Behaviors of Female *Eretmocerus* sp. nr. *californicus* (Hymenoptera: Aphelinidae) Attacking *Bemisia argentifolii* (Homoptera: Aleyrodidae) on Cotton, *Gossypium hirsutum,* (Malavaceae) and Melon, *Cucumis melo* (Cucurbitaceae)

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Behaviors of *Eretmocerus* sp. nr. *californicus* females attacking *Bemisia argentifolii* Bellows & Perring infesting cotton, *Gossypium hirsutum* L., and melon, *Cucumis melo* L., were quantified. Adult female behaviors were described and quantified for *Eret.* sp. nr. *californicus* to establish a behavioral time budget analysis. Females readily searched for host whitefly nymphs on cotton leaves with walking speeds averaging 0.5 mm/s. Females remained infrequently on melon leaves; those that did remain and search for hosts averaged walking speeds of 0.33 mm/s. The duration of host assessment by antennation was related to subsequent behaviors. Rejecting a host was a shorter process than accepting it for further evaluation irrespective of plant species or nymphal stage. Probing the margins of the host nymph the ovipositor was repeated less frequently on an individual host on melon leaves than on cotton. Evidence for a behavioral preference for oviposition under early instars was documented for *Eret.* sp. nr. *californicus* females on both plant species. Oviposition for the females that remained and searched for nymphs on leaves in 1-h-long laboratory trials on cotton measured 18%, while on melon, oviposition measured 55%; this higher percentage was attributed to oviposition sites being more accessible under nymphs on melon leaves. Superparasitism was not observed on either host plant species. Twenty-six percent of a female's time on cotton leaves was spent in searching, host assessment, probing, and oviposition, while on melon leaves these behaviors accounted for 44% of the total time. The remainder of the time was spent host feeding, grooming, and resting.

INTRODUCTION

Eretmocerus sp. nr. *californicus* Howard is the principle species of parasitic Hymenoptera attacking *Bemisia argentifolii* in the Imperial Valley, an important agricultural area in southeast California. Percentage parasitism of *Eret.* sp. nr. *californicus* varied greatly among host plant species present in this area (T.S.B., unpublished data). One cause for this variation may be different searching effectiveness on different host plants as shown for *Encarsia formosa* Gahan attacking *Trialeurodes vaporariorum* (Westwood) (van Lenteren *et al.,* 1980; van Lenteren and Woets, 1988; Dowell, 1989).

Comparative studies describing *Eretmocerus* spp. searching behavior on different host plant species have not been reported previously. A study on the effects of two different host plant species, each infested with *Bemisia tabaci* (Gennadius), on the fitness of two populations of *Eretmocerus* sp. was conducted. The two populations, one an arrhenotokous population from southern California and a thelytokous one from Hawaii, showed significant differences in the preimaginal development rate, survival, and fertility between host plant species (Powell and Bellows, 1992).

Research reported in the present paper is part of a larger study which describes searching and ovipositional behaviors of female *Eret.* sp. nr. *californicus* and which quantifies the extent to which these behaviors are influenced by five morphologically and taxonomically diverse plant species. The results for *Eret.* sp. nr. *californicus* behaviors on sweet potato, *Ipomoea batatas* L., a plant with glabrous leaves, serves as the base-

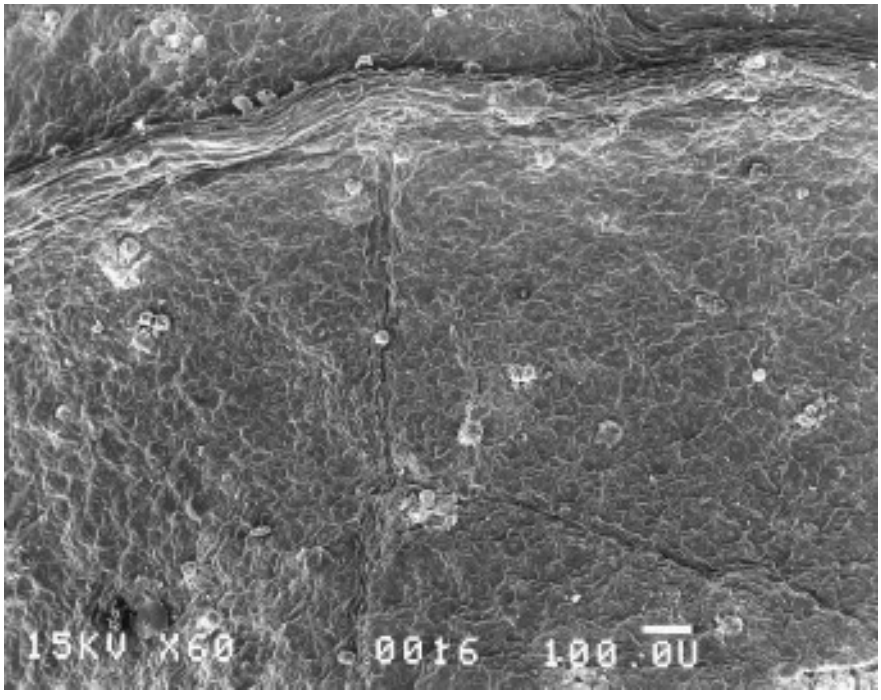

FIG. 1. Scanning electron micrograph of the abaxial surface of a cotton leaf.

line for comparisons among the other plant species (Headrick *et al.*, 1995). The behavioral analyses for *Eret.* sp. nr. *californicus* attacking silverleaf whitefly, *B. argentifolii* Bellows & Perring, on cotton, *Gossypium hirsutum* L. (Malavaceae), and melon, *Cucumis melo* L. (Cucurbitaceae), are presented herein. The leaves of cotton (var.: Delta Pine) are mostly smooth with only a few trichomes (Fig. 1). The leaves of melon are extremely hirsute with large, multicelled trichomes covering the entire leaf surface (Fig. 2).

MATERIALS AND METHODS

Colonies of silverleaf whitefly and *Eret.* sp. nr. *californicus* were maintained on sweet potato at 24 ± 1°C and 60% RH with a 14:10 (light/dark) photoperiod. Noninfested sweet potato plants were maintained in greenhouses at 16–30°C and ca. 60% RH. Silverleaf whitefly colonies were obtained from naturally occurring populations in the Imperial Valley collected from broccoli, melon, and cotton in the fall of 1990 and the summer of 1991. *Eretmocerus* sp. nr. *californicus* colonies were initiated for this study with individuals reared from samples of *Lantana camara* L. infested with *B. argentifolii.* Collections were made on March 1, 1993, at Riverview Cemetary, Western Ave., 3 km N of Hwy 86 (Brawley, Imperial Co., CA). The parasitoid population is arrhenotokous, with a sex ratio of approximately 1:1, and is probably the same population reported earlier from *B. tabaci* from the same area (Powell and Bellows, 1992).

