

**ENCARSIA FORMOSA GAHAN
(HYMENOPTERA: APHELINIDAE)
PARASITIZING BEMISIA ARGENTIFOLII
BELLOWS & PERRING (HOMOPTERA:
ALEYRODIDAE): BEHAVIORAL ANALYSIS
OF TWO GEOGRAPHIC POPULATIONS**

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Abstract

Behavioral evaluation of Greece and Nile strains of *Encarsia formosa* Gahan (Hymenoptera: Aphelinidae), parasitizing the silverleaf whitefly, *Bemisia argentifolii* Bellows & Perring (Homoptera: Aleyrodidae), were performed on Hibiscus (*Hibiscus rosa sinensis*) in the laboratory under four temperature regimes: 16°, 21°, 26°, and 31°C. Behaviors evaluated included: (1) walk-ing, (2) drumming, (3) preening, (4) ovipositor probing, (5) parasitization, (6) stationary, (7) feeding and (8) walking off the lower leaf surface.

Analysis of the mean time and mean percent time that each *E. formosa* strain spends in each behavior, mean frequency of behaviors, and percentage of individuals performing the specified behaviors, all indicated that the *E. formosa* strains represent two climatic-ecotypes, with the Greece strain optimally adapted to a cooler climate (21°C), and the Nile strain optimally adapted to a warmer climate (26°-31°C). This appears to be the first behavioral evidence for two strains of *E. formosa* based upon temperature optima. Biological control implications are discussed.

Introduction

The sweetpotato whitefly, *Bemisia tabaci* (Gennadius), is a polyphagous pest species in the tropics and subtropics throughout the world, infesting approximately 506 host plant species in 74 families (Greathead 1986). This whitefly species causes damage by the removal of plant sap during feeding (Lopez-Avila & Cock 1986), the transmission of numerous viruses and virus-like agents (Brunt 1986), and the excretion of honeydew that becomes a nutrient medium for sooty mold fungus (Horowitz et al. 1984). While *B. tabaci* can still be found in Mexico

(Brown 1992), *Bemisia argentifolii* Bellows & Perring, n. sp.- = the B strain of *B. tabaci* (Perring et al. 1993, Bellows et al. 1994), appears to have largely displaced *B. tabaci* A-strain since 1990 (Nuessly & Perring 1995). Control of pest species in the genus *Bemisia* through the use of insecticides has been very difficult due to increasing pesticide resistance (Byrne et al. 1990, Ditttrich et al. 1990, Prahabker et al. 1992), as well as difficulties associated with proper coverage on the underside of the leaves where the nymphs feed (Bellows & Arakawa 1988).

Introduced whitefly species often become severe pests, and several genera have been effectively controlled through classical biological control programs (Onillon 1990). Therefore, as an introduced pest species in the United States, biological control of *B. argentifolii* offers an environmentally sound and effective means for limiting this pest species. Recently, *B. argentifolii* has inflicted astronomical losses in agricultural and horticultural areas of varying climatic conditions across southern United States (Faust 1992). Similarly, exotic parasitoid species which may be potentially useful in *B. argentifolii* population regulation within the United States will likely be indigenous to areas with vastly different climates. Therefore, as an integral part of the search and evaluation of exotic parasitoid species for control of *B. argen-tifolii*, consideration of climatic-species adaptation or ecotypes is imperative (Onillon 1990), and is the focus of the study reported herein.

Prospects for biological control of *B. argentifolii* using parasitoids remain largely unexplored except for several recently reported studies (Parrella et al. 1991, Heinz & Parrella 1994a,b, Simmons & Minkenberg 1994, Headrick et al. 1995). In glasshouses, however, *Encarsia formosa* Gahan has been essentially the only parasitoid species investigated for control. As a solitary, thelytokous, cosmopolitan endoparasitoid species, *E. formosa* was originally described from collections in Ohio where it was found associated with the greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood)(Gahan 1924). Speyer (1927) subsequently suggested the possibility of using *E. formosa* to control *T. vaporariorum*, and biological control was practiced using *E. formosa* until 1945 when it was replaced by synthetic, organic pesticides. In the early 1970's *E. formosa* use was revived and has steadily increased. Noldus & van Lenteren (1990) reported that *E. formosa* is applied to about 3800 ha of greenhouse crops worldwide.

The bionomics and behavior of *E. formosa* have been studied in association with *T. vaporariorum* and *B. tabaci* (Arakawa 1982, Enkegaard 1993), and other studies have more specifically addressed the effects of temperature on *E. formosa* (Lenteren & Hulsapas-Jordaan 1988, Enkegaard 1993, Roermund et. al. 1993 & 1994). However, similar studies are lacking in association with *B. argentifolii*. While few studies have been reported on different strains or

ecotypes of *E. formosa* (Christochowitz et. al. 1981, Lenteren & Hulspas-Jordaan 1988, Heinz & Parrella 1994b, Heinz et.al. 1994), no investigations have been reported on the differential effects of temperature on various strains of *E. formosa*.

