Influence of Entomopathogenic Fungi on *Serangium parcesetosum* (Coleoptera: Coccinellidae), an Important Predator of Whiteflies (Homoptera: Aleyrodidae)

TADEUSZ J. POPRAWSKI, JESUSA CRISOSTOMO LEGASPI,[1] AND PAUL E. PARKER[2]

Beneficial Insects Research Unit, USDA-ARS Subtropical Agricultural Research Center, 2413 East Highway 83, Weslaco, TX 78596

Environ. Entomol. 27(3): 785-795 (1998)

ABSTRACT The lethal and sublethal effects of the entomopathogenic fungi *Beauveria bassiana* (Balsamo) Vuillemin and *Paecilomyces fumosoroseus* (Wize) Brown & Smith against the coccinellid predator *Serangium parcesetosum* Sicard were studied in the laboratory. We also tested if the ingestion of whiteflies contaminated with *B. bassiana* affected predator survivorship in 3 tests: (1) *S. parcesetosum* larvae were fed contaminated whiteflies for a 10-d period; (2) larvae were fed 1 time-only prey contaminated 24-, 48-, 72-, or 96-h previously; and (3) larvae were fed prey after the conidia were washed off the leaves and prey cuticles. The predator had significantly lower survivorship when sprayed with *B. bassiana* than with *P. fumosoroseus*. However, survivorship was not affected by the dosage rates for each pathogen. Survivorship curves for *P. fumosoroseus* treatments also did not differ significantly from blank and carrier controls. Mean larval duration was longest (≈22.5 d) in *S. parcesetosum* sprayed at the medium and high dosages of *B. bassiana*, intermediate (≈20 d) for the low dosage of *B. bassiana*, and lowest (≈18 d) for the blank and carrier controls and the *P. fumosoroseus* treatments. The pupal stages averaged 6.6-8.0 d. Mean adult body weights ranged from 0.97 mg (*B. bassiana* low dosage) to 1.54 mg (*P. fumosoroseus* medium dosage), but were not significantly different. Analysis of cumulative predation showed that predators sprayed with *P. fumosoroseus* consumed prey at a rate similar to that of the controls (≈130 prey daily per predator), which was significantly higher than that of predators sprayed with *B. bassiana* (≈60 prey daily per predator). Again, dosage was not a significant factor. Feeding on *B. bassiana*-contaminated prey caused ≈86% mortality in *S. parcesetosum* immatures, compared with ≈13% in the controls. Prey contaminated 24-, 48-, 72-, and 96-h previously induced mortalities of 92.5, 71.4, 71.4, and 44.4%, respectively. Washing conidia off the leaves and the cuticle of whiteflies did not result in lowered mortality of the predator relative to the other treatments.

KEY WORDS *Serangium parcesetosum, Beauveria bassiana, Bemisia,* biological control, microbial control, compatibility

THE WHITEFLY *Bemisia argentifolii* Bellows & Perring (Homoptera: Aleyrodidae) is the most important new agricultural pest of the last decade (Henneberry et al. 1993). Economic losses occur from direct feeding damage, honeydew contamination and associated sooty molds, and transmitted viruses. Crop damage attributed to this new pest was estimated at $1 billion in 1992 in the United States alone, primarily concentrated in the southern region of the country (Lacey et al. 1993). Many other areas of the world also are experiencing a dramatic increase in economic impact of *B. argentifolii* (Cock 1993, De Barro 1995). Because the insect is expected to continue to cause widespread and significant damage in many areas of the world, environmentally sound and sustainable methods of control are needed.

Previous experience with other whitefly pests demonstrates that biological control may substantially contribute to the sustainable management of the damage caused by *B. argentifolii* in both greenhouse and field-cropping environments. Although most whitefly biological control research is directed toward insect parasitoids and predators (Gerling 1990, 1996; Onillon 1990; Nordlund and Legaspi 1996), several entomopathogenic fungi have potential as microbial control agents against these pests (Fransen 1990), including *B. argentifolii* (Lacey et al. 1996). Fungi are the only entomopathogens able to invade actively through the cuticle, an advantage against piercing—sucking insects such as Homoptera, and all known pathogens of Aleyrodidae are fungi. Of the fungal species known to infect whiteflies, only *Aschersonia aleyrodis* Webber, *Verticillium lecanii* (Zimmerman) Viégas, *Beauveria bassiana* (Balsamo) Vuillemin, and *Paecilomyces fumosoroseus* (Wize) Brown & Smith (Deuteromyco-

This article reports the results of research only. Mention of a proprietary product does not constitute an endorsement by the USDA nor by the Texas A&M University System.

[1] Texas Agricultural Experiment Station, 2415 East Highway 83, Weslaco, TX 78596.

[2] Mission Plant Protection Center, USDA-APHIS Plant Protection and Quarantine, Moore Air Base, P.O. Box 2140, Mission, TX 78572.

tina: Hyphomycetes) have been commercialized or are currently being developed as microbial agents against *Bemisia* and other whiteflies under greenhouse conditions (Lacey and Fransen 1994, Steinberg and Prag 1994). In addition, *P. fumosoroseus* and *B. bassiana* have been registered as biopesticides for control of Homoptera, including *Bemisia*, in field crops in the United States and Mexico.

Bemisia whiteflies are attacked by a variety of predatory arthropods (Nordlund and Legaspi 1996). Although little investigated, larvae and adults of *Serangium parcesetosum* (= *Catana parcesetosa*) Sicard (Coleoptera: Coccinellidae) are considered important predators of whiteflies (Antadze and Timofeyeva 1975, Timofeyeva and Nhuan 1978, Kuchanwar et al. 1982, Kapadia and Puri 1989, Yigit 1992). *S. parcesetosum* was imported from India into the United States in 1993 (Lacey et al. 1993) and released from quarantine by the USDA–APHIS Plant Protection and Quarantine, Mission Plant Protection Center at Mission, TX, in subsequent years. Recently, Legaspi et al. (1996) evaluated its potential in the laboratory as a biological control agent of *B. argentifolii*.

Currently, there is an incomplete understanding of the potential effects microbial insecticides might have on nontarget invertebrates in natural systems. Predicting the ecological host range (as opposed to physiological host range) is particularly difficult with self-perpetuating organisms that function at the tertiary trophic level, such as entomopathogens and predatory insects.

Mycoses in nature have been observed in a number of predatory insects (Goettel et al. 1990); however, little is known about their epizootiology and resultant effect on predators and other nontarget species. An exception is the common occurrence of *B. bassiana* epizootics in hibernating adult *Semiadalia undecimnotata* (Schneider), *Coccinella septempunctata* L., and *Adalia bipunctata* (L.) (Coleoptera: Coccinellidae), especially under conditions of high humidity (Iperti 1966, Mills 1981, Goettel and Jaronski, 1997). Many species of predatory insects seem refractory to fungal infection when challenged in the laboratory (Goettel et al. 1990). The effects of entomopathogenic fungi on coccinellids have been little studied and this, only recently. Most of these studies have been laboratory screenings primarily aimed at comparing infectivity and pathogenicity of the fungi to the coccinellids, and developing or standardizing bioassay protocols (Magalhães et al. 1988; James and Lighthart 1992, 1994; Tillemans et al. 1992; Todorova et al. 1994). However, effects of insect control agents (see Brooks [1993] for entomopathogens and Croft [1990] for chemical pesticides) other than direct kill also may inhibit the beneficial capacity of nontarget natural enemies of pests.

Although many reports are available on the short-term, detrimental effects of entomopathogenic fungi on nontarget organisms (Goettel et al. 1990), reports on indirect effects are few. The goals of this laboratory study were to determine the lethal and nonlethal effects of *B. bassiana* and *P. fumosoroseus* on *S. parcesetosum*, and the suitability of *B. bassiana*-contaminated whiteflies as prey for *S. parcesetosum*.

