Sociobiology
An international journal on social insects

SHORT NOTE

# First Record of *Cyphoderus innominatus* Mills, 1938 (Collembola: Paronellidae) in Early Colonies of the Leaf-Cutting Ant *Atta sexdens*

Tarcísio Marcos M Mota Filho[1], Kátia Kaelly Andrade Sousa[1], Roberto S Camargo[1], João Victor Lemos Cavalcante Oliveira[2], Nadia Caldato[1], Douglas Zeppelini[2], Luiz Carlos Forti[1]

1 - Laboratory of Social Insects-Pests, Plant Protection Department, School of Agronomic Sciences, São Paulo State University, Botucatu-SP, Brazil
2 - Laboratory of Collembola Systematics and Conservation, Biology Department, Center for Applied Biological and Social Sciences, Paraiba State University, Campus V, João Pessoa-PB, Brazil

**Article History**

**Edited by**
Paulo Cristaldo, UFRPE, Brazil
Received 20 October 2020
Initial acceptance 28 December 2020
Final acceptance 23 February 2021
Publication date 20 May 2021

**Keywords**
Attini, *Cyphoderus*, Myrmecophily, Cyphoderinae.

**Corresponding author**
Tarcísio Marcos Macedo Mota Filho
Laboratory of Social Insects-Pests
Plant Protection Department
School of Agronomic Sciences
São Paulo State University
Botucatu, São Paulo, Brasil.
E-Mail: tarcisio972010@hotmail.com

**Abstract**
*Cyphoderus innominatus* Mills, 1938 (Collembola: Paronellidae) was first observed in early colonies of *Atta sexdens* leaf-cutting ants (Hymenoptera: Formicidae). The colonies were collected on February 6, 2019, from a transition area between the Atlantic Forest and the Cerrado, located in the municipality of Botucatu, São Paulo, Brazil. A total of four colonies collected had an average population density of 227 ± 212 *C. innominatus* individuals, and most of the latter were found in peripheral areas inside the fungus-garden-growing chamber of the colony. In addition, we observed a possible defensive behavior on the part of workers when *C. innominatus* individuals were present in the fungus garden chamber. Thus, this is the first record of *C. innominatus* living in association with early colonies of *A. sexdens*.

Leaf-cutting ants of the genus *Atta* Fabricius, 1805 (Hymenoptera: Formicidae) are eusocial insects found exclusively in the Neotropical region (Hölldobler & Wilson, 1990). These ants build stable and long-lasting nests that protect themselves against abiotic and biotic factors, besides growing the fungus *Leucocoprinus gongylophorus* Heim, 1957, on which they feed (Sudd, 1982; Hölldobler & Wilson, 1990; Rocha et al., 2020).

However, due to favorable conditions, like temperature and humidity, that these nests offer (Camargo et al., 2016), they also become favorable habitats for a multitude of inquilines, from invertebrates such as beetles, cockroaches, mites, crickets, dipterans and springtails (Waller & Moser, 1990; Dekoninck et al., 2007; Forti et al., 2007), to vertebrates such as frogs, lizards and snakes (Hölldobler & Wilson, 1990; Baer et al., 2009; Lima Barros et al., 2016). According to Hölldobler and Wilson (1990), these ant guests are commonly known as myrmecophiles and may be present in ant nests temporarily engaging in predation or commensalism, or may also depend on such nests for a while or throughout their life cycle. Information about their ecology, behavior, life cycles and dispersion is scarce for most of these myrmecophilous species because they are difficult to observe and can only be found when specific studies are conducted (Dekoninck et al., 2007).

Nonetheless, springtails are inquilines commonly reported and studied in nests of fungus-growing ants (Castaño-Meneses et al., 2017). Even a specific behavior known as jigging, performed by *Cyphomyrmex costatus* workers, Mann, 1922, and *Myrmicocrypta buenzlii* workers, Borgmeier, 1934, (Formicidae: Myrmicinae), has been reported as a defense strategy to keep springtails away from the fungus-garden-growing chamber (Weber, 1957, 1972; Kweskin, 2004).

