has been suggested that *B. conops* distribution in the Cerrado biome is fragmented in a range of local populations, with size and sculpture of queen frontal disk varying from one population to the other (Brandão et al., 2001). Therefore, *Blepharidatta* sp. could be a local variation of *B. conops* adapted to semi-arid conditions and consequently represent one extreme of the phenotypic variations observed in populations of *B. conops*. Ant populations in species that have lost mating flight have low levels of gene flow and can evolve independently, as it could happen in fragmented populations of *B. conops* or its ancestor since the early Quaternary.

Future studies should use cytogenetic and molecular tools to compare these *Blepharidatta* populations (e.g. Mariano et al., 2008; Resende et al., 2010 with *Dinoponera lucida* Emery 1901).

Finally, our study extends the known geographic range of the *Blepharidatta* group and the range of biomes where it is found, as it is the first time that the genus, until now found only in wet forests and savannas (Cerrado), is reported for the Neotropical semi-arid region.

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