Oviposition Behavior and Development of *Goniozus triangulifer* Kieffer (Hymenoptera: Bethylidae)

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**ABSTRACT** Bionomics and behavior of the bethylid parasitoid, *Goniozus triangulifer* Kieffer, were studied in the laboratory with leaffolders, *Cnaphalocrocis medinalis* (Gueneée) and *Marasmia patnalis* (Bradley), as hosts. Female parasitoids lived twice as long as males (22 and 11 d, respectively) and laid about 80 eggs during their lifetime. Single male broods occurred in 64% of the samples and were significantly more frequent than expected from the binomial distribution only when the brood size was six. Females exhibited complex behavior upon encountering a host. Malaxation occurred before the female parasitoid paralyzed the host. Parasitoids laid an average of 4.5 eggs on unparasitized hosts and 2.4 eggs on previously parasitized ones, but commonly did not lay at all on these. Total handling time for experienced parasitoids on parasitized hosts was significantly shorter than that for unparasitized hosts. *G. triangulifer* displays the ability to discriminate between unparasitized hosts and those parasitized by conspecific females.

**KEY WORDS** Insecta, *Goniozus triangulifer*, *Cnaphalocrocis medinalis*, *Marasmia patnalis*

*Goniozus triangulifer* Kieffer is a gregarious external parasitoid of leaffolders, *Cnaphalocrocis medinalis* (Gueneée) and *Marasmia patnalis* (Bradley), on rice. We found no reports of the biology of *G. triangulifer* although it is a common leaffolder parasitoid in the Philippines.

We conducted studies of *G. triangulifer*, including bionomics, sex ratio, and behavior, especially in the context of oviposition and host discrimination.

**Materials and Methods**

*G. triangulifer* was established from leaffolder larvae, *C. medinalis* and *M. patnalis*, collected from rice fields in Laguna Province, Philippines, and reared on these hosts under laboratory conditions at 27 ± 5°C, 50-60% RH with a 14:10 (L:D) photoperiod. Mated female parasitoids were placed individually into test tubes (14 by 1.5 cm) plugged with cotton and fed daily with honey solution. Third- and fourth-instar hosts of *C. medinalis* were introduced into the test tubes on single cut rice leaves. A small amount of water in the bottom of the tube kept the leaf turgid. After oviposition by the parasitoid, each host larva and a section of leaf were placed into a test tube.

**Bionomics.** Records were made of the time between oviposition until the adults emerged (n = 196). Adult longevity was measured by placing 20 newly emerged mated wasps of both sexes individually into test tubes. This group was not given hosts. Longevity was determined for another group of females that were given hosts throughout their lives. Fecundity was measured by placing newly emerged, mated females singly into test tubes and providing them with one host larva daily. All adult parasitoids, including those in longevity experiments, were fed daily with honey solution. An overall estimate of sex ratio was obtained from the wasps that emerged in culture.

Another method of determining sex ratio was conducted using the method of Green et al. (1982). Broods were classified according to size, using only those with complete emergence. The number of single-male broods for each size brood was counted. The probability of the occurrence of these numbers was calculated assuming a binomial distribution of males using the equation:

\[
\alpha = \sum_{i=0}^{N} \binom{n}{x} \hat{p}^x (1 - \hat{p})^{n-x}
\]

where \(\hat{p} = (1 - 1/N)^{k-1}\), N is brood size, n is the frequency of brood size occurrence, k is the number of single-male broods of size N, and x is the number of males.

**Behavior.** We observed the behavior of *G. triangulifer* from emergence of adults to oviposition. One male and three or four females were kept together in tubes, and a single host was introduced in the manner described earlier. In addition, individual mated females presented with a host larva were observed.

Oviposition behavior was monitored to detect qualitative and quantitative differences. Mated females were allowed to oviposit on hosts from ca. 1000 to 1600 hours; then larvae were removed and...
numbers of eggs counted. At ca. 1000 hours the next day, each wasp was presented with the same host she had parasitized the day before. A host was considered rejected if the wasp failed to attack it for a period of 1 h despite repeated encounters.

