

Full Length Research Paper

## Development and parasitism of *Encarsia hispida* (Hymenoptera: Aphelinidae) on *Bemisia tabaci* biotype B in cotton

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The biological control of pests is essential for the use of Integrated Pest Management in agricultural environments. In this context, the objective of this study was to identify biological parameters and quantify the parasitism index of *Encarsia hispida* on *Bemisia tabaci* biotype B nymphs in cotton plants. The research was conducted at the Entomology Laboratory of the Federal University of Paraíba, in Areia, Paraíba State, Brazil. For the first bioassay, the treatments consisted of cotton cultivars BRS H8 and BRS Topázio to evaluate the biological development of the parasitoid in its host. In the second bioassay, these cultivars were used to assess the impact of the biological agent in a greenhouse. In the first experiment, there were only female parasitoids with longevity of 24.61 and 22.61 days in BRS H8 and BRS Topázio, respectively. However, they were not statistically different. The life cycle of the parasitoid (egg to adult) was 35.68 and 33.71 days in BRS H8 and BRS Topázio, respectively, and they did not differ from each other. In the second bioassay, there were *E. hispida* parasitism indexes around 34.33 and 29.63% in BRS H8 and BRS Topázio, respectively. The parasitoid *E. hispida* develops properly when the nymphs of the host were from the two cotton cultivars. The parasitoid *E. hispida* has an application potential in the biological control of *B. tabaci* biotype B whiteflies.

**Key words:** Biological control, whitefly, biological parameters, parasitoid.

### INTRODUCTION

The cotton plant (*Gossypium hirsutum* L. var. latifolium Hutch) is significant in the Brazilian Agricultural Theater. It is produced in all five regions of Brazil, distributed in more than half of its federal units (Oliveira et al., 2012). The states of Mato Grosso, Bahia and Goiás are the

most relevant (IBGE, 2014). However, the whitefly *Bemisia tabaci* Gennadius (Hemiptera: Aleyrodidae) biotype B is a very important pest, for it causes significant losses in the agricultural production of various cultures in the world (Begum et al., 2011).

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The whitefly weakens the host plant during feeding and excretes sugary substances on the surface of leaves and fruits (Horowitz et al., 2011), causing sooty mold, which interferes with photosynthesis (Xu et al., 2013). In addition, it may inoculate the host plant with viruses, causing diseases in various cultures (Navas-Castillo et al., 2011). The control of *B. tabaci* biotype B is made exclusively by chemical methods, inducing the selection of resistant populations (Shadmany et al., 2013) and interfering with the survival of beneficial agents.

Among the most promising contributors to the control of the whitefly population, the species *Encarsia hispida* De Santis (Hymenoptera: Aphelinidae) may be mentioned (Hernández-Suárez et al., 2003; Lourencão et al., 2007; Torres et al., 2014). However, studies related to the influence of the whitefly's (*B. tabaci* biotype B) host plant on the biological aspects of *E. hispida* are needed. Takahashi et al. (2008) evaluated the biology of *Encarsia formosa* (Hymenoptera: Aphelinidae) in *B. tabaci* biotype B in different host plants and found that, when whitefly nymphs fed on tomato, they provided a more prolonged development of the parasitoid compared to those fed on cabbage. The information in this context is a relevant prior knowledge for the adoption of biological control programs.

The species *E. hispida* has an application potential in biological control of whiteflies. However, in the literature, there is no information about its biology as parasitoids of whitefly nymphs from agricultural and ornamental plants, although the potential of this parasitoid has been noted in records of success in controlling different species of whiteflies in field and at greenhouses. The objective of this study was to identify biological parameters and quantify the parasitism index of *E. hispida* in *Bemisia tabaci* biotype B nymphs in cotton plants.

## MATERIALS AND METHODS

The research was conducted at the Entomology Laboratory of the Federal University of Paraíba (LEN/UFPB), Areia Campus, Paraíba State. BRS H8 (white) and BRS Topázio (colored) cotton cultivars used in this study were from the Brazilian Agricultural Research Company at the National Center for Cotton Research (EMBRAPA/CNPA). The whitefly *B. tabaci* biotype B and the parasitoid *E. hispida* were obtained from cabbage plants (*Brassica oleracea* L. var. *acephala*) at Campus II of UFPB.