In this study, we identify, quantify and report the first species of springtails of the genus *Cyphoderus* living in early colonies of *A. sexdens*. In addition, we provide information on a possible defensive behavior performed by workers when springtails are present in their fungus garden chamber.

The study design relied on sampling, six hundred colonies of *A. sexdens* (Hymenoptera: Formicidae), aged approximately four months old, were collected on February 6, 2019, in a transition area between the Atlantic Forest and the Cerrado, located in the municipality of Botucatu (22° 48' 23.5" S 48° 25' 52.8" W), São Paulo, Brazil.

In the laboratory, a high population density of springtails was observed in four of the 600 collected colonies. These colonies were separated, and then, with the aid of an aspirator, all springtails present were removed by suction and stored in 98% alcohol. Afterwards, population density was quantified, and samples were taken for species identification. The identification was carried out by the Laboratory of Collembola Systematics and Conservation of the State University of Paraíba (UEPB). The methodology adopted for identification consisted of clearing and mounting of the specimens, following the procedures described by Jordana et al. (1997), with clearing being done using Nesbitt's solution and a phase-contrast microscope (Zeiss Axioskop). Then, to identify the species, the classification key provided by Oliveira et al. (2017) was used. The specimens are deposited in the UEPB's Soil Fauna Reference Collection [*Coleção de Referência da Fauna de Solo*] (CRFS).

*Cyphoderus innominatus* Mills, 1938 (Collembola: Paronellidae) was the species of springtails found living in association with early colonies of *A. sexdens* (Fig 1). *Cyphoderus innominatus* were found in four (0.67%) of the 600 collected colonies, with an average population density of 227 ± 212 individuals (Table 1). Most of the individuals were present in peripheral zones inside the fungus garden chamber of the colony, where some debris accumulated, but the springtails did not make direct contact with the ants (Fig 1B and video: https://youtu.be/EObSZYkZs30).

The video showed that the springtails (https://youtu.be/EObSZYkZs30) were initially present in the fungus garden chamber of the ants, apparently without causing any disturbance. Until a medium worker (ant with head width of approximately 2.2 mm), between the 38$^{th}$ and 46$^{th}$ seconds, presented a defensive behavior against the springtails. With its jaws open, it jumped three consecutive times towards some springtails. However, the attacks were unsuccessful, and they managed to escape.

The present study recorded, for the first time, the occurrence of *C. innominatus* living in association with early colonies of *A. sexdens* leaf-cutting ants collected in a transition area between the Atlantic Forest and the Cerrado. Previously, in Paraguay, springtails of the genus *Cyphoderus* had been reported living in colonies of *A. sexdens* subjected to laboratory conditions (Fowler, 1981). However, despite the occurrence of this genus being recorded, no species was identified. In addition, three other species of springtails had already been found in association with fungus growing carried out by leaf-cutting ants of the genera *Att*a and *Acromyrmex*, namely: *Cyphoderus inaequalis* with *Acromyrmex octospinosus*; *Pseudosinella violenta* (Lepidocyrtidae) with *Atta texana*; and *Seira edmanni* (Seiridae) with *A. sexdens* (Eidmann, 1937; Weber, 1958; Kistner, 1982; Waller & Moser, 1990; Castaño-Meneses et al., 2017). The presence of springtails of the genus *Cyphoderus* in early colonies of fungus-growing ants can be explained by the large amount of food found in these colonies; being myrmecophilous species, they feed on the mycelium of the fungus grown by the ants (Kistner, 1982). Moreover, colonies of fungus-growing ants also indirectly provide favorable conditions, such as temperature, moisture and protection from predators, to enable the growth, development and multiplication of the tenant population (Hughes et al., 2008).