In another experiment, parasitized hosts were presented to mated but inexperienced females that had not oviposited. Behavior was monitored as in the previous treatments.

Data were analyzed statistically by one-way analysis of variance (ANOVA). When F values were significant, means were compared using Duncan’s multiple range test (DMRT) (Duncan 1955). Longevity of males was analyzed by t test.

Results and Discussion

Bionomics. Average (±SD) longevity of males (n = 20) was 11.2 ± 4.4 d, and longevity of females (n = 20) given hosts was 22.1 ± 8.0 d. The latter time did not differ statistically from that of 20.4 ± 8.9 d for females deprived of hosts (t = 0.53; df = 19; P < 0.05).

An average (±SD) of 80.7 ± 27.2 eggs was laid during the lifetime of a female (n = 10). Average number of eggs per host was 4.8 ± 2.0 (n = 455) and development from oviposition to adult emergence was 12 ± 1 d (n = 196). Females often ate their own eggs so total fecundity is probably underestimated. Overall male/female sex ratio was 1:1.9 (n = 698). Mortality during development was high, resulting in only 33.2% emergence.

The frequency of single-male broods is shown in Table 1. For brood sizes two to seven, the proportion of single-male broods was 64%. However, the number of single-male broods was significantly higher than the binomial only when the brood size was six. Although juvenile survival was low, we conclude that sex ratios in G. triangulifer were not as precisely controlled as those found for G. gordhi.

Sex ratios in several species of Goniozus are not random. As in other arrhenotokous parasitoids, small broods are female-biased because only one male is required to inseminate cohort females. As brood size increases, more unfertilized (male) eggs are laid. Green et al. (1982) found that the number of single-male broods in G. gordhi that attacked Deo- clona yuccaeca Busck. differed significantly from that predicted by a binomial distribution. Brood sizes from two to nine were predominantly single-male broods, for larger broods more males emerged. Some evidence of nonrandom sex ratios also was found for Goniozus emigratus (Rohwer) on the navel orangeworm, Amyelois transitella (Walker) (Gordh & Hawkins 1981). Broods were larger and not usually single-male but numbers of males within a brood were not random. Furthermore, the proportion of females declined over 15 sequentially offered hosts. Eventual sperm depletion resulted in all-male broods.

The optimal reproductive strategy hypothesis predicts that in small broods, only one male is required to maximize future number of offspring (Hamilton 1967). Green et al. (1982) reported this for G. gordhi.

Behavior. Males were not observed to copulate with preemergent females as in other species (e.g., Gordh et al. [1983]). Female parasitoids attacked a host immediately after emergence even before mating. The initial attack consisted of the parasitoid malaxating the anterior dorsal region of the host larva. Simultaneously or immediately afterwards, the parasitoid arched its abdomen and injected venom ventrally in the host’s venter, paralyzing it. Frequently, she left the host while the host became paralyzed. Upon returning, she moved up and down the host, occasionally using her antennae and probing with the tip of her abdomen. We interpreted this as an assessment of the quality of the host in terms of size, or whether or not the host was paralyzed. Frequently, the female parasitoid injected more venom in the middle and posterior of the host. Also, she injected venom ventrally in the cervical region.

Immediately before oviposition, the female again moved up and down the host, with the tip of the abdomen or ovipositor quickly moving from one side of the host to the other. We designated this as “ovipositor probing” although we could not determine if the stinger was inserted. Upon selecting a suitable oviposition site, the wasp stopped moving the ovipositor laterad of the host and remained stationary. We refer to this as “oviposition preparation” which immediately preceded oviposition.