Therefore, in 1993 we undertook a comparative study of the bionomics and behavior of two wild populations of *E. formosa*, as potential parasitoids of *B. argentifolii* under specific climatic conditions. We are investigating the hypothesis that there may be climatic-adaptations or ecotypes of *E. formosa* such that the climate in its geographic origin strongly influence its bionomics and behavior, or predisposes different populations of *E. formosa* to specific climatic regimes. The specific study reported herein is limited to evaluation of the effects of temperature on key behavioral parameters of two populations of *E. formosa*, which are indigenous to areas differing significantly in climatic conditions.

Materials and Methods

Parasitoids Uniparental strains of *E. formosa* were collected from two environmentally distinct geographic locations: (1) one strain from *Trialeurodes vaporariorum* (Westwood) on *Phaseolus vulgaris* (common bean) in Angelohori, Greece (Greece strain, M92017); (2) the other strain from *B. tabaci* on *Lantana camora* in Cairo, Egypt (Nile strain, M92030). Cultures were maintained on *B. argentifolii*-infested *Hibiscus rosa sinensis* plants (var. Kona Pink) at 27°C and 18:6 L:D photoperiod. Parasitoids had been in culture for 36-42 generations.

Whiteflies Colonies of *B. argentifolii* were maintained in pure culture on hibiscus plants (var. Brilliant Red) at 27°C and 14:10 L:D photoperiod.

Protocol Under identical environmental conditions as those utilized during evaluation, parasitoids were allowed to emerge and were then held for 24h on SLWF-infested hibiscus leaves (25-35 third instar nymphs/leaf) in a 120mm X 100mm petri plate and provided honey. This allowed for acclimation and conditioning. Individual parasitoids were then observed for 20 minutes, and a total of 30 individuals of each strain were evaluated under each of four temperatures (16°, 21°, 26°, and 31°C). Tests were performed between 8:30 am and 2:30 pm CST at Stoneville, Mississippi (33° 26'N latitude; 90°55'W longitude).

Behavioral Parameters Behaviors evaluated were those considered to be involved in short range host location, host examination and parasitization (Alphen & Vet 1986). These included: (1) walking, (2) drumming, (3) preening, (4) ovipositor probing, (5) parasitization, (6) stationary, (7) feeding, and (8) walking off the lower leaf surface.

Drumming or antennation is defined as tapping the antennae against a substrate (leaf surface or host) at a 90° angle. Those individuals recorded as walking were nearly always drumming as well. Those individuals recorded as drumming were stationary. Preening or grooming was generally preceded by feeding, but also occurred intermittently with walking. In both ovipositor probing and parasitization, the parasitoid assumes a posture in which the abdomen is tucked underneath the body of the parasitoid and the ovipositor is inserted into the host, while the antennae are generally stationary at ca. a 45° angle. However, ovipositor probing is defined as ovipositor insertions of less than 60 seconds, while parasitization is ovipositor insertions of greater than 60 seconds. Initiation of antennation while ovipositor probing or during parasitization generally signaled the termination of these behaviors and the removal of the ovipositor from the host. Feeding occurred on both honeydew and hosts, but both responses are grouped together herein as feeding. Parasitoids walking off the lower leaf surface either walked onto the upper leaf surface where there were no hosts, or off the leaf entirely. Behaviors were evaluated based upon: (1) mean time, (2) mean percent time, (3) mean frequency, and (4) percent parasitoids performing specified behaviors. For the latter, χ^2 tests ($P \leq 0.05$) were used to compare temperatures.

Results and Discussion

Mean period of time and mean percent period of time that each *E. formosa* strain spent in each behavior (Figures 1 & 2, respectively), mean frequency of behaviors (Figure 3), and percentage of individuals performing the specified behaviors (Figure 4) show that: (1) for the Greece strain, walking, drumming, ovipositor probing and parasitization are maximal at 21°C, standing still is minimal at 21°C, and maximal at 26°C and 31°C, and walking off the leaf is highest at 31°C; and (2) for the Nile strain, drumming, ovipositor probing and parasitization are optimal at 26°C and 31°C, feeding and stationary are minimal at 16°C, and walking off the leaf is highest at 16°C.

