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Impact of Host Size and Honey Availability on *Trichogramma atopovirilia* (Hymenoptera: Trichogrammatidae) Longevity¹

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Egg parasitoids of the genus *Trichogramma* (Hymenoptera: Trichogrammatidae) are used as an augmentative biological control tactic in millions of hectares of crops worldwide, where they are released for combating lepidopteran pests (van Lenteren and Bueno 2003, BioControl 48: 123-129). For this tactic to be effective, it is essential to use factitious hosts, such as Sitotroga cerealella (Oliver) (Lepidoptera: Gelechiidae) or Anagasta kuehniellla (Zeller) (Lepidoptera: Pyralidae), to reduce rearing costs and improve profitability of Trichogramma commercialization (Bernardi et al. 2000, Rev. Bras. Biol. 60: 45-52). Biological fitness of egg parasitoids is influenced by the host because the host egg is the only source of nutrients for the development of the parasitoid (Lampson et al. 1996, Environ. Entomol. 25: 283–294; Dias-Pini et al. 2014, J. Appl. Entomol. 138: 677–682). Host guality could affect adult parasitoid size, longevity, and parasitic capacity (Marston and Ertle 1973, Ann. Entomol. Soc. Am. 66: 1155-1162; Corrigan and Laing 1994, Environ. Entomol. 23: 755–760; Kuhlmann and Mills 1999, Biocontrol Sci. Technol. 9: 335–346). For example, larger adults of Trichogramma pretiosum (Riley) were obtained from larger hosts, and their size was positively correlated with other biological attributes (Bai et al. 1992, Entomol. Exp. Appl. 64: 37-48). Large Trichogramma adults live longer and have greater male search success rate in contrast to smaller parasitoids (Kazmer and Luck 1995, Ecology 76: 412-425). The objective of this work was to determine the effect of host size, natural or factitious, and the availability of a supplemental carbohydrate source (honey) on the longevity of Trichogramma atopovirilia (Oatman and Platner).

Naturally occurring *T. atopovirilia* were collected initially from a crop production field in Guanajuato, Mexico, in October 2018. A laboratory colony was established and has been reproduced on its natural host, *Spodoptera frugiperda* (J.E. Smith)

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(Lepidoptera: Noctuidae), since then. The host Spodoptera frugiperda was reared on a meridic diet according to Jaraleño-Teniente et al. (2020, Insects 11: 157). Eggs of Sitotroga cerealella were obtained weekly from a commercial rearing facility (Organismos Benéficos para la Agricultura, Autlán, Jalisco, Mexico) and separated in a series of sieves (20, 40, and 60) with apertures of 0.71, 0.38, and 0.25 mm. Three relative sizes of eggs were obtained in this process. Only the small (60-sieve) and large (40-sieve) eggs were used in our assays. We therefore maintained a colony of T. atopovirilia on its natural host (Spodoptera frugiperda) and established two additional colonies with the large and the small eggs of Sitotroga cerealella. Each colony was maintained in $26 \times 13 \times 13$ -cm plastic containers with 5×5 -cm holes covered with silk screen mesh to allow for ventilation. Each day, a cotton wick saturated in a solution of water and honey (10:1) along with an opaline paper card $(4 \times 4 \text{ cm})$ with about 100 T. atopovirilia eggs corresponding to each aforementioned colony were placed in each plastic container. The eggs were exposed for 24 h to T. atopovirilia, then they were removed and placed in a container similar to that described above for maintaining the three colonies.

The experimental assays were initiated after the third generation on the factitious host. All experimental assays were maintained in a rearing chamber at $25 \pm 2^{\circ}$ C, $75 \pm 5\%$ relative humidity, and on a 12:12-h light/dark photo regime. To measure size of the host, two samples were taken at random (e.g., 15 eggs on each occasion) from each host colony (*Spodoptera frugiperda* host, large and small eggs of *Sitotroga cerealella*). Using a professional camera (Canon EOS 50D) on a stereomicroscope (Carl Zeiss[®] Discovery V20), photographs were taken and individual images were recorded to later measure them with the Java-based ImageJ program (Softonic International S.A., Barcelona, Spain).

Trichogramma atopovirilia longevity was evaluated with wasps placed individually in 1-cm-diameter Eppendorf tubes for each treatment. After inserting the adults, each tube was covered with screen mesh to allow for ventilation. In addition, the variable of carbohydrate availability (honey) was added. Thus, every third day, small drops of honey were placed on the inner wall of the tube according to the assigned treatment (with or without honey). Parasitoid sin each tube were observed twice a day (9 a.m. and 5 p.m.), and parasitoid death was recorded. The six treatments were replicated three times using 7, 8, and 20 individuals for each treatment (n = 35). Data from host size (width) and longevity were analyzed using analysis of variance. If these analyses showed statistical differences among treatment means, the means were separated using Tukey's honestly significant difference comparison ($P \le 0.05$). A Student's *t* test ($P \le 0.05$) was used to compare the length of large and small eggs of *Sitotroga cerealella*. Analyses were conducted using the Statistix 8.1 (Analytical Software 2003, Tallahassee, FL).