The average density of *C. innominatus* individuals found in the early colonies of *A. sexdens* was 227 ± 212 individuals. It is a relatively high density compared to the population density of workers (small, medium and large) from an early nest (up to 4 months) of *A. sexdens*, which, according to Camargo and Forti (2013), comprises approximately 121 individuals. According to Kweskin (2004), a high population density of springtails ends up consuming a large amount of the mycelium grown in the fungus garden, consequently disturbing the colony (Kweskin, 2004). Some behavioral acts performed by ants against these disturbances have been reported. Among said acts, jigging is a peculiar rhythmic swinging behavior, and there are two hypotheses as to why it is performed: alerting mates about disturbances, or expelling competitors (Weber, 1957;1972). However, this behavior was not observed in our study. On the other hand, the behavioral act also performed by ants in the presence of springtails in their fungus gardens, and which was observed in our study, is jumping, when ants jump and try to capture springtails with their jaws and legs. According to Kweskin (2004), this type of behavior seems to be exclusively meant for defense against invaders, and the same behavior was also performed by *C. costatus* workers when springtails approached their fungus garden. The actual reason and the efficiency of this behavioral act against these disturbances are still not well understood.

In light of our findings, future studies should be carried out to elucidate the effect of the presence of *C. innominatus* in fungus gardens of *A. sexdens*, and whether their presence causes disturbances and induces defensive behaviors. Thus, our study provides important information about a species of myrmecophilous springtails that live in association with early colonies of fungus-growing ants, since we reported here, for the first time, the occurrence of *C. innominatus* in early colonies of *A. sexdens*. Furthermore, we provided information about a possible defensive behavior performed by *A. sexdens* workers when springtails are present in their fungus garden chamber.