Materials and Methods

Insects. A cohort of *S. parcesetosum* larvae was obtained from eggs supplied by the USDA–APHIS Plant Protection and Quarantine at Phoenix, AZ. They were reared on eggs and 1st-instars of *B. argentifolii* infesting leaves of *Hibiscus rosa-sinensis* L. 'Kona Pink', or of eggplant, *Solanum melongena* L. 'Florida Market 50', obtained from the USDA–APHIS Mission Plant Protection Center at Mission, TX. Rearing was conducted in environmental growth chambers at 25°C, 50–55% RH, and under a photophase of 16:8 (L:D) h. Three-d-old larvae (1-d-old 2nd instars) were used in the tests.

Fungi. *B. bassiana* (strain GHA, lot 921114) and *P. fumosoroseus* (strain 612, lot 940322) were provided by Mycotech, Butte, MT, as technical, unformulated, dry conidial powders containing 8.7×10^{10} (*B. bassiana*) and 1.2×10^{11} (*P. fumosoroseus*) conidia per gram with >90% claimed viability. *P. fumosoroseus* strain 612 originated from *B. argentifolii* (Weslaco, TX, in 1993). *B. bassiana* strain GHA is under commercial development and information on its origin is therefore restricted.

Effect of Direct Sprays of Conidia of *B. bassiana* and *P. fumosoroseus* on Mortality and Predation Rate of *S. parcesetosum* Larvae. Test fungi were applied to larvae as 1-ml aliquots of conidial suspensions by using a Potter precision spray tower (Burkard Manufacturing Co. Ltd, Rickmansworth, Hertfordshire, England) equipped with a fine spray nozzle operating at a pressure of 0.7 kg/cm^2. A 1-ml aliquot of conidial suspension applied in the tower gives a deposition of 0.01 μl of conidial suspension per square millimeter, corresponding to a theoretical field application volume of ≈100 liters/ha of flat surface. The activity of each fungal strain on *S. parcesetosum* was assessed using 3 application rates (dosages), and 20 insects per dosage. The high dosage (≈1,000 conidia per square millimeter, simulating field rates of $\approx 10^{13}$ conidia per hectare) was prepared by suspending 15 mg of conidia in 10 ml of water with 0.02% Silwet L-77 (an organosilicone nonionic surfactant; Loveland Industries, Greeley, CO). A 5-fold and 25-fold dilution in 0.02% aqueous Silwet was used as the medium and low dosage, respectively. Conidial suspensions were vortexed for 5 min and used immediately. Actual conidial dosages were determined for each fungus from blocks of 1.5% agar placed in the spray tower target arena. Four 0.05-mm^2 microscope fields (400×) were scanned on each block of agar, conidia were counted, and counts were averaged and expressed as dosages applied per square millimeter. Sabouraud dextrose agar plates were sprayed at the time of treatment, incubated for 24 h at 25°C, and conidial germination was assessed using a microscope (400×). A random sample of 100 conidia per plate was taken 3 different times and conidia were scored for germination, counts were

averaged, and percentage of germination calculated. This latter value was then used to transform dosage applied per square millimeter to adjusted dosage per square millimeter (e.g., number of viable conidia actually used in each assay). Blank controls (no treatment) and carrier controls (0.02% aqueous Silwet) were included with each fungal treatment.

Treated and control larvae were then isolated individually in vented plastic petri dishes (4 cm diameter). Each larva was provided with a known number (≈320) of eggs, 1st instars, or both of *B. argentifolii* on an eggplant leaf, and incubated for 24 h at 25°C and 100% RH under a photophase of 16:8 (L:D) h. Thereafter, larvae were maintained under similar temperature and light regimes, but at 50–55% RH. At 24-h intervals, the leaf was replaced with a fresh leaf infested with a known number of prey.

Dead insects were removed daily from the dishes, surface-sterilized in 0.13% Zephiran chloride (benzalkonium chloride; Winthrop, NY) for 1 min, rinsed twice in sterile distilled water, and finally plated on 2% agar supplemented with 0.5% gentamycin. The plates were incubated at 25°C for 48 h and cadavers were scored for overt mycosis (sporulation). Differences in the proportion of larvae surviving between the treatments were determined using chi-square tests.

After 13 d (onset of pupation in the blank control), all surviving larvae were fed ad libitum until they pupated. Additional measurements taken included duration of the larval and pupal stages, and body weight of adults 24 h after emergence. The duration of the larval stage of insects that survived treatment was calculated starting with 1-d-old 2nd instars and ending with pupation. The data were subjected to analysis of variance (ANOVA).

Daily prey consumption was recorded until larval death or pupation. A regression model was used to define cumulative predation as a function of time and treatment, by using treatment as a categorical variable (general linear model [GLM] analysis, Systat package version 5.2; Wilkinson et al. 1992). Percentage of reduction in predation was calculated by $R = ([P_c - P_t]/P_c)\ 100$, where R is percentage of reduction, P_c is predation by control larvae, and P_t is predation by fungus-treated larvae. Larvae that died during the first 24 h of incubation or larvae that escaped from the petri dishes during the tests were not included in the above analyses.

Suitability of Prey Contaminated by *B. bassiana* Conidia for *S. parcesetosum*. *Test A: Continuous Exposure to Prey Contaminated 24-h Previously.* Twelve eggplant leaves each infested with ≈800 1st instars of *B. argentifolii* were sprayed with 1-ml aliquots of a *B. bassiana* suspension. A 2-fold concentrated high dosage was applied using the Potter spray tower. Adjusted conidial counts were determined as previously described. Twelve control leaves were sprayed with 1 ml of aqueous 0.02% Silwet. Leaves were allowed to air-dry before incubation for 24 h at 25°C and 100% RH under a photophase of 16:8 (L:D) h. Leaves were then placed individually into vented plastic petri dishes (15 cm diameter), and 5 randomly picked *S. parcesetosum* larvae were introduced into each dish. The experimental units (dishes) were maintained at 25°C and 50–55% RH under a photophase of 16:8 (L:D) h. After 24 h the leaves were replaced with new leaves that had been treated 24 h earlier. This protocol was repeated daily for 10 d, at which time the 1st pupae were found in the control units. Thereafter, dishes still containing larvae were provisioned with untreated leaves. Dead insects were processed as previously described. Larvae that died during the first 24 h and escapees were not included in the chi-square tests performed on these data. The test was terminated when the last adult emerged. In addition to larval mortality, the number of emerged adults was recorded.

To calculate the mean time of mortality, the number of larvae, prepupae, and pupae that died each day after treatment was divided by the total mortality after 10 d. This value was then multiplied by the respective day. Values for day 1–10 were summed to produce a weighted average time of mortality.

Percentage of reduction in number of emerged adults was calculated by $R = ([A_u - A_t]/A_u)\ 100$, where R is percentage of reduction, A_u is number of adults developing from larvae fed on untreated prey, and A_t is number of adults developing from larvae reared on *B. bassiana*-treated leaf or prey.

Standard leaves were used to determine levels of mycosis in *B. argentifolii*. In addition to the above treatments, 12 infested leaves were sprayed with the same dosage, and in the same manner, as the 1st-d treatment in the assay. *B. bassiana* infection was easily diagnosed by the presence of a red pigment in the hemocoel of infected whitefly nymphs that persisted until after host death (Eyal et al. 1994).

Test B: One-time Exposure to Prey Contaminated 24-, 48-, 72-, or 96-h Previously. This test was conducted similarly to test A. However, eggplant leaves were infested with ≈400 *B. argentifolii* 1st instars, and were sprayed on only 1 occasion with a *B. bassiana* conidial suspension (2-fold concentrated high dosage). These leaves were incubated for 24 h at 25°C and 100% RH under a photophase of 16:8 (L:D) h, and thereafter humidity was reduced to 50–55%. *S. parcesetosum* larvae were placed individually in 80 vented plastic petri dishes (4 cm diameter). Then, 20 larvae were each exposed for 24 h to 1 leaf infested with prey sprayed with conidia either 24 (larvae were then 4 d old), 48 (5 d old), 72 (6 d old) or 96 (7 d old) h earlier. In the time intervals before and after exposure to contaminated prey on contaminated leaves, the coccinellid larvae were fed untreated prey ad libitum. Carrier controls (0.02% aqueous Silwet) were included with each fungal treatment. Dead insects were processed as previously described. The test was replicated 3 times. Standard leaves were included in the test (see test A). Data were recorded and analyzed as in test A.