Significant differences were obtained with the comparison of egg width of hosts (F = 1,893; df = 2,87; P < 0.0001). The diameter of *Spodoptera frugiperda* eggs (520.85 ± 3.7 µm) was almost twice the width of the factitious host eggs (large: 265.7 ± 3.6 µm; small: 247.4 ± 3.2 µm). Furthermore, no differences were found in the length (around 622 µm) of large and small *Sitotroga cerealella* eggs (t = 0.81, df = 58, P = 0.419). In general, host size and availability of honey significantly affected longevity of *T. atopovirilia* adults (F = 31.97; df = 5,186; P < 0.0001), with the natural host (*Spodoptera frugiperda*) and addition of honey yielding the longest *T. atopovirilia* adult longevity (12.8 ± 0.9 d). *Trichogramma atopovirilia* that emerged

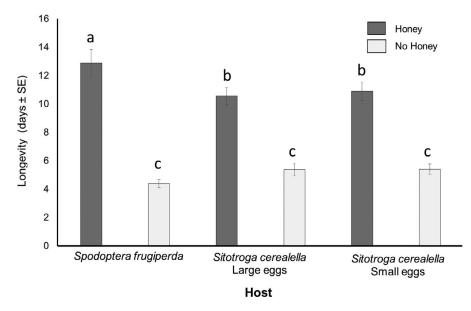


Fig. 1. *Trichogramma atopovirilia* longevity in response to a natural host and two sizes of a factitious hosts under laboratory conditions ($25 \pm 2^{\circ}C$, $75 \pm 5\%$ relative humidity, 12:12 h light/dark). Bars with the same letter are not significantly different (P > 0.05; Tukey's honestly significant difference).

from large and small eggs of *Sitotroga cerealella* lived 2 d less compared to those that emerged from *Spodoptera frugiperda* eggs (10.5 and 10.9 d, respectively). Parasitoids that emerged from the large host or the large or small factitious host without honey had the shortest longevity (4.4–5.4 d), and no significant differences were detected among those treatments (Fig. 1).

Availability of a carbohydrate source for parasitoids (e.g., honey, nectar, or honeydew from hemipteran insects) is vital to extend longevity of egg parasitoids in the field (Wäckers et al. 2008, Biol. Control 45: 176-184). In this work, T. atopovirilia that developed in the largest host (Spodoptera frugiperda) and with the provision of honey increased its longevity by 180% compared to without honey (4.4 d versus 12.8 d). Other authors have reported increasing longevity around 70% in T. pretiosum (6.7 d versus 11.4 d) due to honey availability (Bai et al. 1992; Cañete and Foerster 2003, Rev. Bras. Entomol. 47: 201–204). This reinforces the need for availability of a carbohydrate source in the field for the parasitoid to attain its reproductive potential. Without this supplemental source of carbohydrates, there would not be sufficient energy to maintain vital functions, much less to reserve the carbohydrates in the form of glycogen to increase longevity, flying, and parasitism (Bernstein and Jervis 2008, pp. 129-171 in Wajnberg et al. (eds.), Behavioral Ecology of Insect Parasitoids: From Theoretical Approaches to Field Applications, Blackwell Publishing; Strand and Casas 2008, pp.113–128 in Behavioral Ecology of Insect Parasitoids: From Theoretical Approaches to Field Applications).

Although a favorable effect on longevity was found in parasitoids that emerged from the largest host (*Spodoptera frugiperda*) with honey availability, no favorable effect was found for *T. atopovirilia* when there was no honey. In other words, a parasitoid developing in *Spodoptera frugiperda* but without availability of honey as an adult was not more resistant and did not live longer than the parasitoids that developed in the smaller hosts (*Sitotroga cerealella*). Host quality influenced the longevity of *T. atopovirilia*, as has been reported by others working with *Trichogramma* (Marston and Ertle 1973; Kuhlmann and Mills 1999), but no effect was obtained when a carbohydrate source was absent. A small variation in host size, and the size and biology of *T. atopovirilia* itself, are likely important under ideal conditions (availability of carbohydrates) but, under conditions of stress, there was no advantage in increasing longevity. Additional studies on this topic need to be conducted to evaluate the advantages of larger hosts on valuable traits of this parasitoid, such as fecundity and parasitism.

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