# Interactions of the Whitefly Predator *Delphastus pusillus* (Coleoptera: Coccinellidae) with Parasitized Sweetpotato Whitefly (Homoptera: Aleyrodidae)

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**ABSTRACT** Delphastus pusillus (LeConte), an indigenous coccinellid predator of whiteflies, including sweetpotato whitefly, Bemisia tabaci (Gennadius), feeds on all stages of whiteflies. In laboratory tests, fourth-instar and adult female D. pusillus exhibited a marked tendency to avoid fourth-instar B. tabaci parasitized by the aphelinid endoparasitoids Encarsia transvena (Timberlake) and Eretmocerus sp. nr. californicus Howard in favor of unparasitized whiteflies. The age of the parasitoid within the whitefly influenced the extent of the avoidance. Whiteflies with first-instar parasitoids were not avoided, but those containing third instars and pupal parasitoids were significantly avoided. The presumed causes of the avoidance include parasitism-induced hardening of the whitefly cuticle and introduction of air into the whitefly around maturing parasitoids. D. pusillus and parasitoids tend to attack different stages of the whitefly. This attribute increases temporal separation, enhancing the options for the use of D. pusillus in pest management programs in conjunction with parasitoids.

KEY WORDS Delphastus pusillus, Aphelinidae, predation

Delphastus pusillus (LeConte) is a native coccinellid predator often associated with large populations of arboreal whiteflies in Florida (Muma 1956, Cherry & Dowell 1979). It also feeds readily on the sweetpotato whitefly, *Bemisia tabaci* (Gennadius), and has been considered a potential biocontrol agent of this insect (Parrella et al. 1992, Hoelmer et al. 1993), which has become a major pest of numerous crops in many parts of the United States (Perring et al. 1991).

Adult *D. pusillus* are small, shiny, black beetles 1.3–1.4 mm in length; the larvae and pupae are pale yellowish white. Adult beetles usually feed and oviposit on leaves with high densities of whitefly eggs. As a result, *D. pusillus* feeds primarily on eggs, although all stages of *B. tabaci* may be attacked and consumed (Hoelmer et al. 1993). Adult and immature beetles feed by piercing the integument of their prey and extracting the contents. Death of a whitefly caused by feeding can easily be distinguished from death by other causes. Complete consumption will leave only a flattened, empty whitefly cuticle, whereas an incompletely consumed whitefly will exhibit visibly disrupted internal organs. During studies of the feeding behavior and reproduction of *D. pusillus*, we noticed that fourth-instar whiteflies containing late instars of *Encarsia transvena* (Timberlake), a solitary aphelinid endoparasitoid often found attacking *B. tabaci* in central Florida, appeared to suffer less from predation than did unparasitized whiteflies. The ability of a predator to avoid parasitized prey and to select unparasitized prey is a useful attribute that could facilitate its use in management programs in conjunction with parasitoids. In this paper, we document this avoidance and selection behavior with laboratory studies.

## **Materials and Methods**

Insectary colonies of *D. pusillus* were started with feral beetles collected from various locations in central Florida and maintained in a greenhouse on *B. tabaci* reared on *Phaseolus limensis* Macfady 'Henderson', *Euphorbia pulcherrima* Willd. ex Klotzsch, *Hibiscus rosa-sinensis* L., and on papaya whitefly, *Trialeurodes variabilis* (Quaintance) reared on *Carica papaya* L.

Laboratory studies were conducted at  $28 \pm 3^{\circ}$ C with a 12:12 (L:D) h photoperiod. Polystyrene petri plates (8 mm by 30 mm) were used as experimental arenas. Immature whiteflies were

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provided as prey on leaf disks cut from bean or hibiscus leaves with a cork borer.

High humidity was maintained in the arenas to prevent the leaf disks from curling and dislodging the whiteflies or hiding them from the beetles. This was accomplished by one of two methods. In early experiments, a disk of moistened filter paper was placed in each petri plate and the leaf disk placed on the paper. In later experiments, petri plates were partially filled with a layer of 1% agar. Methyl parabenzoate (methyl P-hydroxy-benzoate) was added to the agar gel to retard growth of bacterial and fungal contaminants. A 10-mm-diameter circular hole was cut in the lid and covered with organdy mesh to allow air movement and prevent condensation that could trap beetles.

Unparasitized whiteflies offered as prey were fourth-instar *red-eyed nymphs*. At this stage of nymph development, red compound eyes are clearly visible, but no wing pad development is apparent. Whitefly nymphs of this age are more convex in profile than early fourth instars, similar to nymphs containing parasitoid larvae or pupae. Aphelinid parasitoids develop within fourthinstar whiteflies, although their eggs may have been deposited in younger stages (Viggiani 1984).