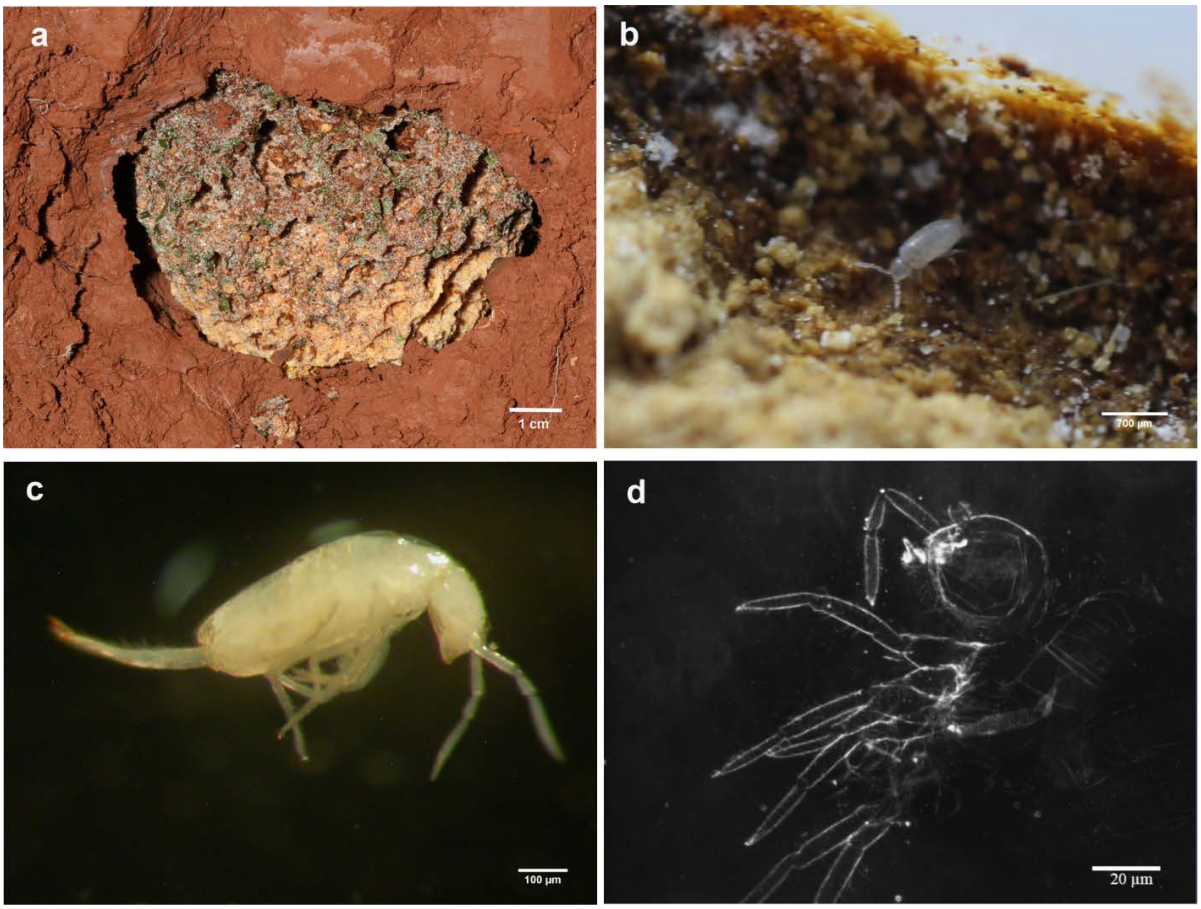


**Fig 1**. (a) Early nest of *Atta sexdens* (Hymenoptera: Formicidae); (b) *Cyphoderus innominatus* Mills, 1938 (Collembola: Paronellidae) present in peripheral zone inside the fungus-garden-growing chamber of *A. sexdens* fungus in laboratory condition [temperature of 22 ± 2 ºC, relative humidity of 70 ± 10% and photoperiod of 14:10 h (L: D)]; (c) Side view of *C. innominatus* in 98% alcohol; (d) Slide view with Zeiss Axioskop microscope.

**Table 1**. Number of individuals of *Cyphoderus innominatus* Mills, 1938 (Collembola: Paronellidae) found in early colonies of *Atta sexdens* (Hymenoptera: Formicidae).

| Colony | Number of individuals of *Cyphoderus innominatus* Mills, 1938 |
|---|---|
| 1 | 539 |
| 2 | 133 |
| 3 | 165 |
| 4 | 70 |
| Mean ± standard deviation | 227 ± 212 |

## Acknowledgments

TMM Mota Filho and KKA Sousa thank the support of the Coordination for the Improvement of Higher Education Personnel [Coordenação de Aperfeiçoamento de Pessoal de Nível Superior] – Brazil (CAPES) – Finance code 001. LC Forti gratefully acknowledges the support of the National Council for Scientific and Technological Development [Conselho Nacional de Desenvolvimento Científico e Tecnológico] (CNPq-PQ) (donation No. 301938/2017-2).

## Conflict of interest

The authors declare no conflicts of interest.

## Authors' contributions

TMM Mota Filho, KKA Sousa, RS Camargo, N Caldato and Forti L. C. Conceptualization, investigation, data curation, Writing. JVC Oliveira and D Zeppelini carried out the identification of the species.

## References

Baer, B., Den Boer, S.P.A., Kronauer, D.J.C., Nash, D.R. & Boomsma, J.J. (2009). Fungus gardens of the leafcutter ant *Atta colombica* function as egg nurseries for the snake *Leptodeira annulata*. Insectes sociaux, 56: 289-291. doi: 10.1007/s00040-009-0026-0.

Camargo, R.S. & Forti, L.C. (2013). Queen lipid content and nest growth in the leaf cutting ant (*Atta sexdens rubropilosa*) (Hymenoptera: Formicidae). Journal of Natural History, 47: 65-73. doi: 10.1080/00222933.2012.738836.

Camargo, R.S., Forti, L.C., Matos, C.A.O., Caldato, N. & Fonseca, O.S. (2016). Is the initial nest depth adapted to favorable conditions for the incipient colony in leaf-cutting ants? Sociobiology, 63: 792-799. doi: 10.13102/sociobiology.v63i2.976.

Castaño-Meneses, G., Palacios-Vargas, J.G., Delabie, J.H.C., Zeppelini, D. & Mariano, C.S.F. (2017). Springtails (Collembola) associated with nests of fungus-growing ants (Formicidae: Myrmicinae: Attini) in southern Bahia, Brazil. Florida Entomologist, 100: 740-742. doi: 10.1653/ 024.100.0421.

Dekoninck, W., Lock, K. & Janssens, F. (2007). Acceptance of two native myrmecophilous species, *Platyarthrus hoffmannseggii* (Isopoda: Oniscidea) and *Cyphoderus albinus* (Collembola: Cyphoderidae) by the introduced invasive garden ant *Lasius neglectus* (Hymenoptera: Formicidae) in Belgium. European Journal of Entomology, 104: 159-161. doi: 10.14411/eje.2007.023.

