

**INCREASED TOLERANCE OF FALL ARMYWORMS  
(LEPIDOPTERA: NOCTUIDAE) TO CRY1AC  $\delta$ -ENDOTOXIN  
WHEN FED TRANSGENIC *BACILLUS THURINGIENSIS* COTTON:  
IMPACT ON THE DEVELOPMENT OF SUBSEQUENT GENERATIONS**

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ABSTRACT

Increased tolerance to Cry1Ac protein was found in a population of fall armyworms, *Spodoptera frugiperda* (J. E. Smith), after selection for a single generation with transgenic *Bacillus thuringiensis* Berliner (Bt) cotton foliage. When fed Cry1Ac treated artificial diet, larvae whose parents had fed on transgenic Bt cotton leaves had significantly higher larval weights and a shorter time to pupation than those larvae whose parents had fed on conventional cotton leaves. In addition, there was no evidence to suggest any fitness or vigor differences existed from progeny of fall armyworms that fed previously on conventional or transgenic Bt cotton. Furthermore, tolerance of fall armyworms to Cry1Ac had a heritable component in the subsequent generation based on larval weights and time to pupation. These data show that using a common approach designed to control all intrinsically tolerant lepidopteran species of transgenic Bt cotton identically may not be desirable.

Key Words: *Spodoptera frugiperda*, plant-resistance

RESUMEN

Aumento en tolerancia a la proteína Cry1Ac fue encontrado en poblaciones del gusano de otoño *Spodoptera frugiperda* (J. E. Smith), después de escogimiento para una generación singular de follaje de algodón con *Bacillus thuringiensis* Berliner (Bt) transgénico. Al ser alimentadas una dieta artificial tratada con Cry1Ac, las larvas con padres que se alimentaron de hojas de algodón Bt transgénica mostraron pesos larvales significativamente mayores y menos tiempo a pupación que las larvas quienes padres se alimentaron con hojas de algodón convencionales. También, no hubo evidencia para sugerir que existen diferencias de salud o vigor en progenie de *S. frugiperda* que se alimentaron previamente con algodón convencional o Bt transgénico. Además, la tolerancia de *S. frugiperda* a Cry1Ac tuvo un componente heredable en la generación subsecuente basado en pesos larvales y tiempo a pupación. Estos datos demuestran que pudiera no ser deseable usar una practica común diseñada a controlar idénticamente a todas las especies de lepidóptera intrínsecamente tolerantes al algodón Bt transgénico.

The fall armyworm, *Spodoptera frugiperda* (J. E. Smith), is a destructive migratory pest of many crops in the Western Hemisphere (Sparks 1979; Young 1979). Historically, this pest has been a sporadic, but serious pest of conventional cotton in the southern United States (Bass 1978; Smith 1985). This pest has the potential to damage both conventional cotton bolls and transgenic cotton bolls that contain an insecticidal Cry1Ac  $\delta$ -endotoxin from the soil bacterium, *Bacillus thuringiensis* Berliner (Bt). The damage to bolls of transgenic Bt cotton caused by the fall armyworm can be more extensive than other lepidopterous pest of cotton including bollworms, *Helicoverpa zea* (Boddy), tobacco budworms, *Heliothis virescens* (F.), and beet armyworms, *Spodoptera exigua* (Hübner) (Bagwell 1994; Adamczyk et al. 1998a). Although application rates of foliar insecticides are often reduced for bollworms found on transgenic Bt cotton, possibly due to reduced fitness or vigor of individuals (Brickle et al. 1999), local outbreaks

of fall armyworms on transgenic Bt cotton often need full application rates of foliar insecticide treatments to keep these populations below economic injury levels (Hood 1997; Smith 1997).

Although certain lepidopterous pests of cotton are very susceptible to current transgenic Bt technology [e.g. tobacco budworms and pink bollworms, *Pectinophora gossypiella* (Saunders)], fall armyworms, bollworms, and soybean loopers, *Pseudoplusia includens* (Walker) are only sub-lethally effected by the Cry1Ac  $\delta$ -endotoxin (MacIntosh et al. 1990, Wilson et al. 1992, Halcomb et al. 1996, Adamczyk et al. 1998b, and Sumerford & Solomon 2000a). It seems that the Cry1Ac  $\delta$ -endotoxin found in current transgenic Bt cotton varieties does not provide sufficient mortality to fall armyworm larvae, but only slows larval development (Adamczyk et al. 1998b). Thus, application of foliar insecticides must be used to control this pest on current transgenic Bt cotton varieties (Smith 1997).