Previous studies (Hoelmer et al. 1993) have shown that an average of 11–12 young fourthinstars are eaten per day per adult *D. pusillus*. To give beetles sufficient prey items to choose among, we offered each predator 20 whiteflies. One *D. pusillus* was placed in an arena with 10 unparasitized and 10 parasitized fourth-instar whiteflies, which were placed in four rows of five nymphs with parasitized and unparasitized whiteflies alternating in a uniform distribution. Both adult and larval beetles were tested. Only female adults were tested; the sex of larval beetles was not determined. Only larvae and adults who were observed feeding in the insectary were used in the experiments.

Studies with Adult Beetles. Three developmental stages of E. transvena within prey were offered to adult D. pusillus: (1) first-instar, detectable externally only by a displacement of one or both of the host's paired yellowish mycetomes (n = 16 trials); (2) third instar, visible through the host integument as a C-shaped body occupying a major part of the host (n = 26); (3) pupal parasitoids, with the black pupal cuticle visible through the host cuticle (n = 10). To determine if beetle response to another parasitoid species was comparable, adult beetles were also exposed to pupae of *Eretmocerus* sp. nr. californicus Howard (n = 10), which is another aphelinid parasitoid commonly attacking *B. tabaci* in Florida (Hoelmer & Osborne 1990).

After 24 h, the beetles were removed from the arenas, and each petri plate was examined with a stereomicroscope. The number of unparasitized

whiteflies still alive, killed by the beetles, or dead from other causes was recorded. If there was any uncertainty about the cause of death, the individual was scored in the other causes category. Damage to parasitized whiteflies was categorized the same way; third-instar and pupal parasitoids were observed closely to detect movement verifying they were still alive. E. transvena pupae rock from side to side periodically; if no visible damage to pupae was apparent, inspection was continued as long as several minutes to see if these movements occurred. E. sp. nr. californicus pupae do not exhibit this movement; therefore, they were inspected closely for signs of cuticle damage and localized feeding.

To determine if beetle avoidance decreased with exposure time, some of the beetles were left in the arenas with the remaining uneaten whiteflies for longer exposure without providing new prey. Ten adult *D. pusillus* were kept in arenas for a second day of exposure to pupal *E. transvena*; and nine adult beetles were kept an additional 3 d with first-instar *E. transvena*. These beetles were removed on day 2 or 4, respectively, and the condition of the prey was recorded as above.

Studies with Beetle Larvae. Unparasitized whitefly and pupal *E. transvena* were offered to fourth-instar *D. pusillus* (n = 12 trials) using the same arenas and experimental design as above. Younger larvae were not tested because earlier work (Hoelmer et al. 1993) demonstrated their preference for smaller whiteflies at younger stages and avoidance of those at older stages.

Data were recorded as percentages of available prey attacked and subjected to arcsine transformation prior to statistical analysis. Transformed data were analyzed with paired difference *t*-tests; significance was assessed at P = 0.05. Summary statistics are shown for untransformed data.

### **Results and Discussion**

Individual beetles consumed a total of between four and eight unparasitized and parasitized whiteflies each day, which was less than in our earlier study (Hoelmer et al. 1993). However, beetles in that study were given young fourth instars instead of older, less preferred, red-eyed nymphs.

**Response of Adult Beetles to Parasitized Whiteflies.** Our initial observations suggesting that *D. pusillus* tended to avoid parasitized whiteflies were supported by our laboratory experiments. The age of the developing parasitoid within the whitefly influenced the extent of the avoidance behavior (Table 1). Parasitized whiteflies containing young larval parasitoids were not avoided. Mean consumption by adult beetles of these whiteflies was greater than unparasitized ones, although the difference was marginally sig-

Parasitoid stage	Prey attacked			
	n (replicates)	Mean	SD <sup>a</sup>	P <sup>b</sup>
	Adult D. pusi	llus		
Egg/first-instar E. transvena	16	3.50	2.48	0.0582
Unparasitized control	16	2.31	2.63	
Third-instar E. transvena	26	1.54	1.84	0.0002
Unparasitized control	26	3.31	3.10	
Pupal <i>E. transvena</i>	10	0.00	0.0	0.0001
Unparasitized control	10	4.00	2.45	
Pupal E. sp. nr. <i>californicus</i>	10	1.70	1.34	0.0002
Unparasitized control	10	7.00	3.43	
	Fourth-instar D. ‡	ousillus		
Pupal <i>E. transvena</i>	12	1.00	2.26	0.0085
Unparasitized control	12	4.17	3.04	

Table 1. Paired comparisons of predation on unparasitized and different stages of parasitized *B. tabaci* fourth instars during 24-h exposure to individual adult or fourth-instar *D. pusillus*.

" Each replicate consisted of 10 parasitized and 10 unparasitized whiteflies.

