

FIRST OCCURRENCE AND SEXUAL BEHAVIOR OF *Cryptontsira parva* (MUESEBECK, 1941) (HYMENOPTERA, BRACONIDAE) PARASITIZING *Lyctus brunneus* (STEPHENS, 1830) (COLEOPTERA, BOSTRICHIDAE) IN SOUTH AMERICA

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Abstract: The current study describes the first record in South America and the sexual behavior of *Cryptontsira parva* (Muesebeck, 1941) (Hymenoptera, Braconidae, Doryctinae) parasitizing *Lyctus brunneus* (Stephens, 1830) (Coleoptera, Bostrichidae, Lyctinae). We also include a brief report of the ethological reproductive aspects the *C. parva* under laboratory conditions by providing information to support research on biological control of *L. brunneus*.

Keywords: biological control; borer; parasitoid-host relationship; reproductive aspects; wasp.

Lyctus brunneus (Stephens, 1830) (Coleoptera, Bostrichidae) is a coleopteran that the larvae, after hatching, start to feed on wood until fully grown (Beesley 1956), which comes from the sapwood in the stem of angiosperm trees. *Lyctus brunneus* is one of the most destructive and invasive pests worldwide regarding wood and wood products due to this habit (Ide *et al.* 2016).

Lyctus brunneus is native to Malaysia and Japan, but it spreads to other regions due to wood– product trading (Yalçin *et al.* 2017). Currently, this insect is distributed worldwide, and can be found in Brazil, where it damages processed wood and wood products. Chemical products are used to reduce damages caused to infested wood or as preventive measure against them (Peters *et al.* 2002). However, researchers report the possibility of using parasitoid insects to control this insect (Yalçin *et al.* 2017). According to Querner (2015), it is necessary using biological control in museums to stop pests from spreading; there are successful cases in which parasitic wasps were used against several pests in museums, including borers that damage wooden furniture.

There is no information about the parasitoid fauna associated with *L. brunneus* in Brazil, as well as research focused on using biological control to reduce damages caused by this coleopteran. Awareness of the interaction and biological aspects of parasitic wasps and *L. brunneus* is relevant to promote further scientific research about its biological control.

Cryptontsira parva (Muesebeck, 1941) (Hymenoptera, Braconidae, Doryctinae), which Dinoderus originally parasitizes minutus (Fabricius, 1775) (Coleoptera, Bostrichidae), can have the potential to biologically control L. brunneus. Muesebeck (1941) showed that Cryptontsira parva (at that time referred as Doryctes parvus) parasitizes specimens belonging to genus Lyctus in Queensland, Australia. The current study shows the first registered in South America and describes the sexual behavior of C. parva parasitizing L. brunneus, and briefly characterizes some of its reproductive ethological aspects under laboratory conditions.

Cryptontsira parva was recorded as a parasitoid of the coleopteran *L. brunneus* in the Laboratório de Biodeterioração da Madeira (LBM) (22°45'30.9"S; 43°41'52.9"W; altitude 36 meters above sea level) of the Departamento de Produtos Florestais (DPF) at the Instituto de Florestas of Universidade Federal Rural do Rio de Janeiro (UFRRJ), campus Seropédica, Rio de Janeiro, Brazil.

Trees of *Clitoria fairchildiana* R.A. Howard (Angiospermae: Fabaceae) provided the wood used as oviposition site for adults and as food for larvae of host *L. brunneus*. Trees were part of the forest fragment at Campus Fiocruz Mata Atlântica, Fundação Oswaldo Cruz (CFMA/FIOCRUZ). Individuals from this forest species were cut based on cutting authorization n.002920, which was granted by Secretaria Municipal de Ambiente e Cultura do Rio de Janeiro (SMAC).

The wood was processed into test samples (29.0 cm x 5.0 cm x 2.5 cm) and naturally dried. Test samples were acclimatized at 12 % relative humidity (environment equilibrium humidity), stored in closed environment, and sheltered from outside weather. Some samples were expelling sawdust with consistency of "talcum powder" after 12-month storage; this outcome was later confirmed as the result of *L. brunneus* larvae feeding. The samples also presented holes in their external part, which were made by adult specimens of *L. brunneus*. The taxonomic identification of the coleopteran was conducted through the capture of adult specimens and the application of criteria by Liu & Geis (2019).