Behaviors of *Eret.* sp. nr. *californicus* females were recorded on videotape for subsequent description and quantification. The video camera used was a Javelin JE3362, the video recorder was a Gyyr time-lapse TLC2051-232, and the monitor was an NEC PM1271A. The camera was mounted on a Zeiss dissecting microscope with Greenough optics. A Javelin JLUX 150 fiber-optic light was used for illumination. Video recordings were made at approximately 40× magnification. *Eretmocerus* sp. nr. *californicus* females, reared in colony, of known age (0–24 h) were allowed to mate and exposed to hosts, for oviposition and host feeding, continuously for 24–48 h before each trial. A single female then was placed on the surface of an excised leaf bearing a known number of whitefly host stages (3.5–10.5/cm^2 on cotton and 3.8–30.2/cm^2 on melon), and then placed adaxial-side down in a glass, 10-cm diameter petri dish. Video recording began and each trial lasted for up to 1 h per female. All trials were conducted between 1000 and 1600 h. The behaviors of 12 females on cotton leaves and 27 females on melon leaves were recorded, yielding a total recording time of 767.2 min. After each trial the presence or absence of parasite eggs was verified by microscopically examining each nymph probed by a female. Video recordings then were analyzed. Each behavior was listed for each female in sequence and the duration of each behavior was recorded

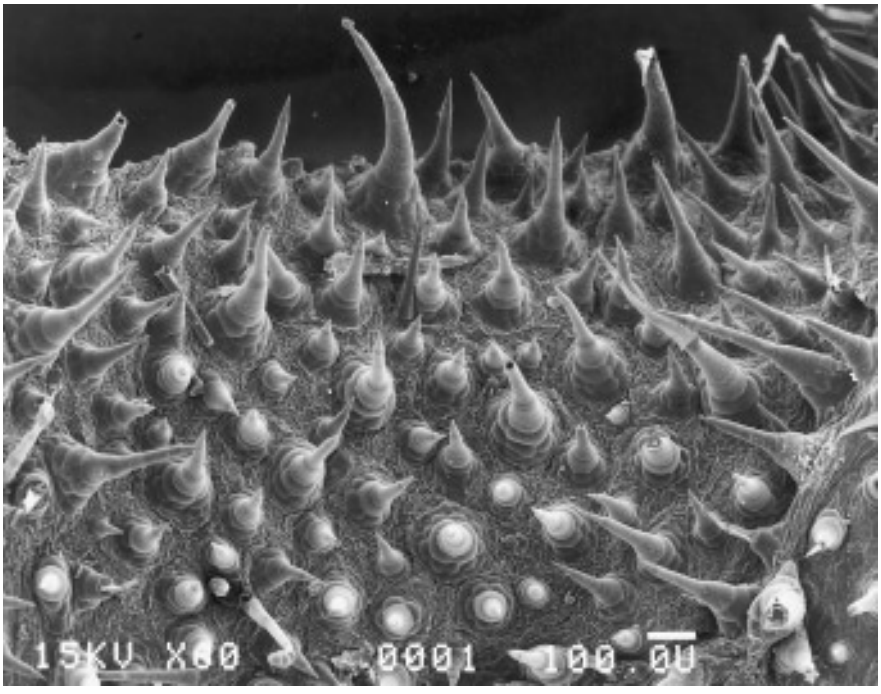

FIG. 2. Scanning electron micrograph of the abaxial surface of a melon leaf.

in seconds. Walking speeds of females were determined by placing acetate sheets over the video monitor, tracing the path of a female, and timing the duration of the walking episode. The relative distance of the path was measured and the absolute distance in micrometers was calculated by use of a stage micrometer.

Behavioral data were entered into a spread-sheet software program (Lotus 123) and collated for statistical analysis in SAS (SAS Institute, 1987). Means are based on total frequencies over all trials for each plant species, unless otherwise noted. Differences among means were examined by analysis of variance (ANOVA) with an observed significance level of 0.05. Differences in frequencies were examined by χ^2 analysis with an observed significance level of 0.05.

Specimens for scanning electron microscopy were fixed in 2% aqueous osmium tetroxide for 24 h, washed in double-distilled water twice, 5 min each, dehydrated in an increasing series of acidulated EtOH, critical-point dried, mounted on stubs, sputter-coated with a gold-palladium alloy, and examined on a JEOL JSM C35 scanning electron microscope in the Department of Nematology, University of California, Riverside. Micrographs were prepared with Polaroid 55P/N film at 15 kV accelerating voltage.

RESULTS

Ethograms for *Eret.* sp. nr. *californicus* females were developed from the recorded sequences. Females typically exhibited behaviors in the following order after being introduced to a leaf: walking on the leaf surface, encountering a host, assessing that host with antennation, and probing the host for oviposition or host feeding; then the process may be repeated. Complexity within this main behavioral pathway occurred through intervening behaviors such as grooming, resting, or resumption of walking. Within the general pathway, frequencies of behavioral sequences were recorded (Figs. 3 and 4). The main behavioral pathway is shown from top to bottom on the left. Arrows are associated with the horizontal lines to the right of each of the main behavioral pathway components and indicate the frequencies of behaviors that led to or were followed by the behavior on the left. For example, in Fig. 3, walking on cotton leaves (W) was followed by grooming 18 times while grooming was followed by walking 51 times over all trials. Antennation was followed by walking 46 times, walking events also led to resting 4 times, and so on. Continuing down the main pathway on the left, walking on cotton leaves led to host encounters (HE) 82% of the time. Host encounters led to arrestment and subsequent circling antennation (CA) 56 times or 41% of all host encounters on cotton leaves led to recognition and assessment of a host. Circling antennation led to an initial probe (P_1) 47% of the time. Initial probes led to successful exsertion of the ovipositor under the host nymph 66% of the time. If an initial probe did not lead to exsertion of the ovipositor, reassessment of the host occurred eight times (Fig. 3, far right, dashed vertical

arrow). Repeated probes (P_R), indicated by dashed lines, followed reassessment by circling antennation 28% of the time and 45% of repeated probes led to successful exsertion under a host nymph. Exsertion of the ovipositor occurred under any given host only once, during which time the decision was made to lay an egg or not. If not, the host was not probed again nor returned to by the same female if searching continued; thus, no superparasitism was observed.