Test C: Simulation of Environmental Degradation of B. bassiana *Conidia.* This test differed from test B by only 1 variable. To simulate the field degradation of fungal inoculum (e.g., by rainfall, solar radiation, desiccation), *B. bassiana* conidia were washed off the leaf surface and cuticle of *B. argentifolii* before presenting

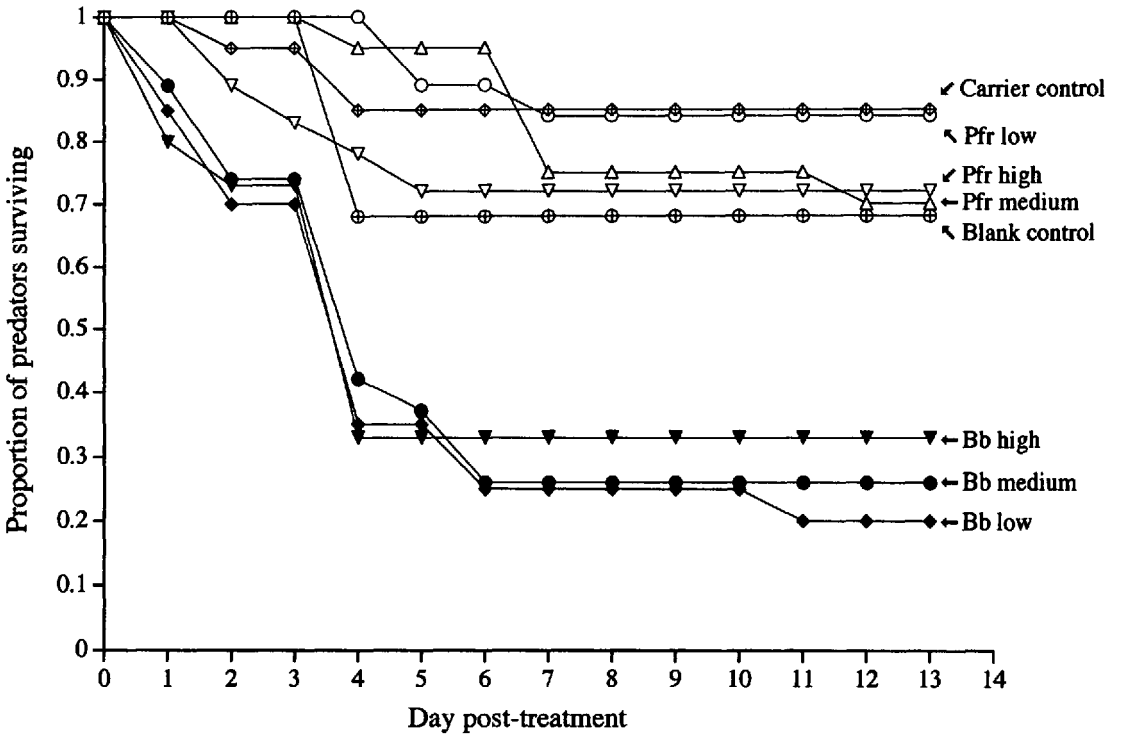

Fig. 1. Effect of *B. bassiana* (Bb) and *P. fumosoroseus* (Pfr) at 3 dosages (low, medium, and high) on survivorship of *S. parcesetosum* larvae. Larvae were 3 d old when sprayed with conidial suspensions.

the treated leaves or prey to *S. parcesetosum* larvae. Either 24-, 48-, 72-, or 96-h after treatment, leaves were introduced into beakers containing 0.13% Zephiran chloride in aqueous 0.04% Silwet, the beakers were agitated for 10 min on a reciprocal shaker (150 rpm), the leaves were rinsed twice in distilled water, and blot-dried with a paper towel. Although a few nymphs of *B. argentifolii* are lost during the process, conidia are effectively removed from the leaf surface and cuticle of *B. argentifolii*. Indeed, 16 standard leaves were used to determine the efficacy of the decontamination procedure. *B. bassiana*-treated leaves were washed as described and then firmly pressed, sprayed surface facing down, onto the surface of dodine-crystal violet agar. The leaves were removed and the agar plates incubated for 96 h at 25°C; by then, only 1 *B. bassiana* colony had developed on the agar in 2 of the 16 plates. Control (0.02% Silwet) leaves were treated as the *B. bassiana*-treated leaves. Dead insects were processed as previously described. Standard leaves used to determine levels of mycosis in *B. argentifolii* also were treated with *B. bassiana*, but not washed. The data were recorded and analyzed as in tests A and B.

Mortalities and mean time of mortality values in tests B and C were subjected to ANOVA. All statistical analyses were performed using the Systat package version 5.2 (Wilkinson et al. 1992). All tests were judged at $\alpha < 0.05$, and means were separated using Tukey highly significant difference (HSD) test.

Results

Effects of the Fungi Directly Sprayed on *S. parcesetosum*. The viability of the *B. bassiana* and *P. fumosoroseus* conidia used in the assays was 93.4 and 95.2%, respectively. After adjusting for viability, the mean ±SE high, medium, and low dosages of *B. bassiana* were 893 ± 56, 260 ± 19, and 30 ± 7 conidia per square millimeter, respectively; the adjusted dosages for *P. fumosoroseus* were 933 ± 62, 228 ± 24, and 20 ± 1 conidia per square millimeter, respectively.

The bioassays conducted with *S. parcesetosum* showed the differential susceptibility of the insect to the test fungi. Survivorship curves are presented in Fig. 1. Thirteen days after treatment, the proportions of larvae surviving in the blank and carrier controls were not significantly different ($\chi^2 = 1.51$, df = 1, $P = 0.219$, $n = 39$), indicating no effect due to the carrier. Survivorship on day 13 did not differ significantly among the 3 *P. fumosoroseus* treatments ($\chi^2 = 1.21$, df = 2, $P = 0.546$, $n = 57$) nor among the 3 *B. bassiana* treatments ($\chi^2 = 0.79$, df = 2, $P = 0.672$, $n = 54$), indicating no dosage effect for either of the fungi. The number of insects surviving at day 13 did not differ significantly among the 3 *P. fumosoroseus* treatments and the 2 controls ($\chi^2 = 2.72$, df = 4, $P = 0.604$, $n = 96$). Survivorship was significantly lower in the 3 *B. bassiana* cohorts than in any of the other cohorts ($\chi^2 = 38.30$, df = 7, $P < 0.001$, $n = 150$). Mycosis was 100% among the larvae that died in the *B. bassiana*

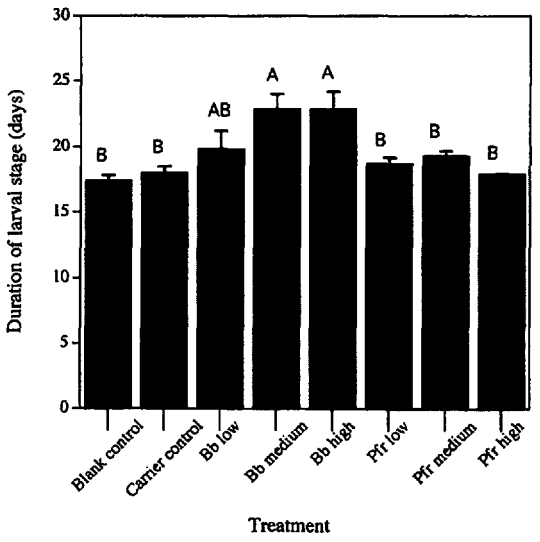

Fig. 2. Mean ± SE developmental time for *S. parcesetosum* larvae treated with 3 dosages (low, medium, and high) of *B. bassiana* (Bb) and *P. fumosoroseus* (Pfr) conidial suspensions. Larvae were 3 d old when treated with conidial suspensions. Letters above bars represent separation of the means (Tukey HSD test at $\alpha < 0.05$).

treatment. No mycosis was recorded in the *P. fumosoroseus* treatment nor in the blank and carrier controls. Control-corrected (Abbott 1925), pooled (e.g., over the 3 dosages) mortality was 66.4% in the *B. bassiana* cohorts and 2.2% in the *P. fumosoroseus* cohorts.