Few studies have examined the impact Cry1Ac  $\delta$ -endotoxin has on subsequent generations of Lepidoptera. Lambert et al. (1998) showed that the increased tolerance of bollworm larvae to transgenic Bt cotton can occur in subsequent generations, although complex interactions (i.e., genetic vs. environmental) were not sufficiently resolved. Furthermore, in a similar study using fall armyworms, the effect of selection for a single generation with transgenic Bt cotton foliage on survival and development of fall armyworms could not be fully characterized (Adamczyk et al. 1998b).

It seems that tolerance to Bt is heritable among certain species of Lepidoptera. Sumerford & Solomon (2000b) showed a genetic component for variation in larval development among *H. zea* feeding on Cry1Ac diet. The authors also found that selecting for more optimally growing larvae was correlated with improved survivorship when larvae were exposed to Cry1Ac. This study was conducted in two parts: 1) to determine if increased or decreased tolerance of Cry1Ac  $\delta$ -endotoxin was found in a population of fall armyworms after selection for a single generation with transgenic Bt cotton foliage, and 2) to determine if offspring of more tolerant individuals also exhibited greater tolerance of Cry1Ac during the subsequent generation.

## MATERIALS AND METHODS

### $P_1$ Generation

A fall armyworm colony (obtained from Dr. Frank Davis (retired), USDA, ARS, CHPRRU at Mississippi State University) was utilized in all tests. Females from this colony are annually outcrossed with wild, pheromone trapped males to maintain genetic heterogeneity and traits present in field individuals. Larval and adult rearing as well as egg harvesting were conducted as described in Adamczyk et al. (1998b).

Three colonies of fall armyworms were established from the original colony mentioned above. Larvae were reared until pupation on artificial diet, conventional cotton leaves, and transgenic Bt cotton leaves as described in Adamczyk et al. (1998b) and modified in Adamczyk et al. (2000). Individual pupae were separated based on larval host (colony designation: NBT, BT, and DIET; reared on conventional leaves, transgenic Bt leaves, and artificial diet, respectively) and equal numbers of pupae (100) were then placed in 3.79 liter cylindrical containers for moth emergence. Adult rearing and egg harvesting were conducted as described in Adamczyk et al. (1998b).

### $G_1$ Experiment

To examine the effects Cry1Ac  $\delta$ -endotoxin had on a subsequent generation of fall armyworms,  $G_1$  neonates from all colonies were placed on arti-

cial diet incorporated with a lyophilized powder of MVP II containing 19.7% Cry1Ac by weight (purified Cry1Ac; Monsanto Co., St., Louis, MO) using the method described in Sumerford & Solomon (1999). Thirty neonates were placed in 28.6 ml cups (1 per cup) containing approximately 5.0 ml of Cry1Ac diet and replicated twice. In addition, the same cohort of individuals from the same three colonies was reared on non-Cry1Ac diet as a control to determine if vigor differences existed among colonies. In a preliminary experiment, it was determined that a dose of 10.0  $\mu$ g/ml of Cry1Ac slowed larval development of fall armyworms very similar to transgenic Bt cotton. Therefore, this diagnostic concentration was used in all tests. Survival of larvae at 7 days after exposure (DAE), survival to pupae, larval weights at 7 DAE, and time to pupation were recorded. Survival analysis between colonies for each dose was conducted with G-tests using PROC FREQ (SAS Institute 1998). All mean weights and times were log transformed before analyzed using REML-ANOVA (PROC MIXED; Littell et al. 1996).

### $G_2$ Experiment

To determine if tolerance of fall armyworms to Cry1Ac had a heritable component, moths from the above colonies were pooled and the subsequent generation tested. Regardless of what the  $P_1$  larvae fed upon, equal numbers of  $G_1$  larvae from all three colonies that fed on non-Cry1Ac diet were allowed to pupate, pooled, and adults mated as described above. This  $G_2$  colony (REG) served as a control again to account for any fitness or vigor differences among colonies.