### Rearing of *B. tabaci* biotype B

The rearing of *B. tabaci* biotype B was in a greenhouse with poinsettia plants (*Euphorbia pulcherrima* Willd) using pots with a 10-liter capacity and a substrate with a blend of vegetable soil, manure and sand (1:2:1 ratio, respectively). The host plants were obtained by vegetative propagation. Three months after the emergence of shoots, they were infested with whiteflies. The plants were involved in circular galvanized metal frame cages with an aphid "voil" tissue mesh (50 × 26 cm). As for the suspension of cages, a wooden structure with galvanized wire on the sides was mounted, thus allowing cage height adjustment following the development of the plant.

After approximately 15 days, numerous whitefly nymphs were collected to find the colony. The environmental conditions were 26 ± 2°C, relative humidity 70 ± 10%, and photoperiod of 12 h. However, the maintenance of the whitefly population was made by free poinsettia plants.

### Rearing of *E. hispida*

Female parasitoids were collected from poinsettia using 00 gelatin capsules (Medeiros, 2009) released among poinsettia plants that would be colonized. They contained whitefly nymphs in their 3<sup>rd</sup> and 4<sup>th</sup> instars in the above-mentioned environment. After the release of the parasitoid, 3<sup>rd</sup> instar larvae of this biological agent were expected to excrete the meconium so that the darkening process to pupation would occur. After this process, the pupae were transferred along with leaves to the laboratory. Then, the pupae were removed with an entomological pin and placed in Petri dishes (9.0 × 1.5 cm) coated with plastic film until emergence.

After emergence, the adult insects were captured in 00 gelatin capsules and accommodated in test tubes (2.5 × 8.5 cm) with a honey solution (20%) distributed on the sides of the tubes to feed the parasitoid. Food was supplied every three days and the exchange of containers was made every 15 days. The containers were sealed with plastic wrap.

### Development of *E. hispida* on *Bemisia tabaci* biotype B in cotton

The bioassay was made by adapting the methodology proposed by Antony et al. (2003). Cotton cultivars BRS H8 and BRS Topázio, 30 days after planting, were placed in plastic bags with 1 kg of the above-mentioned substrate. The plants were placed at the laboratory at 25 ± 2°C, relative humidity 70 ± 10%, and photoperiod of 14 h. These plants were infested with 20 couples of whiteflies for oviposition using a cage (18 × 13 cm) with a "voil" tissue involving the leaves of the two cultivars for 24 h. After infestation, it was expected that whitefly nymphs reached the 3<sup>rd</sup> instar. Four nymphs on each leaf were selected for parasitoid infestation.

For the oviposition of the parasitoid, one individual with up to 24 h of age fed with honey was used. It was collected and released using 00 gelatin capsules (Medeiros, 2009) in clip cages (2.0 cm), allowing contact with the hosts for 24 h. After oviposition, parasitized nymphs were daily recorded using stereomicroscopy through the cuticle of whitefly nymph. Upon reaching the pupal stage, they were removed with an entomological pin and placed in containers (9.0 × 1.5 cm) waiting for the emergence of the parasitoid. After emergence, they were captured and transferred to test tubes (8.5 × 1.5 cm) containing food.

The parameters identified were the corresponding development periods of oviposition to larva, pupa period, pre-imago period, female longevity, oviposition to adult and sex ratio.

### Parasitism of *E. hispida* on *Bemisia tabaci* biotype B in cotton

For the record of the incidence of parasitoids in *B. tabaci* biotype B nymphs in cotton cultivars BRS H8 and BRS Topázio in a greenhouse, the methodology proposed by Simmons and Abb-Rabou (2005) suffered adaptations. Three leaves of each cultivar per pot were collected randomly. Each container contained three plants, totaling 30 leaves. The cotton plants were used 60 days of age in an environment at a temperature of 26 ± 2°C and relative humidity of 70 ± 10% with a 12 h photoperiod. The leaves were taken to the LEN/CCA for stereomicroscopy.

The observation of parasitism on nymphs by the parasitoids was recorded by their holes with their ovipositor on the host integument

**Table 1.** Biological parameters of *Encarsia hispida* parasitizing *Bemisia tabaci* biotype B in two cotton cultivars.