Treatment had a significant effect on the duration of the larval stage ($F = 8.03$; df = 7, 91; $P < 0.001$). However, only the 2 highest dosages of the *B. bassiana* treatment resulted in longer developmental times (Fig. 2).

The pupal stage lasted from 6.6 ± 0.2 d ($n = 25$; carrier control) to 8.0 ± 0.4 d ($n = 4$; *B. bassiana* medium dosage). No significant difference due to treatment was found in the duration of the pupal stage ($F = 1.98$; df = 7, 82; $P = 0.067$).

Mean adult body weight ranged from 0.97 ± 0.19 mg ($n = 3$; *B. bassiana* low dosage) to 1.54 ± 0.06 mg ($n = 10$; *P. fumosoroseus* medium dosage). Although they were lowest at the 3 dosages of *B. bassiana*, body weights did not differ significantly among the treatments ($F = 1.78$; df = 7, 71; $P = 0.100$).

Both time ($F = 562.56$; df = 1, 95; $P < 0.001$) and treatment ($F = 27.35$; df = 7, 95; $P < 0.001$) were found to be significant factors affecting predation rates (Fig. 3). Separation of the means (Tukey HSD tests on both daily and cumulative predation rates) indicated a significantly higher intake by *S. parcesetosum* in the 2 controls and the 3 *P. fumosoroseus* treatments than in the 3 *B. bassiana* treatments, but there were no significant differences between the 2 controls, or among the 3 *P. fumosoroseus* treatments, or among the 2 controls and the *P. fumosoroseus* treatments.

A general linear model (GLM) analysis with dead *S. parcesetosum* larvae removed from the analysis also indicated that both time ($F = 129.33$; df = 1, 95; $P < 0.001$) and treatment ($F = 30.68$; df = 7, 95; $P < 0.001$) were significant factors affecting food intake. Separation of the means (Tukey HSD tests on both daily and cumulative predation rates) indicated a significantly higher intake by *S. parcesetosum* in the controls and the 3 *P. fumosoroseus* treatments than in the 3 *B. bassiana* treatments. No significant difference among the 3 *B. bassiana* treatments was found. This indicated a possible fitness reduction in larvae surviving the *B. bassiana* treatments.

Suitability of Contaminated Prey. *Test A: Continuous Exposure.* Conidial counts, adjusted for 92.5% conidial viability, averaged 1,876 ± 85 conidia per square millimeter. Mycosis in *B. argentifolii* nymphs on standard leaves, and mortality, mycosis, and mean time of mortality in *S. parcesetosum* 10 d after treatment are given in Table 1.

Nine of the predators died from latent infection in the prepupal and pupal stages during the 10-d observation period. The 10-d Abbott-corrected mortality in *S. parcesetosum* immatures (larvae, prepupae, and pupae) in the *B. bassiana* treatment was significantly higher than in the control ($\chi^2 = 44.78$, df = 1, $P < 0.001$, $n = 104$). Only 9.2% ± 4.5 of the larvae (initial $n = 60$) became adults compared to 77.1% ± 5.8 of the larvae ($n = 60$) in the controls. The percentage of reduction in number of emerged adults in the *B. bassiana* treatment was 84.2%.

Test B: One-time Exposure. The conidial counts, adjusted for 94% conidial viability, averaged 1,672 ± 148 conidia per square millimiter. Mycosis in *B. argentifolii* nymphs on standard leaves, and mortality, mycosis and mean time of mortality in *S. parcesetosum* 10 d after treatment are listed in Table 2.

One predator in the 24-, 4 in the 48-, 3 in the 72-, and 1 in the 96-h series died from fungal infection in the pupal stage during the duration of the test. None died as pupa in the control. The 10-d Abbott corrected mortalities in *S. parcesetosum* immatures (larvae and pupae) in the 4 *B. bassiana* treatments were significantly different from control mortalities ($P < 0.001$ in the 4 χ^2 tests); there were no significant treatment (time series) differences ($F = 2.90$; df = 3, 8; $P = 0.102$). There were no significant treatment (time series) differences ($F = 0.37$; df = 3, 8; $P = 0.777$) among the 4 mean times of mortality. The reduction in number of emerged adults in the *B. bassiana* treatments was 88.9, 71.4, 75.0, and 55.5% in the 24-, 48-, 72-, and 96-h series, respectively. In all 4 series, a small proportion (<3%) of the treated insects died from latent mycosis or from unknown causes in the pupal or adult stage after the 10-d observation period.

Test C: Environmental Degradation. Conidial counts, adjusted for 94% conidial viability, averaged 1,613 ± 137 conidia per square millimeter. Mycosis in *B. argentifolii* nymphs on standard leaves, and mortality, mycosis and mean times of mortality in *S. parcesetosum* 10 d after treatment are given in Table 3.

Three predators in the 96-h series died from fungal

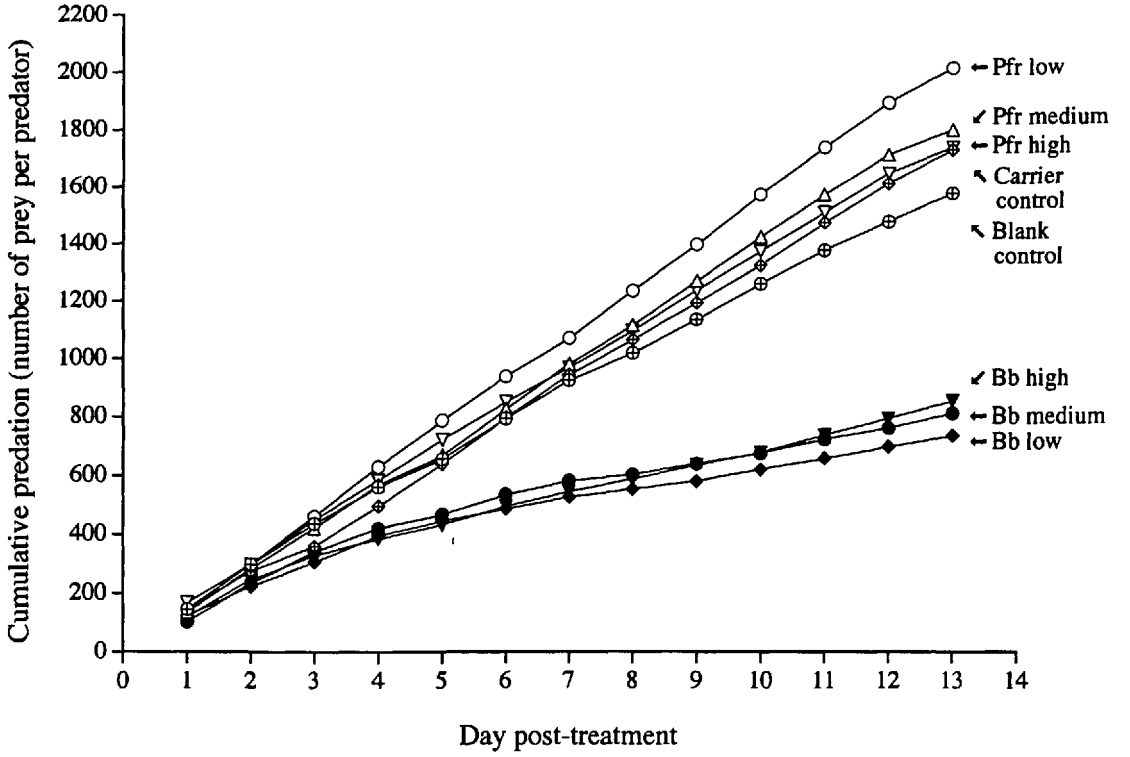

Fig. 3. Cumulative number of prey eaten per *S. parcesetosum* larva as affected by *B. bassiana* (Bb) and *P. fumosoroseus* (Pfr). Conidial suspensions were used at 3 dosages (low, medium, and high). Larvae were 3 d old when treated with conidial suspensions. Prey was not treated.

