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THE PARASITOID FLY *ORMIA OCHRACEA* (DIPTERA: TACHINIDAE) CAN USE JUVENILE CRICKETS AS HOSTS

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ABSTRACT

The parasitoid fly *Ormia ochracea* uses the calling song of its host *Gryllus* spp. to locate an area inhabited by potential hosts. Once a calling male has been located, *O. ochracea* deposits live larvae on the host, and on substrates surrounding the host to enable the larvae to attach to and enter individuals in the vicinity of the calling male. In Texas, where *O. ochracea* parasitizes the Texas field cricket *Gryllus texensis*, we observed juvenile crickets in the mating aggregations that form around calling males. Juvenile *G. texensis* crickets are, therefore, potentially susceptible to parasitism by *O. ochracea*. Here we investigated whether laboratory reared juvenile field crickets could successfully host *O. ochracea* larvae. We found that juvenile crickets were appropriate hosts for *O. ochracea*.

Key Words: parasitoid, cricket, host, parasite, juvenile

RESUMEN

La mosca parasitoide *Ormia ochracea* usa el canto de cortejo del hospedero *Gryllus* spp. para ubicar las áreas habitadas por hospederos potenciales. Una vez que el canto del macho ha sido localizado, *O. ochracea* deposita larvas vivas sobre el hospedero y sobre los sustratos alrededor del hospedero para que larvas pueden atar y entrar los individuos en la vecindad del canto del macho. En Texas, donde *O. ochracea* parasita el grillo de campo Tejano, *Gryllus texensis*, observamos grillos juveniles en las agregaciones de apareamiento que se forma alrededor de los machos cantando. Los grillos juveniles de *G. texensis* entonces son potencialmente susceptibles al parasitismo por *O. ochracea*. Aquí, investigamos si los juveniles de los grillos de campo criados en el laboratorio pueden exitosamente ser hospederos de las larvas de *O. ochracea*. Encontramos que los juveniles de los grillos fueron hospederos apropiados para *O. ochracea*.

Parasites and parasitoids are often limited in host range because of 4 factors: (1) parasites and potential hosts may not overlap in their ranges; (2) they may not come into direct contact for a variety of behavioral or ecological reasons; (3) potential hosts fail to support the essential spatial or metabolic demands of the parasite or parasitoid; and (4) potential hosts may employ a variety of defence mechanisms to avoid being parasitized (Euzet & Combes 1980, referenced in Combes 2001). These limits on host availability may explain why parasites often utilize a specific life stage of their hosts as it is not often that larval and adult, or juvenile and adult life stages of hosts coalesce with respect to all 4 of these limiting factors. However, if a species changes its distribution or behavior, it may put itself at risk of becoming host to a previously absent parasite. How species traverse these boundaries and whether parasites are capable of exploiting these boundary breaches is an exciting area of investigation for parasitologists. Here we investigate whether crickets and their parasitoid fly demonstrate such a violation of host specificity.

Singing males of the Texas field cricket, *Gryllus texensis* (Orthoptera: Gryllidae), are acoustically stalked and parasitized by gravid females of the parasitoid fly *Ormia ochracea* (Diptera: Tachinidae) (Cade 1975). *Ormia ochracea* are larviparous; once they locate their host, the gravid female deposits her planidia (1st instar *Ormia*) on and around the cricket (Cade 1975). Planidia laid around the cricket wave their anterior ends in the air in an attempt to come in to contact with a host. Once contact is established, the planidia burrow into the host's body. Inside the host, the larvae feed on the cricket's fat body, and abdominal and thoracic muscles (Adamo et al. 1995). Parasitized crickets initiate an encapsulation response to kill the planidia before they have a chance to establish themselves (Vinson 1990). However, encapsulation responses are often co-opted by the parasitoid larvae and used to construct a trachea that they attach to the cricket's abdominal wall (Vinson 1990). After 7 to 10 d, the larvae emerge from the host (Adamo et al. 1995). Larval emergence invariably results in the death of the host cricket.

The observation that gravid flies deposit larvae around calling males (Cade 1975) as well as observations made during behavioral trials conducted in our laboratory, suggests that all crickets in the vicinity of a calling male are in danger of

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parasitization by *O. ochracea*. In fact, several authors have observed parasitized females in the wild even though female crickets do not acoustically signal (Walker & Wineriter 1991; Zuk et al. 1993; Adamo et al. 1995). To our knowledge, parasitized juvenile crickets have yet to be observed, although juvenile crickets have been observed in the vicinity of calling males. While collecting *G. texensis* in the field in Texas during Sep 2007, more than 30% of the individuals that we collected from the area immediately around calling male crickets were penultimate and last instar juveniles. Thus, these juvenile *G. texensis* were in danger of becoming parasitized. Using 3rd generation lab reared *G. texensis*, we compared the progress of an *O. ochracea* infestation in penultimate juveniles to that of adults to determine whether juvenile *G. texensis* are suitable hosts for *O. ochracea* larvae.