 $^{b}$  P values shown are for paired comparison *t*-tests, arcsine-transformed data as proportion of available prey attacked. Means and SD are given untransformed.

nificant (P = 0.0582). Eight of 16 beetles tested attacked more parasitized whiteflies, 4 attacked equal numbers of each, and 4 attacked more unparasitized whiteflies. Of the nine beetles kept with the uneaten whiteflies for an additional 3 d, the percentage of remaining whiteflies attacked was similar for parasitized (47.6%) and unparasitized (56.7%) whiteflies.

Avoidance of whiteflies containing older parasitoids increased significantly as the age of the parasitoid increased. Whiteflies containing thirdinstar parasitoids were significantly less likely to be attacked (P < 0.0002) than unparasitized whiteflies, although avoidance was not absolute. Eleven of 16 beetles killed more unparasitized than parasitized whiteflies, 3 killed more parasitized ones, and 2 attacked equal numbers of each.

None of the 10 beetles tested fed on any of the pupal *E. transvena* (P < 0.0001), while they attacked 40% of the unparasitized whiteflies. Moreover, when 10 beetles were left in the test arenas with the remaining whiteflies for an additional 24 h, parasitized whiteflies were also completely avoided. Of the remaining unparasitized whiteflies, 48.4% were consumed.

Avoidance of pupal *Eretmocerus* sp. nr. californicus was significant, although not absolute as with *E. transvena*. All beetles tested, excluding one that did not feed, attacked more unparasitized than parasitized whiteflies, but these beetles all attacked some whiteflies with parasitoid pupae as well.

**Response of Larval Beetles to Parasitized** Whiteflies. Fourth-instar *D. pusillus* also avoided pupal *E. transvena* and fed instead on unparasitized whiteflies (Table 1). Only 1 of the 12 larvae tested fed on more parasitized than unparasitized whiteflies. Six did not attack parasitoid pupae; one larva did not feed.

In similar fashion, Delphastus sp. feeding on wooly whitefly, Aleurothrixus floccosus (Maskell), in Central America, the Dominican Republic, and the Yucatan Peninsula in Mexico was observed avoiding whiteflies parasitized by several species of aphelinids (J. Quezada, personal communication). Quezada & DeBach (1973) also found that the vedalia beetle, Rodolia cardinalis (Mulsant), feeding on cottony cushion scale, Icerya purchasi Maskell, exhibited similar discrimination and avoidance of prey parasitized by Cryptochaetum iceryae (Williston). The beetles would starve if left with only scales containing pupae of the parasitic fly. It has been suggested (Quezada & DeBach 1973; Quezada, personal communication) that the discriminatory feeding exhibited by these predators may be related to the relative specificity of their diets compared with predators having more varied diets.

The avoidance of whiteflies with advanced parasitism exhibited by D. pusillus is most likely a result of the extraoral digestion often found in small coccinellid predators (Savoiskaya 1960). Following penetration of the host cuticle by D. *pusillus*, fluids are gradually sucked out of the prey, with periods of regurgitation that apparently help to stir up and digest the contents of the prey. As parasitoid larvae develop within whitefly nymphs, physiological changes harden the whitefly cuticle. This may make cuticle penetration more difficult for small predators. In addition, as a parasitoid larva grows and approaches pupation, the fluid contents of the whitefly are used up and air spaces form inside the cuticle. These air spaces are likely to interfere with fluid uptake by D. pusillus. Both phenomena probably influence beetle choice by making it more difficult to obtain food from whiteflies containing advanced stages of parasiFebruary 1994

toids. The differences in avoidance of E. transvena and E. sp. nr. californicus pupae support this suppostion; pupae of E. transvena are more compact than those of E. sp. nr. californicus, with a larger air space between the pupal and host cuticle.

In naturally occurring field populations of whiteflies, the impact of avoiding parasitism is probably not great because beetles concentrate their feeding among high densities of eggs, while parasitoids oviposit and develop in older instars. The greatest impact on parasitoid populations would most likely be in low density whitefly populations, when greater proportions of older parasitized whitefly instars would be discovered by beetles searching for prey.

Only the portion of immature parasitoids in the egg or early instar stage would be at significant risk from predation by *D. pusillus*; as larvae grow, the risk of predation would decrease significantly. There are insufficient data to conclude that hunger does not influence avoidance of parasitized prey, but the results of the 2-d exposure to pupal *E. transvena* suggest that avoidance does not diminish quickly.

These results suggest that because the beetles and parasitoids tend to attack different stages and because parasitized whiteflies are avoided by *D*. *pusillus*, there is potential for integration of both types of natural enemies into management programs for whiteflies. Similar examples of complementary natural enemies include Nephaspis amnicola Wingo and Encarsia nr. haitiensis Dozier in Hawaii to control Aleurodicus dispersus Russell (Kumashiro et al. 1983), and Serangium parcesetosum Sicard and Encarsia lahorensis (Howard) in Corsica for control of Dialeurodes citri (Ashmead) (Malausa et al. 1988).

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