infection in the pupal stage during the duration of the test; none died in the other series nor in the control. The 10-d Abbott-corrected mortalities in *S. parcesetosum* immatures (larvae and pupae) in the 4 *B. bassiana* treatments (time series) were significantly higher than control mortalities ($P < 0.001$ in the 4 χ^2 tests); there were no significant treatment differences ($F = 3.13$; df = 3, 8; $P = 0.088$). The 4 mean times of mortality were not significantly different from each other ($F = 1.28$; df = 3, 8; $P = 0.345$). The reduction in number of emerged adults in the *B. bassiana* treatments was 33.3, 59.3, 74.1, and 77.8% in the 24-, 48-, 72-, and 96-h series, respectively. In all 4 series, a small proportion (<5%) of the treated insects died from latent mycosis or from unknown causes in the pupal or adult stages past the 10-d observation period. ANOVA on the mean times of mortality of tests A, B, and C combined revealed no statistical differences due to treatment ($F = 1.73$; df = 7, 27; $P = 0.136$).

Discussion

To evaluate 2 fungi used to control whiteflies, we looked at how a direct application of conidia to a predatory coccinellid affects survival, development time, and feeding rate. For 1 fungus, *B. bassiana*, we also looked at how exposure from feeding on contaminated prey affects the predator. In addition, we looked at the effect of killed *B. bassiana* on the predator.

Serangium parcesetosum larvae were highly susceptible to *B. bassiana* (80% uncorrected mortality at our lowest dosage of ≈30 conidia per square millimeter) but not to *P. fumosoroseus*. Goettel et al. (1990) listed coccinellid beetles of 16 genera that can be naturally infected by *B. bassiana*, but none by *P. fumosoroseus*. Magalhães et al. (1988) reported that *B. bassiana* caused mycosis in 60% of adult *Coleomegilla maculata lengi* Timberlake (Coleoptera: Coccinellidae) and in 35% of adult *Eriopis connexa* (Coleoptera: Coccinellidae) when conidia were applied directly to the insects. However, both coccinellid species were not infected following exposure to spore showers of the entomopathogenic fungus *Zoophthora* (= *Erynia*) *radicans* (Brefeld) Batko (Zygomycetes: Entomophthorales), although this fungus was highly pathogenic to one of its natural hosts, *Empoasca kraemeri* (Homoptera: Cicadellidae). James and Lighthart (1994)

Table 1. Suitability of *B. argentifolii* nymphs as prey for *Serangium parcesetosum* immatures under conditions of continuous exposure to prey and leaves contaminated with *B. bassiana*

B. argentifolii	*S. parcesetosum*		
% ± SE overt mycosis on standard leaves (*n* nymphs)	10-d Abbott percent ± SE mortality[a]	% overt mycosis	Mean time ± SE of mortality (days)
89.4 ± 4.2 (4,534)	85.9 ± 4.9	100	5.3 ± 0.3

[a] Control mortality was 13.3 ± 3.9%.

Table 2. Suitability of *B. argentifolii* nymphs as prey for *S. parcesetosum* immatures under conditions of exposure for 24 h to prey and leaves previously contaminated with *B. bassiana*

Time (h) after spray at which *Serangium* was exposed	*B. argentifolii*	*S. parcesetosum*		
	% ± SE overt mycosis on standard leaves (*n* nymphs)	10-d Abbott % ± SE mortality[a]	% ± SE overt mycosis	Mean time ± SE of mortality
24	79.1 ± 5.1 (2,429)	92.5 ± 6.7	76.3 ± 8.5	5.5 ± 0.4
48	73.5 ± 4.2 (1,938)	71.4 ± 10.7	63.3 ± 9.4	4.9 ± 0.8
72	89.3 ± 4.8 (3,788)	71.4 ± 2.2	56.7 ± 8.8	5.1 ± 0.5
96	68.4 ± 4.7 (2,732)	44.4 ± 5.8	30.0 ± 4.7	4.4 ± 0.6

[a] Overall control mortality was 8.3 ± 1.1%.

dipped 1st-instar *Hippodamia convergens* Guérin-Méneville (Coleoptera: Coccinellidae) for 10 s in 5 concentrations of 4 entomopathogenic fungi. Two *B. bassiana* strains caused from 75 to 95% mortality. *Metarhizium anisopliae* (Metschnikoff) Sorokin (Deuteromycotina: Hyphomycetes) caused up to 97% and *P. fumosoroseus* up to 56% mortality. *Nomuraea rileyi* (Farlow) Samson (Deuteromycotina: Hyphomycetes) did not kill the larvae. James and Lighthart (1994) concluded that *M. anisopliae, B. bassiana,* and *P. fumosoroseus* have the potential to infect *H. convergens* if used in crops where this predator occurs, but they also acknowledged that further research is needed to determine how direct effects observed in the laboratory play out in the field. Maddox et al. (1992) stated that ideally, introduced entomopathogenic organisms should not infect nontarget organisms; however, because most entomopathogens are not host specific, many unusual hosts may be infected in the laboratory. The different ecological host ranges of different entomopathogens (e.g., coevolution between hosts and pathogens) could partially explain the different susceptibilities found in our study and in previously reported studies. As an entomopathogen, *N. rileyi* is known only from lepidopteran hosts, particularly from Noctuidae, but *B. bassiana, P. fumosoroseus, M. anisopliae,* and *Z. radicans* are ubiquitous entomopathogens of worldwide distribution and they have been isolated from various arthropod (mainly insect) species. However, the host range of *Z. radicans* is restricted principally to aphids and dipterans, although it also is found in several hymenopterous wasps, lepidopteran larvae, and cicadas, *P. fumosoroseus* is known from a few species in the Diptera, Lepidoptera, Homoptera, and Coleoptera. *B. bassiana* and *M. anisopliae* have the widest host ranges (but *M. anisopliae* exhibits specificity within certain insect groups) among the entomopathogenic fungi and also occur in soil as common saprophytes; they are known from >700 host species in at least 9 insect orders.

Little information exists on the biology of *S. parcesetosum* (Legaspi et al. 1996) and on the sublethal or chronic effect of entomopathogenic fungi (when applied directly to the insects) on developmental time of insects. Developmental time of *S. parcesetosum* larvae feeding on eggs of citrus whitefly, *Dialeurodes citri* (Ashmead) (Homoptera: Aleyrodidae), lasts 20–21 d at 20–23°C (Timofeyeva and Nhuan 1978). Mean developmental time of larvae plus pupae is 19.0–32.4 d at 25°C in larvae feeding on eggs and nymphs of *Bemisia tabaci* Gennadius (Homoptera: Aleyrodidae) (Yigit 1992). Although larval developmental times at 25°C were longest in *S. parcesetosum* at the 2 highest dosages of *B. bassiana* in our study, they still were comparable to the times previously reported for uninoculated larvae at similar temperatures. Furthermore, there were no treatment effects on duration of the pupal stage and adult body weight. Thus, we conclude that neither *B. bassiana* strain GHA nor *P. fumosoroseus* strain 612 had sublethal effects on the developmental biology of *S. parcesetosum* surviving direct contamination by the fungi.