Pupae from larvae reared the previous generation on Cry1Ac diet were separated based on time to pupation of  $G_1$  individuals. Those that had pupated at 15 DAE were termed the FAST colony and those individuals that pupated at 19 and 20 DAE were termed the SLOW colony. These pupation times were selected to insure that similar numbers of pupae were available to develop adequate colonies. All  $G_2$  colonies (REG, SLOW, and FAST) were maintained as described above. Survival of larvae at 8 DAE, survival to pupae, larval weights at 8 DAE, and time to pupation were recorded. Survival analysis between colonies for each dose was conducted with G-tests using PROC FREQ (SAS Institute 1998). All means weights and times were log transformed before being analyzed using REML-ANOVA (PROC MIXED; Littell et al. 1996).

## RESULTS AND DISCUSSION

### $G_1$ Experiment

Based on very high (>85%) survival data, rearing fall armyworms on transgenic Bt cotton had

no effect on mortality in the subsequent generation. In addition, there were no significant differences ( $P > 0.05$ ) in larval survival at 7 DAE ( $0 \mu\text{g/ml}$ :  $\chi^2 = 2.21$ ,  $df = 2$ ,  $P = 0.33$ ;  $10 \mu\text{g/ml}$ :  $\chi^2 = 3.33$ ,  $df = 2$ ,  $P = 0.19$ ) and survival to pupae ( $0 \mu\text{g/ml}$ :  $\chi^2 = 1.46$ ,  $df = 2$ ,  $P = 0.48$ ;  $10 \mu\text{g/ml}$ :  $\chi^2 = 0.29$ ,  $df = 2$ ,  $P = 0.87$ ) among all three colonies.

Larvae that were reared on Cry1Ac diet weighed significantly less ( $P < 0.05$ ) and took significantly more time to pupate ( $P < 0.05$ ) than those larvae reared on non-Cry1Ac diet which is a reported sub-lethal effect observed for fall armyworms feeding on transgenic Bt cotton (Adamczyk et al. 1998b) (Figs. 1 and 2). Significant differences ( $P < 0.05$ ) among colonies (larval weights:  $F = 12.27$ ;  $df = 2$ ,  $345$ ;  $P < 0.001$ , time to pupation:  $F = 13.19$ ;  $df = 2$ ,  $319$ ;  $P < 0.001$ ) and diet (larval weights:  $F = 541.24$ ;  $df = 1$ ,  $345$ ;  $P < 0.001$ , time to pupation:  $F = 329.58$ ;  $df = 1$ ,  $319$ ;  $P < 0.001$ ) were observed as well as colony by diet interactions (larval weights:  $F = 5.76$ ;  $df = 2$ ,  $345$ ;  $P = 0.004$ , time to pupation:  $F = 17.47$ ;  $df = 2$ ,  $319$ ;  $P < 0.001$ ). In addition, based on larval weight and time to pupation for larvae feeding on non-Cry1Ac diet, again there was no evidence to suggest any fitness or vigor differences existed among the three colonies ( $P > 0.05$ ).

It appears that rearing fall armyworms on transgenic Bt cotton caused increased tolerance in the subsequent generation to Cry1Ac. When fed Cry1Ac diet, larvae whose parents had fed on transgenic Bt cotton leaves (BT) had significantly ( $P < 0.05$ ) higher larval weights at 7 DAE and a shorter time to pupation than those larvae whose parents had fed on conventional cotton leaves (NBT) (larval weights:  $t = 5.24$ ,  $df = 345$ ,  $P < 0.001$

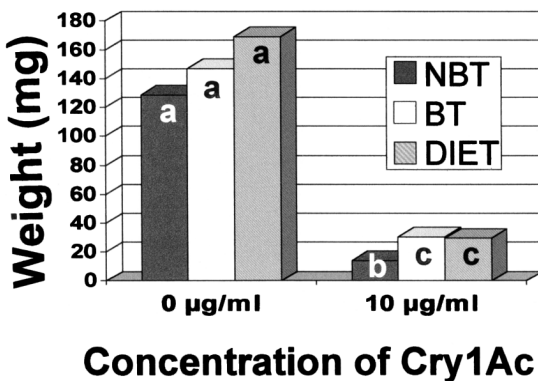


Fig. 1. Mean larval weights at 7 days after exposure (DAE) for fall armyworms fed non-Cry1Ac diet ( $0 \mu\text{g/ml}$ ) or Cry1Ac diet ( $10 \mu\text{g/ml}$ ). NBT, BT, and DIET colonies = previous generation reared on conventional cotton leaves, transgenic Bt cotton leaves, and non-Cry1Ac diet, respectively. Columns with the same letter are not significantly different ( $\alpha = 0.05$ ) from one another (REML-ANOVA; PROC MIXED; Littell et al. 1996).