The main pathway on melon began with walking which led to host encounters (HE) 83% of the time (Fig. 4). On melon, host encounters led to circling antennation 74 times or 62% of all host encounters. Circling antennation led to an initial probe (P_1) 82% of the time. Initial probes led to successful exsertion of the ovipositor under the host nymph 90% of the time. If an initial probe did not lead to exsertion of the ovipositor, reassessment of the host occurred only twice (Fig. 4, far right, vertical arrow). Repeated probes (P_R), indicated by dashed lines, followed reassessment by circling antennation 5% of the time and 60% of repeated probes led to successful exsertion under a host nymph.

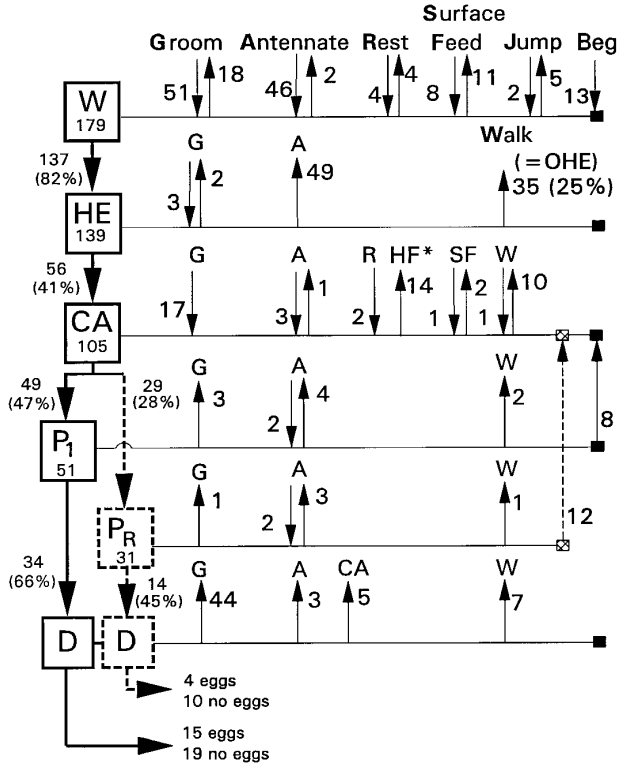

FIG. 3. Ethogram for behaviors of *Eret.* sp. nr. *californicus* attacking *Bemisia argentifolii* on cotton. Arrows indicate subsequent behavioral events and numbers indicate the frequency of observation collated over all trials. The main behavioral pathway begins at the top left and moves down the left margin. The number in a box indicates the total number of events for that behavior, and the sum for the arrows leading to or away from a main behavior are equal. The behavioral loops for host assessment, probing and reassessment, and their frequencies are indicated by the dashed arrows. A, antennation; Beg, beginning (indicating time preceding a walking event at the initiation of a trial); CA, circling antennation; D, disengagement, G, grooming; HE, host encounter; HF*, host feeding loop (which incorporates several behaviors not listed); OHE, oblivious host encounter; P_1, initial probe; P_R, repeat probe; R, resting; SF, surface feeding; W, walking.

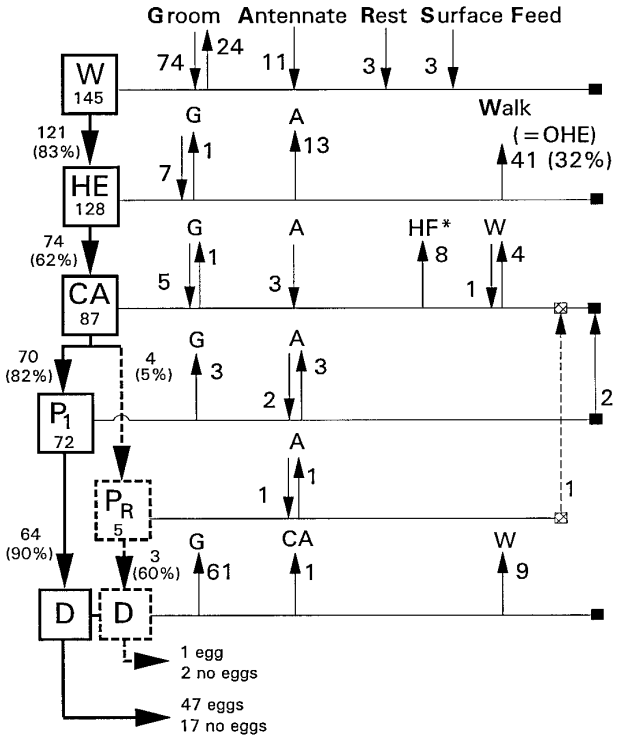

FIG. 4. Ethogram for behaviors of *Eret.* sp. nr. *californicus* attacking *Bemisia argentifolii* on melon. Arrows indicate subsequent behavioral events and numbers indicate the frequency of observation collated over all trials. The main behavioral pathway begins at the top left and moves down the left margin. The behavioral loops for host assessment, probing and reassessment, and their frequencies are indicated by the dashed arrows. Acronyms are the same as in legend to Fig. 3.

All behaviors were analyzed to determine which of these three factors: (1) host nymphal stage; (2) preceding or subsequent behaviors; and (3) host plant species, had any impact on their frequency or duration.

Walking speed. In this study 23% of the females left the surfaces of cotton leaves without exhibiting any searching behavior and on melon 63% of the females left the leaves without any searching. Walking speeds by *Eret.* sp. nr. *californicus* females averaged 0.5 mm/s on cotton, while on melon they averaged 0.33 mm/s, compared to 1.08 mm/s on sweet potato (Headrick *et al.*, 1995).

Host encounters. Searching females walked and asynchronously drummed the apices of their antennae on the surfaces of cotton and melon leaves. When a female came into contact with a whitefly on cotton one

of two events took place. Either the female antennated the host (Fig. 3 "A") or females continued searching without any observable change in behavior to suggest recognition of the host. These "oblivious host encounters" occurred 35 (25%) times over all trials (Fig. 3 "OHE"). Antennation of the host involved continued drumming of the apices of her antennae asynchronously on the dorsum of the host. Antennation was followed by abandonment of the host (walking) or continued assessment of the host by circling antennation (Fig. 3 "CA"). The frequency of host encounters leading to circling antennation on cotton was 56 of 137 (41%) and host encounters led to antennation without circling 49 of 105 (47%) over all trials.

The movements of the females that remained on melon leaves were hampered by the resinous secretions from the glandular hairs and therefore they stopped often to groom. Although searching led to host encounters 121 of 145 (83%) times (Fig. 4), we perceived that females often missed recognizing host nymphs because of the difficulty in maneuvering around the leaf hairs and the accumulated resins and debris on their antennae. On melon, continued searching without any observable arrestment after a host encounter, or an oblivious host encounter, occurred 41 (32%) times (Fig. 4 "OHE") over all trials. The frequency of host encounters leading to circling antennation was 74 of 128 (62%) and host encounters led to antennation without circling only 13 of 128 (10%) times.