However, predation rate in our *B. bassiana* treatments was affected through mortality directly attributable to the treatment, through fitness reduction (e.g., reduced voracity) in infected, moribund larvae (Fargues et al. 1994), and possibly by sublethal dosages of the pathogen (Maddox 1992, Ranaivo et al. 1996). Timofeyeva and Nhuan (1978) reported that each larva of *S. parcesetosum* consumes 900–1,000 cit-

Table 3. Suitability of *B. argentifolii* nymphs as prey for *S. parcesetosum* immatures under conditions of exposure to leaves and prey contaminated with *B. bassiana* conidia after the conidia were killed

Time (h) after spray at which conidia were killed	*B. argentifolii*	*S. parcesetosum*		
	% ± SE overt mycosis on standard leaves (*n* nymphs)	10-d Abbott % ± SE mortality[a]	% ± SE overt mycosis	Mean time ± SE of mortality
24	71.2 ± 3.4 (1,824)	34.8 ± 3.9	16.7 ± 8.8	7.4 ± 0.6
48	60.5 ± 5.1 (2,127)	60.1 ± 5.6	10.0 ± 0	7.1 ± 0.7
72	86.1 ± 2.8 (1,411)	67.4 ± 3.9	6.7 ± 3.3	5.5 ± 0.5
96	68.4 ± 4.7 (1,602)	66.8 ± 4.8	0	5.3 ± 0.3

This experiment was designed to simulate environmental degradation of *B. bassiana* conidia.
[a] Overall control mortality was 8.1 ± 2.0%.

rus whitefly eggs (as many as 200 daily) during its 20–21 d development at 20–23°C. Rates of predation in the controls and the *P. fumosoroseus* treatments in our study are comparable to those reported by Timofeyeva and Nhuan (1978). However, the percentage of reduction in predation (e.g., beneficial capacity) in *B. bassiana*-treated larvae (3 dosages pooled) was 51.6% compared with predation by control larvae (blank and carrier controls pooled). No comparable data are available on such indirect effects of entomopathogenic fungi on predatory insects.

Mortality in *S. parcesetosum* larvae exposed to contaminated leaves and prey was high. Entomopathogenic fungi normally invade via the external cuticle and need not be ingested to initiate infection (Ferron 1978, Ferron et al. 1991, Tanada and Kaya 1993, Feng et al. 1994 and references therein). Despite this general mode of infection there is evidence that strains of *B. bassiana* (Gabriel 1959, Bao and Yendol 1971, Broome et al. 1976, Yanagita 1987, references in Feng et al. 1994) and possibly of other species of entomopathogenic fungi may infect their hosts, particularly insects with chewing mouthparts, via the alimentary tract. It is known that conidia of *B. bassiana* can germinate in the gut of certain insects regardless of the gut microflora (Allee et al. 1990).

Few studies have investigated the suitability of conidia of entomopathogenic fungi and of fungus-contaminated or fungus-infected prey for predatory arthropods. Where it has been studied, findings are inconsistent. For example, *Aschersonia*-infected and sporulating whiteflies can be an alternative (to pollen) food source for predators in greenhouse crops and citrus orchards, and thereby help with the establishment of these beneficial insects (see Lacey et al. 1996). Adult *Cryptolaemus montrouzieri* Mulsant (Coleoptera: Coccinellidae) and adult and larval *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae) were not affected after feeding on gelechiid larvae sprayed with the *B. bassiana*-based mycoinsecticide Boverin, but 50% of larvae of *C. montrouzieri* died (Kiselek 1975). Furthermore, conidia of *B. bassiana* had no toxicity following their ingestion (method of exposure not specified) by larvae or adults of the coccinellid *C. septempunctata* (Kiselek 1975). Blastospores of 1 *B. bassiana* strain mixed with wildflower pollen were toxic to larvae of *C. maculata lengi* (from 55.6 to 77.8% mortality depending on the dosage), but blastospores of another *B. bassiana* strain caused little mortality (Todorova et al. 1994). Pollen or larval *Leptinotarsa decemlineata* (Say) (Coleoptera: Chrysomelidae) treated with blastospores of one *B. bassiana* strain increased the developmental time of *C. maculata*, whereas *L. decemlineata* larvae treated with another *B. bassiana* strain reduced developmental time and pollen treated with the same strain had no effect (Todorova et al. 1997). In their review of the then existing literature Flexner et al. (1986) remarked that the only tests to show >10% fungus-induced mortality in parasitoids and predators were some of those in which natural enemies consumed the conidia.

In the test simulating environmental degradation of conidia, killed *B. bassiana* had a negative effect on *S. parcesetosum*, possibly due to toxin production. The dibenzoquinone pigment oosporein is produced by many isolates of *Beauveria* (Roberts 1981). It is a red antibiotic pigment that colors the insect cadaver red and curbs growth of bacteria upon the host death, thus allowing the fungal mycelium to compete with the intestinal bacterial flora (see Ferron [1981] and Eyal et al. [1994] for references). Oosporein is potently mycotoxic to mammals and avians, and exhibits inhibitory growth and phytotoxic effects on plants (Eyal et al. 1994). Its toxicity to insects is unknown (Roberts 1981), but it has been postulated (Eyal et al. 1994) that it plays a significant role in the infectivity process, especially at the stage where the fungus has already penetrated the host insect. Many other compounds toxic to insects are produced by isolates of *Beauveria* (Roberts 1981). Many entomopathogenic fungi overcome their hosts after only limited growth in the body cavity and poor sporulation on the host cadaver, so mycotoxins are presumed to cause host death. Although our data did not produce any information that would suggest a plausible mode of action of oosporein and possibly other mycotoxins on *S. parcesetosum*, we hypothesize that these toxic chemistries played a role in the death of the insects (leaves and whiteflies in test C were sporeless and very few cadavers of *S. parcesetosum* sporulated). Further studies are necessary with other isolates of *B. bassiana*, both toxin producing and toxin nonproducing, and with other species of entomopathogenic fungi.

Because of the variability among entomopathogens and their effects on their hosts, and because they occupy numerous and diverse ecological niches, laboratory experiments such as ours can provide information only on selected populations of nontarget insects submitted to maximum challenge tests (e.g., under conditions of temperature and humidity favoring the entomopathogens). Evaluation and prediction of effects on nontarget organisms from these laboratory studies may not be realistically applicable to agroecosystems. For example, Wilding (1981) stated that it is common that entomopathogens can infect hosts in the laboratory that are never found infected in the field, and Hajek et al. (1996) reported that data from laboratory bioassays are poor estimators for predicting nontarget impact. Fungi infect most genera of insects and mites but different pathogenic species or strains have different pathogenicities and virulences and can be quite specific and may only infect 1 type of host. Thus, more specific species or strains can be used to control pests without significantly affecting populations of beneficial predators and parasitoids (small numbers of these may, however, become infected). Theoretical information, and empirical studies on specific interactions, are needed (Maddox et al. 1992). Selecting the most appropriate insect pathogen for release with minimal hazards imposed on nontarget beneficial arthropods will require an indepth knowledge of how the pathogen will interact with other biological control agents.

There have been few attempts to elucidate the interactions among an insect, its entomopathogens, and its predators (Maddox et al. 1992). Steinberg and Prag (1994) report on the combined use of *A. aleyrodis* and *Delphastus pusillus* LeConte (Coleoptera: Coccinellidae) to control *B. tabaci* in greenhouse cucumber, *Cucumis sativus* L. No antagonistic interaction between the 2 agents was detected. *Aschersonia* species are specific pathogens of whiteflies and coccids and are unable to infect other insects (Lacey et al. 1996). Entomopathogenic fungi are important in the natural regulation of many insect pests. Several species are commercially produced in several countries and numerous other species have potential for development as microbial control agents and are in varying stages of development (Lacey and Goettel 1995). Their relative safety and selectivity (Goettel et al. 1990, Vinson 1990, Goettel and Johnson 1992, Lacey et al. 1996) should facilitate their integration into integrated pest management programs where their effects on other natural enemies will be minimal compared with most currently used chemical insecticides (Lacey and Goettel 1995).