Twenty-eight test samples with evident presence of coleopteran action were selected and transferred to a rectangular glass vessel (60 cm x 30 cm x 30 cm) sealed with voile fabric. Samples were stored in environment under controlled temperature (27 ± 3 °C) and relative humidity (65 ± 10 %) in order to create a laboratorial *L. brunneus* population. Once this population was established, the conditions to produce *L. brunneus* generations were assured. There were several hymenopterans flying inside the vessel and on wood surface, 60 days later. Sample of that specimens were collected and stored in flasks filled with 70 % ethanol for taxonomic identification.

Ethological aspects regarding the reproduction of *C. parva* were registered in *L. brunneus* populations through observations *in situ* and through video recording couples during copulation and oviposition, using a Canon PowerShort A650 IS camera. Such events were observed in 10 couples of the sample population kept inside the vessel, for five days.

The hymenopteran specimens were sent to Departamento de Ecologia e Biologia Evolutiva of Universidade Federal de São Carlos in São Carlos, São Paulo state, Brazil, for identification.

Samples of the coleopteran host and hymenopteran parasitoids were dried, mounted in pin, and deposited at the Coleção Taxonômica do Departamento de Ecologia e Biologia Evolutiva, Universidade Federal de São Carlos (DCBU) (referred to DCBU-419891 to DCBU-419906, females, and DCBU-419907 to DCBU-419918, males). A total of 28 parasitoids (12 males and 16 females) collected inside the glass vessel where the L. brunneus population was kept on C. fairchildiana wood samples were identified as Cryptontsira parva. The specimens were identified based on Belokobyskij (2008) and by comparing with the figures of the holotype (Figure 1). The holotype is deposited at United States National Museum (USNM), in Washington (referred to as n. 55678).

Cryptontsira parva adult specimens were found on wood samples and flying inside the vessel (Figure 2). It is noteworthy that there was no other insect besides *L. brunneus* on the wood samples. Therefore, *C. parva* adults could only have developed as parasitoid on this coleopteran.

Information regarding the reproductive ethological aspects of *C. parva* copulation and oviposition was gathered. The observed processes happened exclusively on *C. fairchildiana* wood samples infested by *L. brunneus*. All events were observed during the photophase.

Only the C. *parva* male sought the female for mating. There was no evidence of dispute between males for a receptive female. Mating started when the male touched a female body with its antennae. If the female was receptive to mating, the male then assumed the mating position by positioning itself on a plane higher than the female's lower metasoma, and then curving its own metasoma towards the female genitalia, remaining there for about 22 seconds, while vigorously flapping its wings.

Six mating attempts failed, mainly when males sought females that were probably already fertilized, which reacted by moving its antennae very fast, forcing the male to give up. The female antennae movement ceased after the male left. The receptive female did not move its antennae, suggesting that one male can mate with more than one female.

Females were observed inserting their ovipositor in the wood samples. *C. parva* specimens entered the holes produced by *L. brunneus*; assumingly, they were females searching for oviposition sites.

By the first time *C. parva* parasitizing *L. brunneus* is recorded in South America. Belokobylskij (2008) reported that *C. parva* specimens were only found in Hawaii, Australia, Indonesia, Vietnam, Japan, India, Puerto Rico, Cuba, and Panama. Although Muesebeck (1941)



Figure 1. Holotype (female) of *Cryptontsira parva* (Muesebeck, 1941) (originally described as *Doryctes parvus*).

reported that *C. parva* (at that time referred as *Doryctes parvus*) can parasite specimens belonging to genus *Lyctus*, there are no references in the literature about the relationship between *C. parva* and *L. brunneus*.

On the original description, *Cryptontsira parva* was regarded as a parasitoid of the bustrichid *D. minutus*, that is a widespread and destructive pest on felled culms and finished bamboo products. This species was also reared in China from *D. japonicus* Lesne from bamboo stems, and in India, from *Sinoxylon* sp. (Coleoptera, Bostrichidae), infesting cotton starks. In Australia, *Cryptontsira laemosacci* (Nixon, 1943) was collected from *Laemosaccus* sp. (Coleoptera, Curculionidae) (Muesebeck 1941, Belokobylskij 2008).