Eidmann, H. (1937). Die gäste und gastverhältnisse der blattschneiderameise *Atta sexdens* L. Zeitschrift für Morphologie und Ökologie der Tiere, 32: 391-462.

Forti, L.C., Camargo, R.S., Verza, S.S., Andrade, A.P.P., Fujihara, R.T. & Lopes, J.F. (2007). *Microdon tigrinus* Curran, 1940 (Diptera, Syrphidae): Populational fluctuation and specificity to the nest of *Acromyrmex coronatus* (Hymenoptera: Formicidae). Sociobiology, 50: 909-919.

Fowler, H.G. (1981). Behaviour of two myrmecophiles of Paraguayan leaf-cutting ants. Revista Chilena de Entomologia, 11: 69-72.

Hölldobler, B. & Wilson, E.O. (1990). The Ants. Cambridge: Harvard University Press, 732 p

Hughes, D.P., Pierce, N.E. & Boomsma, J.J. (2008). Social insect symbionts: evolution in homeostatic fortresses. Trends in Ecology and Evolution, 23: 672-677. doi: 10.1016/j.tree.2008.07.011.

Jordana, R., Arbea, J.I., Simón, C. & Luciáñez, M.J. (1997). Collembola Poduromorpha. Museo Nacional de Ciencias Naturales, Madrid. 807 p.

Kistner, D.H. (1982). The social insects bestiary. Social insects. In: H.R. Hermann (ed), Social Insects. Vol. 3. (p. 1-245) New York: Academic Press.

Kweskin, M.P. (2004). Jigging in the fungus-growing ant *Cyphomyrmex costatus*: a response to collembolan garden invaders?. Insectes sociaux, 51: 158-162. doi: 10.1007/s00040-003-0712-2.

Lima Barros, A., López-Lozano, J.L. & Lima, A.P. (2016). The frog *Lithodytes lineatus* (Anura: Leptodactylidae) uses chemical recognition to live in colonies of leaf-cutting ants of the genus *Atta* (Hymenoptera: Formicidae). Behavioral Ecology and Sociobiology, 70: 2195-2201. doi: 10.1007/s00265-016-2223-y.

Oliveira, J.V.L.C., Alves, J.L.S. & Zeppelini, D. (2017). Two new *Cyphoderus* (Collembola: Paronellidae) of" *tridenticulati*" and" *bidenticulati*" groups from Brazilian Amazon. Zootaxa, 4350: 47-60. doi: 10.11646/zootaxa.4350.1.2.

Rocha, F.H., Lachaud, J.P. & Pérez-Lachaud G. (2020). Myrmecophilous organisms associated with colonies of the ponerine ant *Neoponera villosa* (Hymenoptera: Formicidae) nesting in *Aechmea bracteata* bromeliads: A biodiversity hotspot. Myrmecological News, 30: 73-92. doi: 10.25849/myrmecol.news_030:073.

Sudd, J.H. (1982). Ants: foraging, nesting, brood behavior, and polyethism. In: H.R. Hermann (Ed.). Social Insects. Vol. 4. (p. 107-155). New York: Academic Press.

Waller, D.A. & Moser, J.C. (1990). Invertebrate enemies and nest associates of the leaf-cutting ant *Atta texana* (Buckley) (Formicidae, Attini). In: Meer, R.K.V., Jaffe, K. & Cedeno, A. (eds). Applied Myrmecology – A world perspective (p. 255-273). San Francisco: Westview Press.

Weber, N.A. (1957). Fungus-growing ants and their fungi: *Cyphomyrmex costatus*. Ecology, 38: 480-494. 10.2307/1929893.

Weber, N.A. (1958). Evolution in fungus-growing ants. In: Proceedings of the Tenth International Congress of Entomology, 2: 459-473.

Weber, N.A. (1972). Gardening Ants, the Attines. The American Philosophical Society, Philadelpha, Penn. 146 p.