At this time, we suggest that the integration of *P. fumosoroseus* strain 612 and of *S. parcesetosum* to manage *B. argentifolii* is feasible. The data are not as conclusive for *B. bassiana* strain GHA. A large proportion of *S. parcesetosum* died following the 4 different methods of exposure to *B. bassiana*. At the dosages used in tests A, B, and C, *S. parcesetosum* larvae began to die 3 d after initial exposure to conidia via leaf surfaces (the presumed predominant mode of exposure in the field [Malgalhães et al. 1988]) and via prey cuticle and presumably after ingestion of infected prey tissues or of mycotoxins. Continuous exposure (test A) and 1 time exposure (tests B and C) resulted in comparable mortalities. Mortality was not dependent on the age of the predator when exposed to *B. bassiana* nor on the age of the fungal infection in the prey (tests B and C). The mean time of mortality values in tests A, B, and C were comparable. The predatory species was highly susceptible to *B. bassiana* when conidia were applied directly to the insects or via feeding on contaminated surfaces or contaminated or infected prey, and thus the beneficial capacity of the predator was dramatically affected. Consequently, some adverse effect should be anticipated when *B. bassiana* strain GHA is applied on a crop supporting populations of the coccinellid. Further knowledge is needed to adjust timing of various releases of both biological control agents to obtain maximum additive effectiveness in the field with minimum impact of the fungus on the predator. A search for strains or pathotypes of the fungus with more narrow host ranges is also essential.

Acknowledgments

We thank R. Osterlind and R. Staten (APHIS Plant Protection and Quarantine, Phoenix, AZ) for providing the *S. parcesetosum* used in these experiments. Plants and whiteflies were kindly provided by A. Chavarria and L. Wendel (APHIS Mission Plant Protection Center, Mission, TX) and the conidia by Mycotech, Butte, MT. We are grateful to B. Legaspi (Texas A&M University, Weslaco, TX) for his advice on the statistical analyses, and to G. Elzen (USDA–ARS, Weslaco, TX), L. Lacey (USDA–ARS, Wapato, WA), B. Legaspi, and J. Vandenberg (USDA–ARS, Ithaca, NY) for helpful comments on the manuscript. S. Del Rio, P. Silva, and C. Veland (USDA–ARS, Weslaco, TX) provided technical assistance. This article is published with approval of the Director of the Texas Agricultural Experiment Station.

References Cited

Abbott, W. S. 1925. A method for computing the effectiveness of an insecticide. J. Econ. Entomol. 18: 265–267.

Allee, L. L., M. S. Goettel, A. Gol'berg, H. S. Whitney, and D. W. Roberts. 1990. Infection by *Beauveria bassiana* of *Leptinotarsa decemlineata* larvae as a consequence of fecal contamination of the integument following per os inoculation. Mycopathologia 111: 17–24.

Antadze, A. I., and T. V. Timofeyeva. 1975. An effective predator of citrus whitefly. Subtrop. Kult. 3: 80–81.

Bao, L., and W. G. Yendol. 1971. Infection of the eastern subterranean termite *Reticulitermes flavipes* (Kollar) with the fungus *Beauveria bassiana* (Balsamo) Vuill. Entomophaga 16: 343–352.

Brooks, W. A. 1993. Host-parasitoid-pathogen interactions, pp. 231–272. *In* N. E. Beckage, S. N. Thompson, and B. A. Federici [eds.], Parasites and pathogens of insects, vol. 2. Pathogens. Academic, San Diego.

Broome, J. R., P. P. Sikorowski, and B. R. Norment. 1976. A mechanism of pathogenicity of *Beauveria bassiana* on the larvae of the imported fire ant, *Solenopsis richteri*. J. Invertebr. Pathol. 28: 87–91.

Cock, M.J.W. [ed.] 1993. *Bemisia tabaci*—An update 1986–1992 on the cotton whitefly with an annotated bibliography. CAB International, International Institute of Biological Control, Ascot, Berks, UK.

Croft, B. A. 1990. Arthropod biological control agents and pesticides. Wiley, New York.

De Barro, P. J. 1995. *Bemisia tabaci* biotype B: a review of its biology, distribution and control. Division of Entomology Technical Paper 33. CSIRO, Canberra, Australia.

Eyal, J., A. Mabud, K. L. Fischbein, J. F. Walter, L. S. Osborne, and Z. Landa. 1994. Assessment of *Beauveria bassiana* Nov. EO-1 strain, which produces a red pigment for microbial control. Appl. Biochem. Biotechnol. 44: 65–80.

Fargues, J., J. C. Delmas, and R. A. LeBrun. 1994. Leaf consumption by larvae of the Colorado potato beetle (Coleoptera: Chrysomelidae) infected with the entomopathogen, *Beauveria bassiana*. J. Econ. Entomol. 87: 67–71.

Feng, M. G., T. J. Poprawski, and G. G. Khachatourians. 1994. Production, formulation and application of the entomopathogenic fungus *Beauveria bassiana* for insect control: current status. Biocontrol Sci. Technol. 4: 3–34.

Ferron, P. 1978. Biological control of insect pests by entomogenous fungi. Annu. Rev. Entomol. 23: 409–442.

1981. Pest control by the fungi *Beauveria* and *Metarhizium*, pp. 465–482. *In* H. D. Burges [ed.], Microbial control of pests and plant diseases 1970–1980. Academic, London.

Ferron, P., J. Fargues, and G. Riba. 1991. Fungi as microbial insecticides against pests, pp. 665–706. *In* D. K. Arora, L. Ajello, and K. G. Mukerji [eds.], Handbook of applied mycology, vol. 2. Humans, animals and insects. Marcel Dekker, New York.

Flexner, J. L., B. Lighthart, and B. A. Croft. 1986. The effects of microbial pesticides on non-target, beneficial arthropods. Agric. Ecosyst. Environ. 16: 203–254.

Fransen, J. J. 1990. Natural enemies of whiteflies: fungi, pp. 187–210. *In* D. Gerling [ed.], Whiteflies: their bionomics, pest status and management. Intercept, Andover, Hants.

Gabriel, B. P. 1959. Fungus infection of insects via the alimentary tract. J. Insect Pathol. 1: 319–330.

Gerling, D. 1990. Natural enemies of whiteflies: predators and parasitoids, pp. 147–185. *In* D. Gerling [ed.], Whiteflies: their bionomics, pest status and management. Intercept, Andover, Hants.

1996. Status of *Bemisia tabaci* in the Mediterranean countries: opportunities for biological control. Biol. Control 6: 11–22.

Goettel, M. S., and D. L. Johnson. 1992. Environmental impact and safety of fungal biocontrol agents, pp. 356–361. *In* C. J. Lomer and C. Prior [eds.], Biological control of locusts and grasshoppers. CAB, Wallingford, Oxon.

Goettel, M. S., and S. T. Jaronski. 1997. Safety and registration of microbial agents for control of grasshoppers and locusts. Mem. Entomol. Soc. Can. 171: 83–99.

Goettel, M. S., T. J. Poprawski, J. D. Vandenberg, Z. Li, and D. W. Roberts. 1990. Safety to nontarget invertebrates of fungal biocontrol agents, pp. 209–231. *In* M. Laird, L. A. Lacey, and E. W. Davidson [eds.], Safety of microbial insecticides. CRC, Boca Raton.

Hajek, A. E., L. Butler, S.R.A. Walsh, J. C. Silver, F. P. Hain, F. L. Hastings, T., M. Odell, and D. R. Smitley. 1996. Host range of the gypsy moth (Lepidoptera: Lymantriidae) pathogen *Entomophaga maimaiga* (Zygomycetes: Entomophthorales) in the field versus laboratory. Environ. Entomol. 25: 709–721.