Antennation. Circling antennation involved a female standing centrally upon the dorsum of larger whitefly nymphal instars or over smaller instars, and while rotating, antennated the submarginal perimeter of the host, counter-turning frequently (reversing direction of rotation) along the lateral margins between the anteriolateral tracheal folds and the vasiform orifice. On melon leaves, however, assessment of the host nymph by circling antennation was hampered by the presence of the trichomes and females often were unable to perform counter-turning on smaller instar nymphs. Circling antennation was recorded from the initial host contact up to any subsequent behavior, e.g., probing. The circling antennation behavior typically began immediately upon encountering a host and any preliminary antennation could not be distinguished.

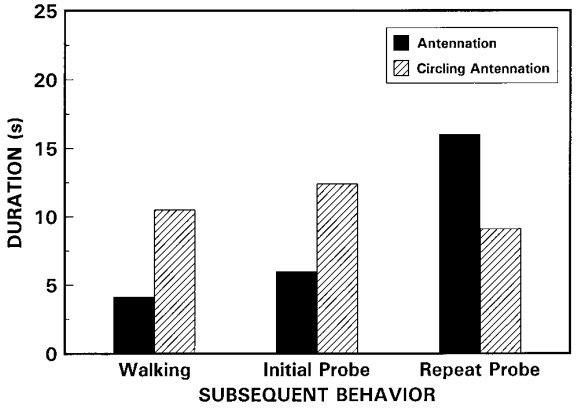

FIG. 5. Mean duration of antennation events on cotton leaves based on the subsequent behavior.

On cotton, circling antennation led to abandonment 10 times and averaged 10.5 ± 2.2 s (range 2.0–24.0 s), led to initial probes 49 (47%) times, and averaged 12.43 ± 0.9 s (range 2.0–31.0 s) with no significant difference ($F = 0.69$, OSL = 0.41). After the initial probe, reassessment of the host by circling antennation before another probe occurred 29 (28%) times and averaged 9.1 ± 0.9 s (range 4.0–22.0 s), and was significantly less than the mean duration of circling antennation leading to an initial probe ($F = 5.72$, OSL = 0.02) (Fig. 5). There were no significant differences detected in the means calculated above among host nymphal stages ($F = 0.68$, OSL = 0.6).

Antennation without circling durations were timed as beginning with initial host contact and ending with abandonment. On cotton, antennation of the host without circling led to abandonment 46 times with a mean duration of 4.15 ± 0.63 s (range 1.0–26.0 s) (Fig. 5). Antennation of the host without circling led to initial probes twice, lasting 1.0 and 11.0 s. Antennation of the host without circling led to repeat probes twice and lasted 16.0 s each (Fig. 5).

On melon, circling antennation by females assessing hosts was disrupted frequently by grooming as resins and debris accumulated on the antennae. On melon, circling antennation led to abandonment four times with a mean of 18.0 ± 6.8 s (range 3.0–31.0 s), led to initial probes 70 times, and averaged 16.2 ± 1.3 s (range 3.0–76.0 s) with no significant differences ($F = 0.10$, OSL = 0.76). After the initial probe, reassessment of the host by circling antennation occurred four times and averaged 14.25 s (range 13.0–16.0 s) and was not significantly different from the mean duration of the initial circling antennation ($F = 0.13$, OSL = 0.72) (Fig. 6).

Antennation of the host without circling on melon led to abandonment 11 times with a mean duration of 11.3 ± 5.8 s (range 1.0–68.0 s). Antennation of the host without circling led to initial probes twice, lasting 5.0 and 7.0 s. Antennation of the host without circling led to repeat probes once for 10 s and was not significantly different from antennations leading to the initial probe (Fig. 6).

Probing and disengagement. The initiation of probing behavior began with a female stepping off the host onto the leaf surface, facing away from the center of the host, and flexing the metasoma upward to expose the appendicular ovipositor. The wings raised ca. 45°

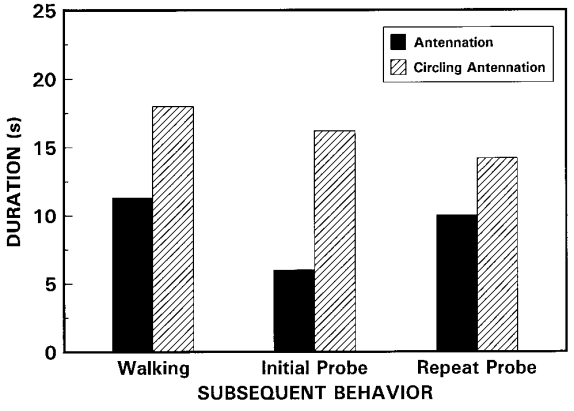

FIG. 6. Mean duration of antennation events on melon leaves based on the subsequent behavior.

above the dorsum and slightly pronated concurrently with metasomal flexure. The apex of the ovipositor was slightly exserted and retracted repeatedly as a female probed against the margin of the host body apparently attempting to find a gap between the host and the leaf surface. The ovipositor was observed to be flexible and bent with the pressure of exsertion.

If a suitable gap between the leaf surface and the host was found, the ovipositor was then fully exserted underneath the whitefly nymph (48 of the 51 hosts probed on cotton, 67 of 72 on melon). Successful exsertion of the ovipositor under a host nymph was accomplished 66% of the time after the initial probe on cotton and 90% of the time on melon. On melon leaves the body of the host nymph was raised above the leaf surface due to the leaf hairs (Fig. 7); thus, exserting the ovipositor under a host nymph on melon was successful on the initial attempt more often than on cotton where the margin of the nymph is nearly contiguous with the leaf surface. The female generally remained quiescent during the time the ovipositor was exserted under the nymph, but slight rhythmic movements of the body were observed just after the female gained purchase and then prior to disengagement.

Disengagement involved a combination of actions which began when the ovipositor was retracted after a variable period of time and females began asynchronously rubbing the hind tarsi against the host and leaf surface. The wings were lowered into their resting position flat over the dorsum. Subsequent to disengagement, females groomed (n = 44 on cotton, n = 61 on melon) or walked away from the host nymph (n = 7 on cotton, n = 9 on melon). Females were not observed to return to any host they had previously probed, thus no superparasitism was recorded.

If a suitable gap between the host and leaf surface was not found during the initial probing, the female then either abandoned the host (n = 2 on cotton) or returned to antennation of the host's margin (n = 8 on cotton, n = 2 on melon). If circling antennation of the same host took place after an initial probe it was always followed by another probing attempt (Figs. 3 and 4, dashed vertical line leading from CA to P_R), either on the same side as the initial probe on or the side opposite. Females irregularly alternated between sides during repeated probing events until a suitable gap was accessed (n = 14 successes on cotton, n = 3 on melon) or the host was abandonded (n = 1 on cotton).