These data show that each *E. formosa* strain is differentially adapted to different climatic conditions, with the Greece strain adapted to a cooler climate (21°C), and the Nile strain adapted to a warmer climate (26°C-31°C). Therefore, these results indicate the existence of a climatic-species adaptation in *E. formosa*, which when coupled with our life history results (unpubl.), leads to the conclusion that these 'ecotypes' are adapted to precise ecological conditions. These data correlate well with the climatic conditions which exist in the geographic regions of origin of each *E. formosa* strain (Lacey & Kirk 1993), where the annual mean monthly minimum and maximum temperatures for Greece are ca. 8.75°C and 9.09°C, respectively, lower than in Egypt.

Messenger & van den Bosch (1971), discussed climatically adapted strains of parasitoids and presented specific cases which demonstrate their potential utility in biological control programs. For example, the French and Iranian strains of *Tioxys pallidus* (Halliday) have been successfully integrated into control programs of the walnut aphid [*Chromaphis juglandicola* (Kaltenbach)] in California, with the former in coastal and intermediate zones, and the latter in northern zones. Therefore, the two *E. formosa* ecotypes reported herein, as well as other wild populations, may offer alternatives to the present *E. formosa* commercial strain, and may have potential utility under various ecological conditions, such as in particular geographical regions, or at different times of the year within a region where climatic conditions change over the season, as well as in cool glasshouses.

It should be noted that most of the laboratory-reared *E. formosa* currently distributed throughout Europe may have originated from a few individuals introduced to England (Nemec & Stary 1984), and *E. formosa* seems to be very uniform, genetically, even relative to other Hymenoptera which are characterized by low genetic variability (Graur 1985). The limited success of *E. formosa* against *B. tabaci* (Parrella et al. 1991, Ruisinger & Backhaus 1994) and whitefly species other than *T. vaporariorum*, as well as the limited or reduced capacity of *E. formosa* to control whitefly species under low temperature conditions (Milliron 1940, Vet 1980, Vet & van Lenteren 1981, Christochowitz et al. 1981, Vianen & van Lenteren 1986, Lenteren and Hulspas-Jordaan 1988, Yano 1989), may be due in part to this ever narrowing genetic diversity of commercially utilized *E. formosa*. Therefore, one possible solution to this narrow genetic base may be the identification of climatic ecotypes from wild populations, as reported herein, which may enhance the limited genetic diversity of the present *E. formosa* commercial strain.

Finally, these data show that walking, drumming, ovipositor probing and parasitization are optimal at the most suitable temperature(s), and may be among the more discriminating behavioral parameters which are indicative of parasitoid efficacy, and that under similar timed evaluations of parasitoid behaviors, the percentage of parasitoids performing specified behaviors may be one of the more informative methods of behavioral analysis.

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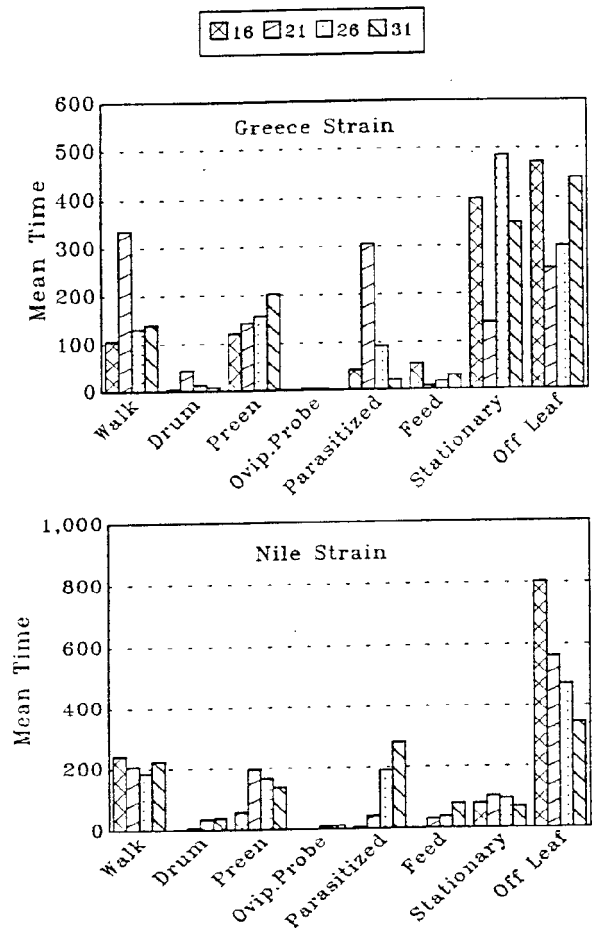


Figure 1. Mean Time (seconds) *Encarsia formosa* Displayed Behaviors at Four Temperatures.