Materials and Methods

Crickets and flies were collected nightly from several locations between Austin and Smithville, Texas, in 2007. Flies were trapped with a sound trap modelled after Walker's (1989) slit trap design. A Durabrand CD-566 compact disc player (Lennox Electronics Corporation, 35 Brunswick Avenue, Edison, NJ 08817) was connected to an Amplified Speaker AMX 18 (RadioShack Corporation, Fort Worth, Texas 76102) and then placed underneath the sound trap. This speaker broadcasted a natural call of a *G. texensis* male at a decibel level of 61db from 30 cm. The broadcasted call was recorded from a laboratory-reared male *G. texensis* and had a dominant frequency of 4.6 kHz. Gravid female *O. ochracea* were attracted to the broadcast and entered the trap via the slit, becoming ensnared. Captured flies were housed in a terrarium and provided with *ad libitum* hummingbird feed. Crickets and flies were brought back to establish laboratory colonies of each at Carleton University, Canada.

We used a manual parasitization method (Vincent & Bertram in press) to parasitize the crickets. Briefly, a gravid *O. ochracea* female was chilled and her abdomen removed. The abdomen was gently teased apart, the reproductive tract opened, and the planidia exposed. Once planidia were exposed to the air, they 'stood' on their posterior ends and moved back and forth in a 'waving' motion. These planidia were gently probed with a blunt dissecting tool until they latched on to the tool. They were then transferred to a cricket. Two actively waving planidia were gently placed on the articular sclerites (located at the anterior end of the thorax where the wings attach) of each host cricket. In total, 74 penultimate juveniles (37 males/37 females) and 57 adults (28 males/29 females) were parasitized in this manner.

Crickets were housed individually and provided *ad libitum* food and water until larval parasitoid emergence. After mature parasitoid larvae emerged from the host and pupated, the pupae were housed separately in an incubator so that the success (eclosion) of each cricket's larvae could be determined. The dates of parasitization and larval emergence were recorded as well as the number of days after larval emergence the cricket survived. All crickets that did not succumb to parasitization were frozen on the 14th d following manual parasitization. Crickets were later thawed and dissected to ascertain whether there was any visible evidence of parasitism (e.g., encapsulated parasitoid larvae or parasitoid trachea).

Comparisons of parasitoid success in juvenile and adult cricket hosts were based on the following measurements: number of days from manual parasitization to first mature larval emergence; the total number of larvae that emerged from each cricket (0, 1, or 2); the number of days that the cricket survived following larval emergence; success of parasitoid pupae (proportion of pupae that eclosed: none-0, half-1, all-2); the number of larvae that established, as determined by counting the number of parasitoid trachea found in cricket's abdomen during dissections; the proportion of crickets with larval emergence; and the proportion of larvae that emerged versus became established in the cricket host.

Results

Ormia ochracea planidia were able to establish themselves in juvenile crickets. We found no significant difference between adult and juvenile crickets in the number of planidia that successfully established, the number of larvae that successfully emerged, or in the success of emerged larvae (Table 1). Juvenile crickets lived significantly longer than adult crickets post larval emergence (juveniles = 1.03 days, adults = 0.61 days, $P = 0.015$; Table 1).

Discussion

The observance of penultimate juvenile crickets in mating aggregations led us to postulate that juvenile *G. texensis* may be parasitized by *O. ochracea* in the wild, and indeed in laboratory studies, juvenile crickets can serve as hosts for *O. ochracea*. Penultimate stage juvenile *G. texensis* appear to be ideal hosts for *O. ochracea*. The number of parasitoid larvae that established within a cricket, the number of larvae that emerged, the proportion of established versus emerged larvae, and the time it took for the larval parasitoids to emerge from the cricket did not differ between juvenile and adult cricket treatments. Our results suggest that *O. ochracea* could successfully use penultimate juvenile crickets as hosts in nature.

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TABLE 1. *ORMIA OCHRACEA* SUCCESS WITHIN JUVENILE AND ADULT CRICKETS.

	ADULT	JUVENILE	
Mean No. Adults Eclosing from Pupae (0-2)	1.71 ± 0.1405	1.92 ± 0.0576	χ^2 = 3.23 n = 45 P = 0.2004
Mean No. Larvae Established (0-2)	1.35 ± 0.104	1.30 ± 0.102	χ^2 = 0.934 n = 107 P = 0.82
Proportion of Crickets with Larval Emergence	0.634	0.763	χ^2 = 3.75 n = 107 P = 0.053
Mean No. Larvae Emerged (0-2)	1.18 ± 0.0969	1.31 ± 0.0778	χ^2 = 4.19 n = 86 P = 0.19
Mean No. Days Until Larval Emergence	6.71 ± 3.45	7.44 ± 2.98	ANOVA: n = 86 F = 1.99 P = 0.16
Mean Proportion of Emerged / Established	0.7804	0.844	χ^2 = 0.579 n = 86 P = 0.45
Mean No. Days Cricket Survived Following Larval Emergence	0.614 ± 0.839	1.03 ± 1.02	ANOVA: n = 86 F = 6.13 P = 0.015

[1]*df* for all analyses was 1.

Our results show that *O. ochracea* has a similar life history in hosts of different stages of development in a laboratory population of crickets. However, it remains to be demonstrated whether juvenile crickets become parasitized in the wild. If juvenile crickets are regularly utilized by *O. ochracea* as hosts in nature, this might impact populations of *G. texensis*.

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