Probing duration was defined for the purposes of this

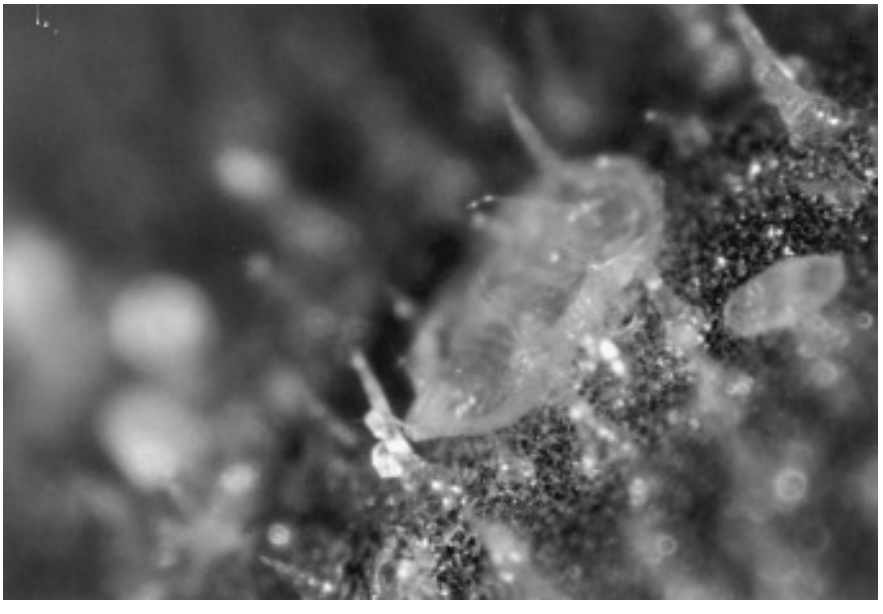

FIG. 7. Photograph of a fourth instar nymph of *B. argentifolii* on abaxial surface of a melon leaf.

study as the initial engagement of the ovipositor until subsequent antennation or abandonment. The durations of initial and repeat probes were analyzed. On cotton, initial probes averaged 60.6 ± 8.04 s (n = 51, range 4.0–298.0 s). Repeat probes averaged 33.9 ± 6.6 s (n = 31, range 4.0–155.0 s); these means were significantly different in duration (F = 5.33, OSL = 0.02). An initial or repeat probe that led to oviposition on cotton averaged 77.8 ± 11.5 s (n = 18, range 24.0–224.0 s) and 70.5 ± 6.4 s (n = 4, range 53.0–80.0 s), respectively, and were not significantly different (F = 0.1, OSL = 0.76). No differences were detected between initial and repeat probes leading to oviposition among whitefly nymphal stages (F = 1.59, OSL = 0.23). An initial or repeat probe that did not result in oviposition averaged 51.0 ± 10.5 s (n = 33, range 4.0–298.0 s) and 28.5 ± 6.9 s (n = 27, range 4.0–155.0 s), respectively, with no significant difference (F = 2.92, OSL = 0.09). There were no significant differences detected between initial and repeat probes, leading to unsuccessful oviposition among whitefly nymphal stages (F = 0.93, OSL = 0.47).

On melon, initial probes averaged 89.1 ± 9.9 s (n = 70, range 10.0–475.0 s). Repeat probes averaged 74.2 ± 25.5 s (n = 5, range 10.0–141.0 s); these means were not significantly different in duration (F = 0.15, OSL = 0.7). Initial probes that led to oviposition on melon averaged 102.4 ± 14.3 s (n = 47, range 48.0–475.0 s). Only one repeat probe led to oviposition and this probed lasted for 141 s. Initial probing durations were longer on fourth instars than on first instar nymphs (F = 4.0, OSL = 0.0075). An initial or repeat probe that did not result in oviposition averaged 62.9 ± 6.7 s (n = 24, range 10.0–128.0 s) and 57.5 ± 24.9 s (n = 4, range 10.0–118.0 s) and were not significantly different (F = 0.09, OSL = 0.76). There were no stage-related differences among initial and repeat probes that did not lead to oviposition (F = 1.86, OSL = 0.15).

Oviposition. On cotton, egg deposition was observed to be near the center of the host irrespective of nymphal stage. Probing and oviposition by females on melon were affected by the presence of the large leaf hairs. Females were observed to probe in areas where there were no host nymphs, under crawlers, which ultimately moved away, and nearby first instars, indicating that the typical stimuli from the host was disrupted, possibly by the fluids and debris accumulated on their antennae. Eggs were laid under crawlers on two occasions, and an egg was laid beside a first instar nymph, instead of under it. A female retracted her ovipositor and began rubbing her hind tarsi asynchronously against the host and leaf surface after egg deposition on cotton, and the wings lowered into their resting position flat over the dorsum. Disengagement behavior on melon was hampered by the leaf hairs.

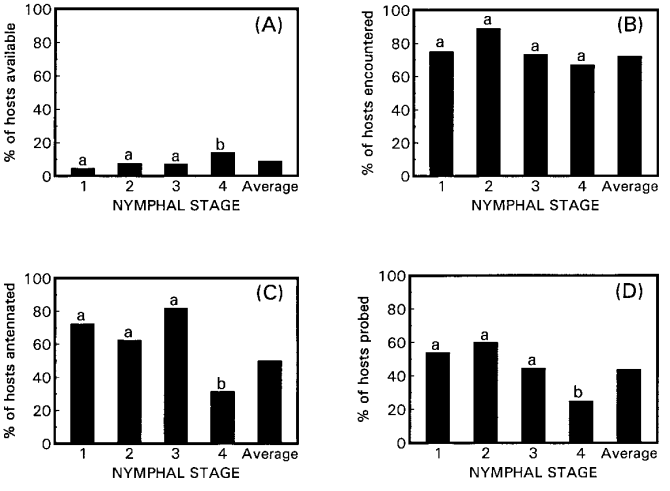

FIG. 8. Frequencies, represented as a percentage, of behavioral events leading to oviposition by nymphal instar on cotton; columns in each figure with different letters differed in frequency with an OSL ≤ 0.05; weighted averages are included. (A) Of the numbers available, those which were encountered; (B) of those encountered, those which were antennated; (C) of those antennated, those which were probed; (D) of those probed, those which received an egg.

Females frequently groomed during probing and oviposition on melon. Grooming following disengagement occurred 83% of the time on melon, 78% of the time on cotton.

Probes that resulted in full exsertion of the ovipositor beneath the host did not indicate successful oviposition. Oviposition occurred under 21 of 48 (44%) nymphs examined for egg deposition on cotton. Oviposition occurred under 48 of 67 (72%) nymphs examined on melon. Probes that resulted in egg deposition were the longest in duration on both cotton and melon.