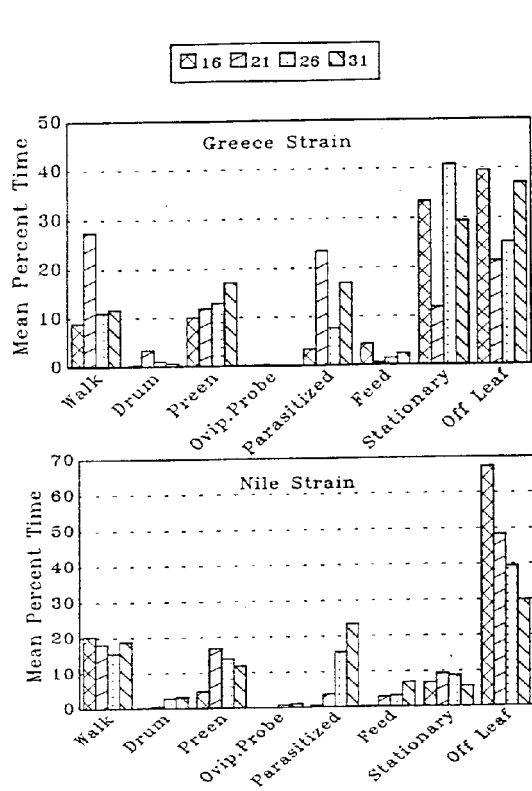


Figure 2. Mean Percent Time *Encarsia formosa* Displayed Behaviors at Four Temperatures.

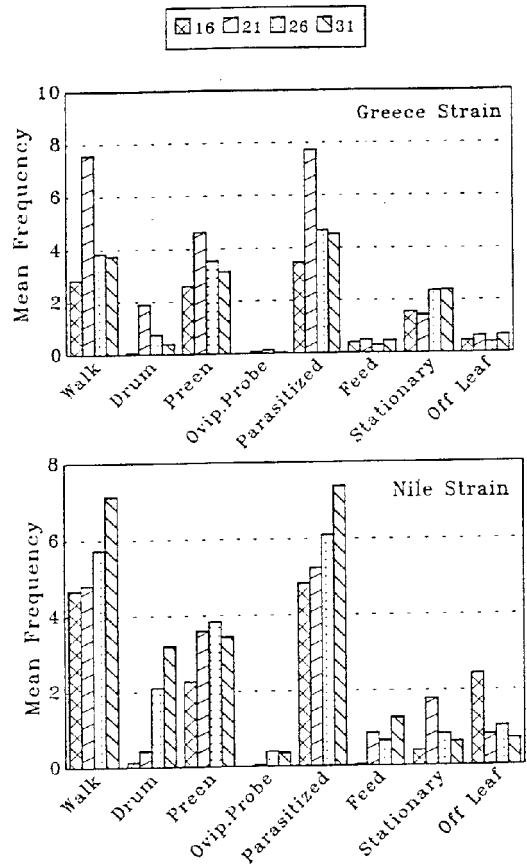


Figure 3. Frequency at which *Encarsia formosa* Displayed Behaviors at Four Temperatures.

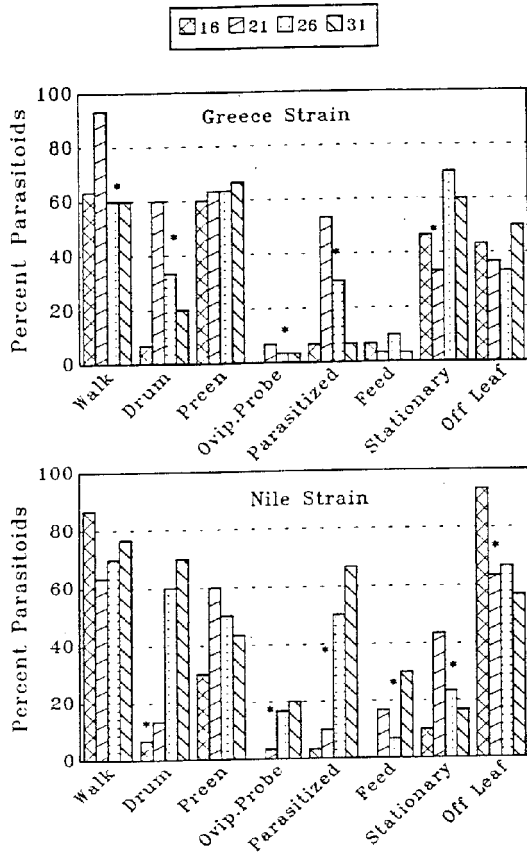


Figure 4. Percent *Encarsia formosa* Performing the Specified Behaviors at Four Temperatures [* = significant temperature effect χ^2 tests $P \leq 0.05$].