Figure 2. a) *Cryptontsira parva* (Muesebeck, 1941) specimen on *Clitoria fairchildiana* R.A. Howard wood infested by *Lyctus brunneus* (Stephens, 1830); b) *Cryptontsira parva* specimen next to exit holes made by *L. brunneus* in *Clitoria fairchildiana* wood.

According to Marsh (1997), most Doryctinae species, which have known biology, are idiobiont parasitoids, solitary or gregarious ectoparasitoids of boring insect larvae. Some Doryctinae species are usually described as parasitoids of beetles living between tree bark and sapwood. Therefore, these species can be used to control damages caused by xylophagous coleopteran (Shaw & Huddleston 1991).

According to Unger *et al.* (2001), *Monolexis fuscicornis* Förster, 1862 (Hymenoptera, Braconidae) is a natural enemy for *Lyctus* and *Hylotrupes bajulus* (Linnaeus, 1758) (Coleoptera, Cerambycidae) larvae. According to Yalçin *et al.* (2017), the parasitism rate of this Doryctinae on *L. brunneus* reaches 63.5 %; thus, it has a great potential to be used as pest control agent against *L. brunneus.* Although the parasitism rate of *C. parva* on *L. brunneus* is unknown, it is possible to assess its use as a biological control against pests infesting wood in several environments.

The register that C. parva reproduction only occurs in the photophasis is supported by Lewis & Whitfield (1999), who highlighted that most Braconidae species have exclusively daytime reproductive behavior. Cryptontsira parva mating behavior is similar to that of other species belonging to Braconidae (Rezende et al. 1995, Freitas et al. 2004, Benelli et al. 2012, Avila et al. 2017). Males in this group show complex sequence of signs during mating, including acoustic (wing vibration) (Bredlau et al. 2013, Canele et al. 2013) and chemical signs (McClure et al. 2007). Therefore, it is possible to expect that C. parva males use both strategies to mate.

Cryptontsira parva females, supposed to be fertilized, likely repeal males attempting to mate them by releasing chemical substances that cause immediate change in the receptor's behavior. According to Xu *et al.* (2014), *Cotesia glomerata* (Linnaeus, 1758) (Hymenoptera, Braconidae) females that are already fertilized repeal males.

According to Matthews (1975), Braconidae start the mating process when the male touches any part of the female's body with its antennae; after being noticed, he moves his antennae very fast and flaps his wings in response to female sexual pheromones. Tagawa & Hidaka (1982) noticed that, in cases when *C. glomerata* males have high sexual excitement, they do not touch the female with the antennae. Vinson (1972) suggests that wing vibration can help *Campoletis sonorensis* (Cameron, 1886) (Hymenoptera, Ichneumonidae) to guide itself to the source of the scent. Male wing vibration in some species belonging to Braconidae leads to higher female receptivity (Field & Keller 1993, Joyce *et al.* 2008).

The typical mating time of 22 seconds was similar to records by Rezende *et al.* (1995) for *Chelonus insularis* Cresson, 1865 (Hymenoptera, Braconidae). Although no dispute for females in *C. parva* was observed, Avila *et al.* (2017) suggested that *Cotesia urabae* Austin & Allen, 1989 (Hymenoptera, Braconidae) have more success mating when there are many males in the environment.

Cryptontsira parva parasitizes *L. brunneus* in a study carried out in laboratory environment in Rio de Janeiro (Brazilian state) and was for the first time recorded in South America. The species presented a mating behavior similar to that of other species belonging to Braconidae, and its outcome suggested that it is a potential organism to be used as biological control against the herein assessed coleopteran.

ACKNOWLEDGMENTS

The scholarship grant to second author by the Coordination for the Improvement of Higher Education Personnel (CAPES) - Finance Code 001 - is gratefully acknowledged. To Fapesp and CNPq for financial support to Project INCT - HYMPAR. To Sergey Alexandrovich Belokobylskij for confirm the identification of the species of *Cryptontsira* and to send the picture of the holotype of *C. parva*.

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Submitted: 20 June 2020 Accepted: 11 December 2020 Published on line: 23 December 2020 Associate Editor: Fernando Farache