Oviposition by *Eret.* sp. nr. *californicus* females also was analyzed for frequency and host nymphal stage effects. A total of 21 eggs were laid over all trials on cotton, 4 of 12 females laid eggs, and the maximum for 1 female was 8 eggs. A total of 48 eggs were laid over all trials on melon, 9 of 27 females laid eggs, and the maximum for 2 females was 10 eggs.

The frequencies of various oviposition behaviors among the different nymphal stages was examined. On cotton, first instar nymphs represented 34.5% of the total stages present in all trials, second instars represented 15.8%, third instars 13.9%, and forth instar nymphs made up 35.8% of all stages present. Of these, 4.65% of first instar nymphs were encountered; 7.6% of second instars, 7.2% of third instar nymphs, and 14.21% of fourth instar nymphs were encountered by searching female wasps (Fig. 8A). Of the 4.65% first instar nymphs encountered, 75.0% were subsequently antennated, while 88.9, 73.3, and 67.1% of the second, third, and fourth instars, respectively, were subsequently antennated (Fig. 8B). Of the first instars anten-

nated, 72.2% were then probed with the ovipositor, 62.5% of the second instars antennated were subsequently probed, and 81.8 and 31.4% of the thirds and fourths, respectively, were probed (Fig. 8C). Of the first instars probed, 53.85% ultimately had an egg laid under them, and 60.0, 44.4, and 25.0% of second, third, and fourth instar nymphs, respectively, had eggs laid under them (Fig. 8D). The differences that occurred among the frequency of nymphal stages available and the frequency of them being encountered by a female searching were highly significant ($\chi^2 = 31.32$, $df = 3$, OSL = 0.0); fourth instar nymphs were found more frequently than expected (Fig. 8A). The average proportion of stages encountered on cotton was 8.9%. There were no stage-related differences between the frequency of host stage encountered and the frequency of that stage being antennated (mean = 72.2%, $\chi^2 = 3.58$, $df = 3$, OSL = 0.31) (Fig. 8B). Of the nymphs antennated, there was a significantly lower frequency of fourth instars probed than any other stage ($\chi^2 = 16.08$, $df = 3$, OSL = 0.0001); there were no significant differences among first, second, and third instar nymphs ($\chi^2 = 1.2$, $df = 2$, OSL = 0.54). Finally, of nymphs probed with the ovipositor, there was a significantly lower frequency of fourth instars receiving an egg ($\chi^2 = 3.89$, $df = 3$, OSL = 0.27). There were no significant differences in the frequency of oviposition among first, second, and third instar nymphs ($\chi^2 = 0.46$, $df = 2$, OSL = 0.79). Thus, females enountered fourth instar nymphs during searching with more frequency than other stages. However, females arrested on all stages with approximately the same relative frequency. Females did discriminate among stages when probing with the ovipositor, probing fourth instar nymphs much less frequently than the other stages. Similarly, fourth instar nymphs received eggs with much less frequency than other stages.

On melon, first instar nymphs represented 54.3% of the total stages present, second instars represented 23.6%, third instars 10.2%, and forth instar nymphs made up 11.9% of all the stages present. Of the nymphs available, 8.4% of the first instars, 6.2% of the second instars, 10.8% of the third instars, and 19.6% of the fourth instars were encountered by searching females (Fig. 9A). Of the 8.4% first instar nymphs encountered, 64.5% were subsequently antennated, while 90.0, 73.3, and 53.1% of second, third, and fourth instars, repectively, were subsequently antennated (Fig. 9B). Of the first instars antennated, 62.5% were then probed with the ovipositor, 88.9% of the second instars were probed, and 90.1 and 82.35% of thirds and fourths, respectively, were probed (Fig. 9C). Of the first instars probed, 92.0% ultimately had an egg laid under them, and 56.25, 90.0, and 42.9% of second, third, and fourth instar nymphs, respectively, had eggs laid under them (Fig. 9D). There were significantly more fourth instar nymphs encountered by searching females ($\chi^2 = 25.07$, $df = 3$, OSL = 0.00001); the average proportion of stages encountered on melon was 9.4% (Fig. 9A). Although fourths were encountered with greater frequency than other stages, significantly fewer were subsequently assessed by antennation ($\chi^2 = 8.49$, $df = 3$, OSL = 0.037); the remaining stages were antennated with the same relative frequency as their relative abundance (Fig. 9B). The differences that occurred among the frequency of nymphal stages antennated and the frequency of them being probed with the ovipositor were slightly significant ($\chi^2 = 7.26$, $df = 3$, OSL = 0.064); fewer first instar nymphs were subsequently probed than expected (Fig. 9C). Of the nymphs probed with the ovipositor, second and fourth instar nymphs received eggs with significantly less frequency than first or third instars ($\chi^2 = 23.74$, $df = 3$, OSL = 0.0). Thus, females encountered fourth instar nymphs on melon leaves with greater frequency than expected. Of the nymphs encountered, females arrested and antennated fourth instar nymphs less frequently than other nymphal stages. However, all stages were subsequently probed with the ovipositor with the same relative frequency, except first instar nymphs were probed slightly less frequently. Females also oviposited eggs under fourth instars less frequently than the earlier stages; of the remaining stages, seconds received fewer eggs than either first or third instars nymphs.

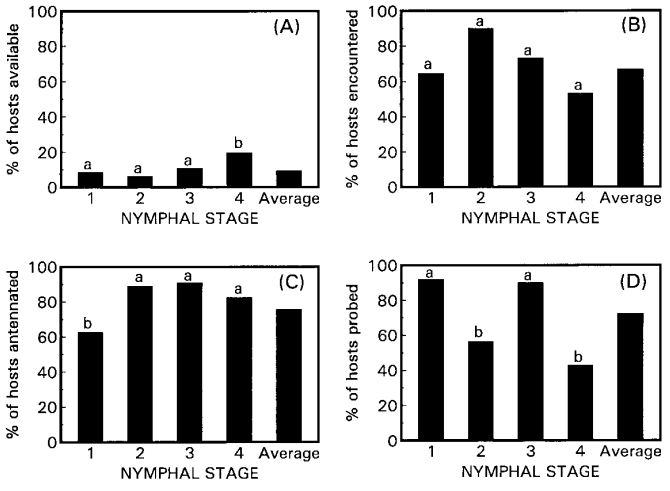

FIG. 9. Frequencies, represented as a percentage, of behavioral events leading to oviposition by nymphal instar on melon; columns in each figure with different letters differed in frequency with an OSL $\leq$ 0.05; weighted averages are included. (A) Of the numbers available, those which were encountered; (B) of those encountered, those which were antennated; (C) of those antennated, those which were probed; (D) of those probed, those which received an egg.