Henneberry, T. J., N. C. Toscano, R. M. Faust, and J. R. Coppedge [eds.]. 1993. Sweetpotato Whitefly: 1993 Supplement to the Five-Year National Research and Action Plan—First Annual Review, Tempe, Arizona, 18–21 January 1993. U.S. Dep. Agric. Agric. Res. Serv. ARS-112.

Iperti, G. 1966. Protection of coccinellids against mycosis, pp. 189–190. *In* I. Hodek [ed.], Ecology of Aphidophaga. W. Junk, Dordrecht.

James, R. R., and B. Lighthart. 1992. Protocol for testing the effects of fungal pesticides on nontarget beetles using *Hippodamia convergens* (Coleoptera: Coccinellidae). National Technical Information Service Publ. PB92-217-488. Springfield, VA.

1994. Susceptibility of the convergent lady beetle (Coleoptera: Coccinellidae) to four entomogenous fungi. Environ. Entomol. 23: 190–192.

Kapadia, M. N., and S. N. Puri. 1989. Seasonal incidence of natural enemies of *Bemisia tabaci* (Gennadius). Indian J. Ecol. 16: 164–168.

Kiselek, E. V. 1975. The effect of biopreparations on insect enemies. Zashch. Rast. 12: 23.

Kuchanwar, D. B., M. G. Harda, M. N. Borle, and B. K. Sharnagat. 1982. *Catana parcesetosa,* a potential predator of the citrus black fly, *Aleurocanthus woglumi* Ashby. Punjabrao Krishi Vidyapeeth Res. J. 6: 74.

Lacey, L. A., and J. J. Fransen. 1994. Fungi as biological control agents of *Bemisia tabaci* s.l. Bemisia Newsl. 8: 34–35.

Lacey, L. A., and M. S. Goettel. 1995. Current developments in microbial control of insect pests and prospects for the early 21st century. Entomophaga 40: 3–27.

Lacey, L. A., A. A. Kirk, and R. D. Hennessey. 1993. Foreign exploration for natural enemies of *Bemisia tabaci* and implementation in integrated control programs in the United States. Proceedings ANPP Int. Conf. Pests of Agric. 1: 351–360.

Lacey, L. A., J. J. Fransen, and R. Carruthers. 1996. Global distribution of naturally occurring fungi of *Bemisia,* their biologies and use as biological control agents, pp. 401–433. *In* D. Gerling and R. T. Mayer [eds.], Bemisia 1995: Taxonomy, biology, damage and management. Intercept, Andover, Hants.

Legaspi, J. C., B. C. Legaspi, Jr., R. L. Meagher, Jr., and M. A. Ciomperlik. 1996. Evaluation of *Serangium parcesetosum* (Coleoptera: Coccinellidae) as a biological control agent of the silverleaf whitefly (Homoptera: Aleyrodidae). Environ. Entomol. 25: 1421–1427.

Maddox, J. V. 1992. The effect of regulations on the use of insect pathogens as biological control agents, pp. 73–81. *In* R. Charudattan and H. Browning [eds.], Regulations and guidelines: Critical issues in biological control. USDA–CSRS National Workshop, Vienna, VA.

Maddox, J. V., M. L. McManus, M. R. Jeffords, and R. E. Webb. 1992. Exotic insect pathogens as classical biological control agents with an emphasis on regulatory considerations, pp. 27–39. *In* W. C. Kauffman and J. E. Nechols [eds.], Selection criteria and ecological consequences of importing natural enemies. Thomas Say Publications, Entomology Proceedings, Entomological Society of America, Lanham, MD.

Magalhães, B. P., J. C. Lord, S. P. Wraight, R. A. Daoust, and D. W. Roberts. 1988. Pathogenicity of *Beauveria bassiana* and *Zoophthora radicans* to the coccinellid predators *Coleomegilla maculata* and *Eriopis connexa.* J. Invertebr. Pathol. 52: 471–473.

Mills, N. J. 1981. The mortality and fat content of *Adalia bipunctata* during hibernation. Entomol. Exp. Appl. 30: 265–268.

Nordlund, D. A., and J. C. Legaspi. 1996. Whitefly predators and their potential for use in biological control, pp. 499–513. *In* D. Gerling and R. T. Meyer [eds.], Bemisia 1995: taxonomy, biology, damage and management. Intercept, Andover, Hants.

Onillon, J. C. 1990. The use of natural enemies for the biological control of whiteflies, pp. 287–313. *In* D. Gerling [ed.], Whiteflies: their bionomics, pest status and management. Intercept, Andover, Hants.

Rainovo, F., M. Welling, G. Zimmermann, and H. Schmutterer. 1996. Fitness reduction of the African migratory locust, *Locusta migratoria,* after application of low concentrations of *Metarhizium flavoviride* blastospores and neem oil. IOBC WPRS Bull. 19(9): 236–239.

Roberts, D. W. 1981. Toxins of entomopathogenic fungi, pp. 441–464. *In* H. D. Burges [ed.], Microbial control of pests and plant diseases 1970–1980. Academic, London.

Steinberg, S., and H. Prag. 1994. Efficacy of the fungus *Aschersonia aleyrodis* and the coccinellid predator *Delphastus pusillus,* used to control *Bemisia tabaci* in greenhouse cucumber. Bemisia Newsletter (special issue) 8: 3.

Tanada, Y., and H. K. Kaya. 1993. Insect pathology. Academic, San Diego.

Tillemans, F., G. Iperti, and F. J. Coremans-Pelseneer. 1992. Sensibilité de certaines coccinelles aphidiphages à *Beauveria brongniartii* (Sacc.) Petch (Fungi Imperfecti). Meded. Fac. Landbouwwet. Rijksuniv. Gent 55 (2a): 369–372.

Timofeyeva, T. V., and H. D. Nhuan. 1978. Morphological and biological characteristics of the Indian coccinellid, *Serangium parcesetosum* (Sicard) (Coleop., Coccinellidae), a predator of the citrus whitefly in Adzharia. Entomol. Obozr. 57(2): 302–308.

Todorova, S. I., J.-C. Côté, P. Martel, and D. Coderre. 1994. Heterogeneity of two *Beauveria bassiana* strains revealed by biochemical tests, protein profiles and bio-assays on *Leptinotarsa decemlineata* (Col.: Chrysomelidae) and *Co-*

leomegilla maculata lengi (Col.: Coccinellidae) larvae. Entomophaga 39: 159–169.

Todorova, S. I., J.-C. Côté, and D. Coderre. 1998. Evaluation of the effects of two *Beauveria bassiana* (Balsamo) Vuillemin strains on the development of *Coleomegilla maculata lengi* Timberlake (Coleoptera: Coccinellidae). J. Appl. Entomol. (in press)

Vinson, S. B. 1990. Potential impact of microbial insecticides on beneficial arthropods in the terrestrial environment, pp. 43–64. *In* M. Laird, L. A. Lacey, and E. W. Davidson [eds.], Safety of microbial insecticides. CRC, Boca Raton.

Wilding, N. 1981. Pest control by Entomophthorales, pp. 539–554. *In* H. D. Burges [ed.], Microbial control of pests and plant diseases 1970–1980. Academic, London.

Wilkinson, L., M. Hill, and E. Vang. 1992. SYSTAT: statistics, version 5.2 ed. SYSTAT, Evanston, IL.

Yaganita, T. 1987. Studies on oral infection of the silkworm, *Bombyx mori*, with *Beauveria bassiana*. J. Seric. Sci. Jpn. 56: 279–284.

Yigit, A. 1992. Method for culturing *Serangium parcesetosum* Sicard (Coleoptera: Coccinellidae) on *Bemisia tabaci* Genn. (Homoptera: Aleyrodidae). J. Plant Dis. Prot. 99: 525–527.

Received for publication 3 June 1997; accepted 10 February, 1998.