Cultivars	Egg to larva (days)	Pupa (days)	Pre-imagó (days)	Egg to adult (days)	Longevity of ♀ (days)	Sex ratio
BRS H8	6.01±0.09 <sup>a</sup>	5.06±0.08 <sup>a</sup>	11.07±0.13 <sup>a</sup>	35.68±1.67 <sup>a</sup>	24.61±1.67 <sup>a</sup>	1.0
BRS Topázio	6.05±0.12 <sup>a</sup>	5.05±0.09 <sup>a</sup>	11.10±0.17 <sup>a</sup>	33.71±1.16 <sup>a</sup>	23.61±1.17 <sup>a</sup>	1.0
CV (%)	7.79	7.56	6.13	18.63	27.37	

Means followed by the same letter in columns do not significantly differ from each other by F test (P = 0.05). Untransformed data ± mean standard error.

and the visualization of the larval development of this parasitoid inside the whitefly.

### Statistical analysis

The experiments were arranged in a completely randomized design (CRD). For the experiment I (development), the repetition consisted of four nymphs of 3<sup>rd</sup> instar whiteflies with 20 repetitions per cultivar. In experiment II (parasitism), using the data analyzed, it was calculated by the Simmons and Abb-Rabou (2005) equation:

$$P = \frac{NPP + NP + NA}{NN2 + NN3 + NN4 + NPP + NP + NA} \times 100$$

NPP = number of pre-pupae of the parasitoid; NP = number of pupae of the parasitoid; NA = number of adults of the parasitoid; NN2 = number of 2<sup>nd</sup> instar nymphs of whitefly; NN3 = number of 3<sup>rd</sup> instar nymphs of whitefly; NN4 = number of 4<sup>th</sup> instar nymphs of whitefly.

Data were subjected to analysis of variance and means of the treatments were compared by F test at 5% probability. Data were analyzed by the software Assistat 7.7 (Silva and Azevedo, 2002).

## RESULTS AND DISCUSSION

### Development of *E. hispida* on *Bemisia tabaci* biotype B in cotton

The evaluated biological parameters of *E. hispida* were not affected when *B. tabaci* biotype B nymphs were developed in cotton cultivars BRS H8 and BRS Topázio. For the larva to egg period, the pupa duration, the pre-imagó period, the egg to adult period and longevity had no significant differences from each other. Regarding the variable sex ratio, only female individuals of *E. hispida* were recorded when host nymphs were from both cotton cultivars (Table 1).

It was found that the parasitoid broke the integument of the host using the ovipositor to feed from the hemolymph. The sucking of the hemolymph of the host by parasitoids enables them to acquire nutrients. However, this process destroys the parasitoid's oviposition opportunity for the development of their offspring (Shah et al., 2015).

The 3<sup>rd</sup> instar larva of the parasitoid, upon releasing meconium, begins the sclerotization process of its cuticle in a matter of days. When this is completed, the pupal stage begins. After its formation, it shows small movements within its host in a matter of hours,

decreasing when the emergence of adults is close. Before emergence of the adult, it is observed that the individual changes its body position to perform an opening in the host. According to Antony et al. (2004), this type of adult insects creates an opening in the cuticle of the host to enable its emergence.

The values corresponding to the periods egg to larva and pre-imagó of this study differ from those found by Azimi et al. (2014) for *Encarsia formosa*, and Pessoa et al. (2016) for *Encarsia desantisi* (Hymenoptera: Aphelinidae) in non-BT cotton. Thus, researchers have shown that species from the genus *Encarsia* may reduce or extend its life cycle in function of the host plant where the whitefly developed.