Feeding, grooming, resting. Feeding was divided into two categories, host feeding and surface feeding. Surface feeding involved arrestment at and antennation of a potential food or water source, then extending the antennae away from the face and lowering

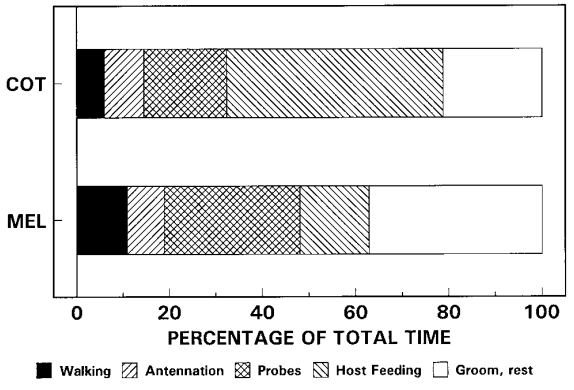

FIG. 10. Mean duration of behavioral feeding, grooming, and resting events by *Eret.* sp. nr. *californicus* on cotton and melon.

the mouthparts to the food or water source. Further antennation of the food or water source occurred sporadically during feeding episodes. Surface feeding events on cotton averaged 25.4 ± 8.1 s (n = 23, range 1.0–133.0 s), while surface feeding events on melon averaged 31.25 ± 6.8 s (n = 4, range 18.0–47.0 s) (Fig. 10). Females also were observed to feed upon accumulated honeydew or water droplets condensed on the upper surfaces of nymphs on cotton; this, however, was not host feeding.

Host feeding by females involved assessment of the host by circling antennation, alignment along the long axis of the host body facing the vasiform orifice, stepping forward to engage the ovipositor in the crevasses of the vasiform orifice, penetrating the host integument, exserting and retracting the ovipositor inside the host's body, and then turning to feed upon the accumulated hemolymph. Host feeding times were recorded from initiation through cessation of feeding on the hemolymph. Host feeding events on cotton averaged 291.0 ± 117.26 s (n = 32, range 1.0–2927.0 s) (Fig. 10). On cotton, females host fed on first instars in 17 of 32 (53%) events; host feeding on second, third, and fourth instar nymphs occurred 12.5, 6, and 28% of the time, respectively. Host feeding durations by stage were analyzed. Host feeding durations on first instars averaged 47.35 ± 16.9 s (range 1.0–249.0 s). Although the fewest host feeding events occurred with second and third instars, they were the longest in duration, averaging just over 1000 s each, and the longest host feeding event was 2927 s on a second instar nymph. Host feeding on fourth instars averaged 198.12 ± 128.39 s (range 1.0–971.0 s). Host feeding by individual females also was analyzed for cotton. Seven of 12 females displayed host feeding on cotton. Host feeding ranged from 2–15 events in a 1-h trial. Two of the females spent the majority of time (ca. 2500–3000 s) host feeding in the 1-h trials.

Host feeding on melon averaged 360.42 ± 117.26 s (n = 7, range 4.0–966.0 s). Females host fed on first instars in 3 of 7 (43%) events; host feeding on second and fourth instar nymphs occurred 28% of the time for each. Third instar nymphs were not fed upon. Host feeding durations on first instars averaged 237.3 ± 52.2 s (range 133.0–293.0 s). Host feeding on second instars lasted 594 and 966 s; the latter was the longest host feeding event recorded in the melon trials. Host feeding on fourth instars lasted 4.0 and 247.0 s. Host feeding by individual females also was analyzed in the melon trials. Three of 27 females displayed host feeding, ranging from 2–3 events per female in a 1-h trial.

Grooming was observed most frequently following an ovipositional episode, but could occur at any time and could be sustained while other events were happening such as antennation or walking, but was exclusive of feeding. Grooming involved use of the fore legs to clean the anterior portion of the thorax, the head, and antennae. The hind legs were used to clean the wings, metasoma, and ovipositor. The fore legs were rubbed together for cleaning and together cleaned the middle legs; the hind legs were rubbed together for cleaning. Grooming episodes averaged 32.1 ± 5.8 s (n = 59, range 1.0–219.0 s) on cotton and 78.8 ± 8.4 s (n = 98, range 1.0–426.0 s) on melon (Fig. 10).

Resting was rarely observed during the trials on cotton (n = 7) and melon (n = 7). The mean duration of resting on cotton was 47.14 ± 17.7 s (range 2.0–116.0 s) (Fig. 10). The mean duration of resting on melon was 93.57 ± 46.18 s (range 8.0–362.0 s) (Fig. 10).

Figure 10 shows the overall behavioral time budget analysis for *Eret.* sp. nr. *californicus* females attacking hosts on cotton and melon leaves in all trials. Twenty-six percent of the total time on cotton and 44% of the total time on melon was spent in walking, antennation, and probing, while the remaining time was spent host feeding, grooming, and resting. Host feeding was the most time consuming activity on cotton, taking up to 53% of the total time; however, this value is largely due to the exceptionally long host feeding events by 2 of the 12 females. Host feeding was significantly less on melon (18%) than on cotton; however, grooming on melon accounted for 38% of the total time, and on cotton, only 21% of the time was spent in grooming.

DISCUSSION

Host plant morphology has been shown to impact the searching efficiency of aphelinid parasitoids by slowing or inhibiting their movements (Hua *et al.*, 1987; Gerling, 1990). Van Lenteren and de Ponti (1990) determined that walking speeds were one of the main factors influencing parasitism efficiency of *Enc. formosa* attacking *T. vaporariorum* on various ornamentals in greenhouses. We found in the present study that walking speeds for *Eret.* sp. nr. *californicus* were relatively

fast on glabrous leaves and relatively slow in hirsute leaves, but that host plant morphology influenced other behaviors such as propensity to commence searching and ability to antennate, recognize, and probe a host—all of which also ultimately affect oviposition frequency.

Data from the silverleaf whitefly survey in the Imperial Valley in southeastern California show that percentage parasitism on melon is extremely low (T.S.B., unpublished data). In the laboratory, most (63%) females introduced onto melon leaves for the behavioral trials left the leaf without displaying any searching behavior. This may be indicative of the field situation, with females initially avoiding plants with leaves that bear numerous hairs or glandular trichomes. Searching and subsequent behaviors expressed in the laboratory of the females that remained on melon leaves may have been elicited due to initial contact with hosts.