Just before an egg was laid, the abdomen contracted. These contractions increased in frequency until an egg was deposited. During oviposition, the stinger protruded. The tip of the ovipositor opened, the body was arched, and the egg was deposited on the outside of the host’s body. There was a slight pause during which the ovipositor closed, then probing with the ovipositor was resumed. Eggs were laid alternately on the side of each abdominal segment beginning from the posterior and moving towards the middle of the host.

Table 1. Frequency of single-male broods in G. triangulifer and probabilities of occurrence

<table>
<thead>
<tr>
<th>Brood size (N)</th>
<th>Frequency (n)</th>
<th>Single-male broods (k)</th>
<th>P*</th>
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<tbody>
<tr>
<td>1</td>
<td>5</td>
<td>2</td>
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<tr>
<td>2</td>
<td>4</td>
<td>1</td>
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<td>12</td>
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<td>0</td>
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* Probability of occurrence of single-male broods of size N, using a binomial function following the method of Green et al. (1982) (see text for explanation).
Females were observed guarding hosts from conspecific females but time spent doing this was not determined.

*G. triangulifer* differed from other species within the genus in two ways: preferred oviposition sites for *G. triangulifer* were near the posterior of the host; the host recovered from the paralysis in about 30 min.

A summary of results from the experiments related to oviposition behavior is given in Table 2. Some females attempted to drag the larva away, especially when more than one parasitoid was present. This behavior lasted \((x \pm SD) 59 \pm 36.65\) s, but our observations were limited to two occasions. In the field this behavior may protect the wasp and the host larva from predators or conspecific females.

When wasps were given parasitized larvae, they spent some time moving across the larvae vibrating their antennae. We described this activity as "antennal vibrations." When presented with a parasitized host, experienced females either rejected the host outright after examining it with their antennae or attacked the host, ate the eggs, and then malaxed, or occasionally, laid a new batch of eggs. Experienced parasitoids rejected parasitized hosts in 22 of 30 replicates. When an inexperienced female was presented with a parasitized host, she always attacked the host, ate all the eggs laid previously, then oviposited. She usually avoided the previous oviposition sites and never laid more eggs than her predecessor.

Mean total handling time for experienced female wasps on unparasitized hosts was not significantly different from that of inexperienced females on parasitized hosts (Table 2). However, there was a significant reduction in handling time by experienced wasps on parasitized hosts. In addition, time spent in preoviposition and oviposition was significantly lower with experienced wasps presented parasitized hosts than with other parasitoid/host combinations. Females spent more time off the larvae when unparasitized larvae were available than when parasitized ones were present, regardless of whether the wasps were experienced or not. Experienced females also spent more time moving over the host, perhaps assessing its quality.

One major difference in duration of ovipositional activities between experienced and inexperienced wasps was in the number of eggs they laid. Average number of eggs for each group of females was calculated only for those which oviposited. Experienced females given unparasitized hosts laid a mean \((\pm SD) 4.5 \pm 0.97\) eggs. Experienced females given parasitized hosts laid a mean of 2.4 \(\pm\) 2.0, although most females did not oviposit at all, and inexperienced females given parasitized hosts laid 4.0 \(\pm\) 0.8 eggs per host. Numbers of eggs laid by experienced females on parasitized hosts were significantly lower than that of experienced females on unparasitized hosts \((t = 2.75; df = 19; P < 0.05)\) or inexperienced females on parasitized hosts \((t = 2.1; df = 19; P < 0.05)\).

**Acknowledgment**

We gratefully acknowledge the assistance of Gordon Gordh in identifying *G. triangulifer*. Voucher specimens of the species are deposited with Gordon Gordh (Division of Biological Control, University of California, Riverside). M. Lava provided additional technical assistance. K. Manalili and J. Crisostomo assisted in the statistical analysis.

**References Cited**


Gordh, C. & B. Hawkins. 1981. *Goniozus emigratus*, a primary external parasite of *Paramyelois transintelata* (Walker), and comments on bethylids attacking...


Received for publication 14 October 1986; accepted 11 August 1987.