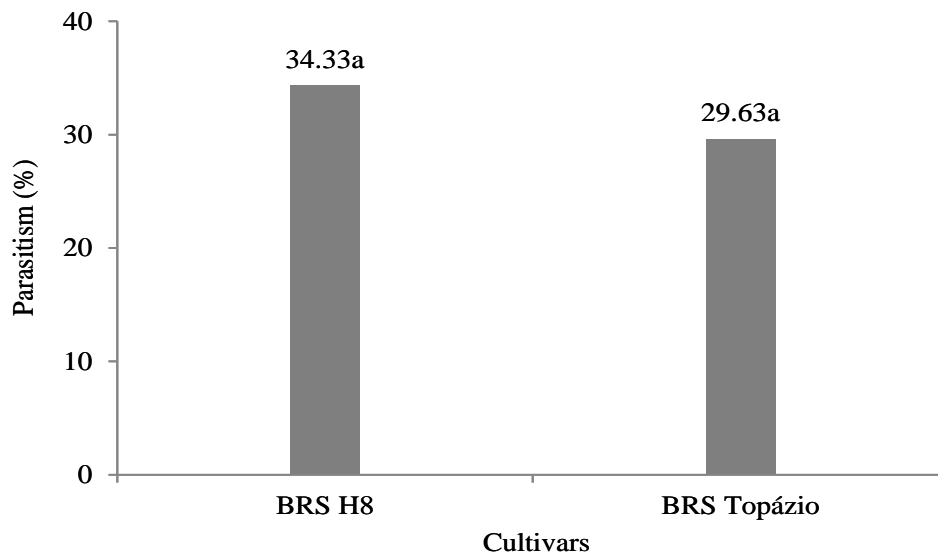
The results for the egg to adult period of *E. hispida* which developed in both cotton cultivars indicate that host nymphs of *B. tabaci* biotype B, allow a proper biological development of the parasitoid. According to Talaei (2009), the plant host is an important factor for the adequacy of hosts to parasitoids.

The longevity of the female parasitoid was 24.61 and 22.61 days for BRS H8 and BRS Topázio, respectively. Pessoa et al. (2016), evaluating the natural agent *E. desantisi* in *B. tabaci* nymphs from cotton cultivars DeltaOpal and FM 993, found longevity values of 19.3 and 22.3 days, respectively. According to Hódar et al. (2002), the quality of food is one of the main factors influencing longevity, body size and abundance of fertility.

According to Giorgini et al. (2009), a characteristic of the genus *Encarsia* is its asexual reproduction, thelytokous parthenogenesis, where the presence of males is rare or unknown. This characteristic of parasitoids, that is, breeding female individuals, is of great importance in biological control programs. There is a possibility of the presence of the symbiont *Cardinium hertigii*, which induces parthenogenesis and breeds exclusively female individuals of *E. hispida*. They are located with a larger quantity in follicular and nutritive cells, and to a lesser extent in parasitoid oocytes (Zchori-Fein et al., 2004). Thus, the symbiont influenced sex ratio, which was 1.0 in both cultivars assessed.

### Parasitism of *E. hispida* on *Bemisia tabaci* biotype B in cotton

The parasitism index of *E. hispida* in *B. tabaci* biotype B



**Figure 1.** Parasitism of *E. hispida* when whitefly nymphs were from the two cotton cultivars.

nymphs from both cotton cultivars were not statistically different from each other ( $F = 3.9623$ ,  $P = 0.0618$ ) (Figure 1). In total, 1,230 and 933 parasitoids from whitefly nymphs in BRS H8 and BRS Topázio cultivars, respectively, were recorded. Among these parasitoids, there was the presence of individuals from both sexes in both cultivars. They are visually different in color. The female has a light yellow color all over the body, while the male has a brown color (Myartseva and Evans, 2008).

Graff et al. (2006) reported the performance of this natural agent regarding its parasitism in *B. tabaci* pests in vegetable plants. They concluded that its effect on the density of this pest's nymphs varied only in tomatoes, while in pepper and cucumber plants it had the same parasitism rate on the host. These authors analyzed the use of *E. hispida* for *B. tabaci* on hibiscus plants from 2004 to 2005 to control the plague, and found high rates. Yet the parasitoid behavior was influenced by abiotic and biotic factors. Furthermore, the use of synthetic products affected this agent regarding the control of *B. tabaci*. In Brazil and in the world, the species *E. hispida* has been reported affecting different host species of several ornamental plants, vegetables and crops, resulting in socioeconomic damage (Hernandez-Suarez et al., 2003; Oliveira et al., 2003; Lourencão et al., 2007; Torres, 2014).

## Conclusion

The parasitoid *E. hispida* develops properly when *B. tabaci* biotype B nymphs are from the two cotton cultivars BRS H8 and BRS Topázio. The parasitoid *E. hispida* has an application potential in the biological control of the whitefly *B. tabaci* biotype B.

## Conflict of Interests

The authors have not declared any conflict of interests.

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