The frequency of assessing an encountered host nymph by circling antennation was significantly different among melon, cotton, and sweet potato (Headrick *et al.,* 1995). Host encounters were less frequent on melon compared to either cotton or sweet potato, but antennation followed host encounters 62% of the time. Similarly, larger proportions of hosts assessed by antennation on melon were subsequently probed for oviposition. Thus, for females on melon, once a host was recognized, engaging in subsequent behaviors on that host was more frequent than on either cotton or sweet potato. This indicates that once a host is recognized as suitable in an environment that makes the process of searching and assessment difficult, the likelihood of continued assessment and subsequent utilization increases.

The results of this study also showed that ovipositions per hosts assessed were higher on melon (55%) than on cotton (18%) or sweet potato (15%), a result which appears contradictory to the field survey data (T.S.B., unpublished data). However, this may be explained by (1) the fact that many females leave a melon plant shortly after having landed on it and (2) the mechanics of oviposition by *Eret.* sp. nr. *californicus* females. The body of the host nymph on sweet potato lies flat and its margins are nearly contiguous with the smooth leaf surface; whereas, on hairy-leafed plants, such as melon, the body of the nymph is raised above the leaf surface and its margins curled upward (Fig. 7). The frequency of initial probing events that led to successful exsertion of the ovipositor under the host on smooth-leaved plants was low compared to the frequency of ovipositor exsertions under the host that required multiple probing events. Thus, finding a suitable place to exsert the ovipositor beneath the host nymph on a glabrous leaf is more difficult than to do so on a plant with hirsute leaves. This most likely accounts for the higher percentage parasitism and more successful probing attempts that occurred on melon leaves in laboratory trials, once searching and assessment behavior was elicited.

Circling antennation times were consistently longer on melon than on cotton or sweet potato (Headrick *et al.,* 1995). Thus, duration of circling antennation that led to abandonment of the host was longer on melon than on either cotton or sweet potato (Headrick *et al.,* 1995). This suggests that the presence of leaf hairs and exudates on melon leaves adversely affects the female's ability to recognize cues normally detected from hosts on other plant species.

Probing durations also were longer on melon irrespective of whether it was an initial or repeat probe or followed by oviposition. No differences or clear trends were detected in probe durations based on the host nymphal stage. Lakin and Bellows (in press) noted that *Eret.* sp. females spent more time probing fourth instar nymphs of citrus whitefly, *Dialeurodes citri* (Ashmead), in trying to find a suitable place for oviposition due to their larger size. Probing durations and frequencies have not been reported for any other species of *Eretmocerus.*

Circumstantial evidence for instar preference in *Eret. californicus* was noted by Gameel (1969) and Gerling (1966a) based on life history analyses. In the present study females did not show a clear preference for probing under second instar nymphs as shown for females on sweet potato (Headrick *et al.,* 1995). However, in the present study females discriminated against older instars for oviposition irrespective of how many more they encountered, which contrasts with the general assumptions that later instars are preferred because they are larger and thus easier to locate (cf. Gerling, 1990). Other aphelinids, e.g., *Enc. formosa* and *Enc. luteola,* appear to prefer later instar nymphs for oviposition (Nell *et al.,* 1976; Headrick and Bellows, unpublished data), or only early instars, e.g., *Amitus hesperidium* and *A. bennetti* (Clausen and Berry, 1932; T.S.B., unpublished data). Similar preferences for oviposition into later instars was noted for *Eret.* sp. (Lakin and Bellows, in press).

There were no discernible differences in searching behaviors leading to oviposition or host feeding among the host plants in this study, and this was also noted for sweet potato (Headrick *et al.,* 1995). During the 1-h trials on cotton, females spent most of their time involved in either ovipositional behaviors or host feeding. The relatively long period associated with host feeding on cotton is attributed to the three females that spent up to 50 min each host feeding. Similar results were reported for females on sweet potato. For females on melon, considerably more time was involved in walking and grooming. This difference can be related to the difficulty in walking on melon leaves and the process of grooming associated with the accumulation

of plant exudates. Durations of antennation events leading to host feeding did not differ significantly among host plants or host nymphal stages, which contrasts with the findings of van Lenteren *et al.* (1980) for *Enc. formosa*. Host feedings by females on cotton were more numerous than on any other plant species ($n = 31$) and showed a distinct preference for first instars as reported for sweet potato (Headrick *et al.*, 1995). However, host feeding by females on melon was infrequent ($n = 7$) and showed no stage preferences. *Encarsia deserti, Enc. formosa, Enc. lahorensis, Enc. pergandiella,* and *Enc. transvena* were also reported to show distinct nymphal instar preferences for host feeding (Gerling *et al.*, 1987; van Alphen *et al.*, 1976; van Lenteren *et al.*, 1980; Viggiani and Mazzone, 1978; Gerling, 1966b; 1983, respectively), whereas other *Eretmocerus* spp. apparently do not (Gerling, 1990). Further research is needed to elucidate the role of the host plant and its affect on feeding frequencies, durations, and host stage preferences. As reported for females on sweet potato, host searching and oviposition occurred directly before and after host feeding, and no host was observed to be used for both oviposition and host feeding, which is consistent with findings for other aphelinids attacking Aleyrodidae (cf. Nell *et al.*, 1976; van Lenteren *et al.*, 1980; Gerling, 1990).

Van Lenteren and de Ponti (1990) stated that host plant resistance and biological control are foundational to an integrated approach to pest management. Leaf morphology, i.e., leaf hairs and glandular trichomes, play an important physical and chemical deterrent role in host plant resistance to herbivore feeding. However, relatively little research has been conducted on describing and quantifying the interrelationships among the host plant, herbivore, and natural enemy with the aim to inhibit the herbivore and enhance the natural enemy. Van Lenteren and de Ponti (1990) cited examples of research which showed that increased numbers of glandular hairs makes certain plants more resistant to herbivores, but also adversely affected the natural enemies, while in other plants increased numbers of hairs limited one herbivore species while other species flourished. Intermediate or partial resistance has been found to give overall management by partially limiting the herbivores and allowing the natural enemies to proceed (van Lenteren and de Ponti, 1990). However, there are also data which show that plants bred for partial resistance have an adverse effect on natural enemy effectiveness because certain chemicals attractive to the natural enemies are modified or no longer present, or the microclimate afforded by the leaf morphology has been altered (van Emden, 1986). Van Lenteren and de Ponti (1990) point out that breeding for resistance does not automatically lead to better overall pest control.

The results of the present study and the silverleaf whitefly survey (T.S.B., unpublished data) show that host plant attractiveness to natural enemies can affect percentage parasitism greatly, and leaf morphology can profoundly affect parasite efficiency. As seen on melon leaves, parasite probing efficiency on nymphs is high due to the ease of accessability under the host, but that, if given the choice, the majority of female *Eret.* sp. nr. *californicus* do not remain to search for hosts on these leaves.

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