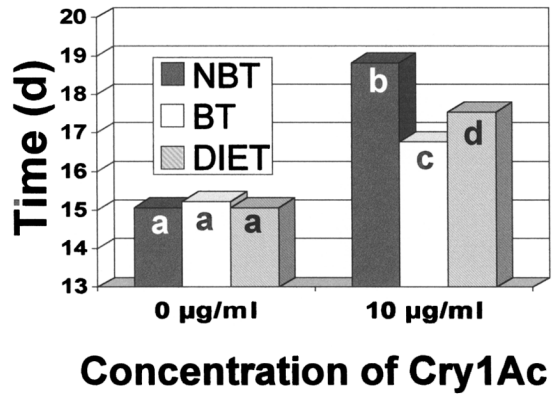


Fig. 2. Mean time to pupation for fall armyworms fed non-Cry1Ac diet ( $0 \mu\text{g/ml}$ ) or Cry1Ac diet ( $10 \mu\text{g/ml}$ ). NBT, BT, and DIET colonies = previous generation reared on conventional cotton leaves, transgenic Bt cotton leaves, and non-Cry1Ac diet, respectively. Columns with the same letter are not significantly different ( $\alpha = 0.05$ ) from one another (REML-ANOVA; PROC MIXED; Littell et al. 1996).

(LSMEANS); time to pupation:  $t = -7.59$ ,  $df = 319$ ,  $P < 0.001$  (LSMEANS)] (Figs. 1 and 2).

#### $G_2$ Experiment

As in the  $G_1$  experiment, there was no indication that  $G_2$  larvae were less fit or vigorous from feeding on Cry1Ac diet than from feeding on non-Cry1Ac diet. In fact, survival of pupae for the FAST colonies was significantly higher than the SLOW or REG colonies (Fig. 3).

Based on larval weights and time to pupation, increased tolerance to Cry1Ac was inherited among individuals in the subsequent generation (Figs. 4 and 5). Significant differences ( $P < 0.05$ ) among colonies (larval weights:  $F = 20.78$ ;  $df = 2$ ,  $348$ ;  $P < 0.001$ , time to pupation:  $F = 38.06$ ;  $df = 2$ ,  $309$ ;  $P < 0.001$ ) and diet (larval weights:  $F = 695.25$ ;  $df = 1$ ,  $348$ ;  $P < 0.001$ , time to pupation:  $F = 1360.96$ ;  $df = 1$ ,  $309$ ;  $P < 0.001$ ) were observed as well as colony by diet interactions (larval weights:  $F = 15.00$ ;  $df = 2$ ,  $348$ ;  $P < 0.001$ , time to pupation:  $F = 24.27$ ;  $df = 2$ ,  $309$ ;  $P < 0.001$ ). When fed Cry1Ac diet, larvae from the FAST colony had significantly ( $P < 0.05$ ) higher larval weights at 8 DAE and a shorter time to pupation than those larvae from the SLOW (larval weights:  $t = 8.06$ ,  $df = 348$ ,  $P < 0.001$  (LSMEANS); time to pupation:  $t = -9.66$ ,  $df = 309$ ,  $P < 0.001$  (LSMEANS)] or REG colonies (larval weights:  $t = -5.46$ ,  $df = 348$ ,  $P < 0.001$  (LSMEANS); time to pupation:  $t = 4.55$ ,  $df = 309$ ,  $P < 0.001$  (LSMEANS)]. Furthermore, based on time to pupation, a heritability estimate was calculated that further suggests tolerance to Cry1Ac was inherited in the subsequent generation ( $h^2_{\text{FAST}} = 0.49$ ). In addition, based on larval

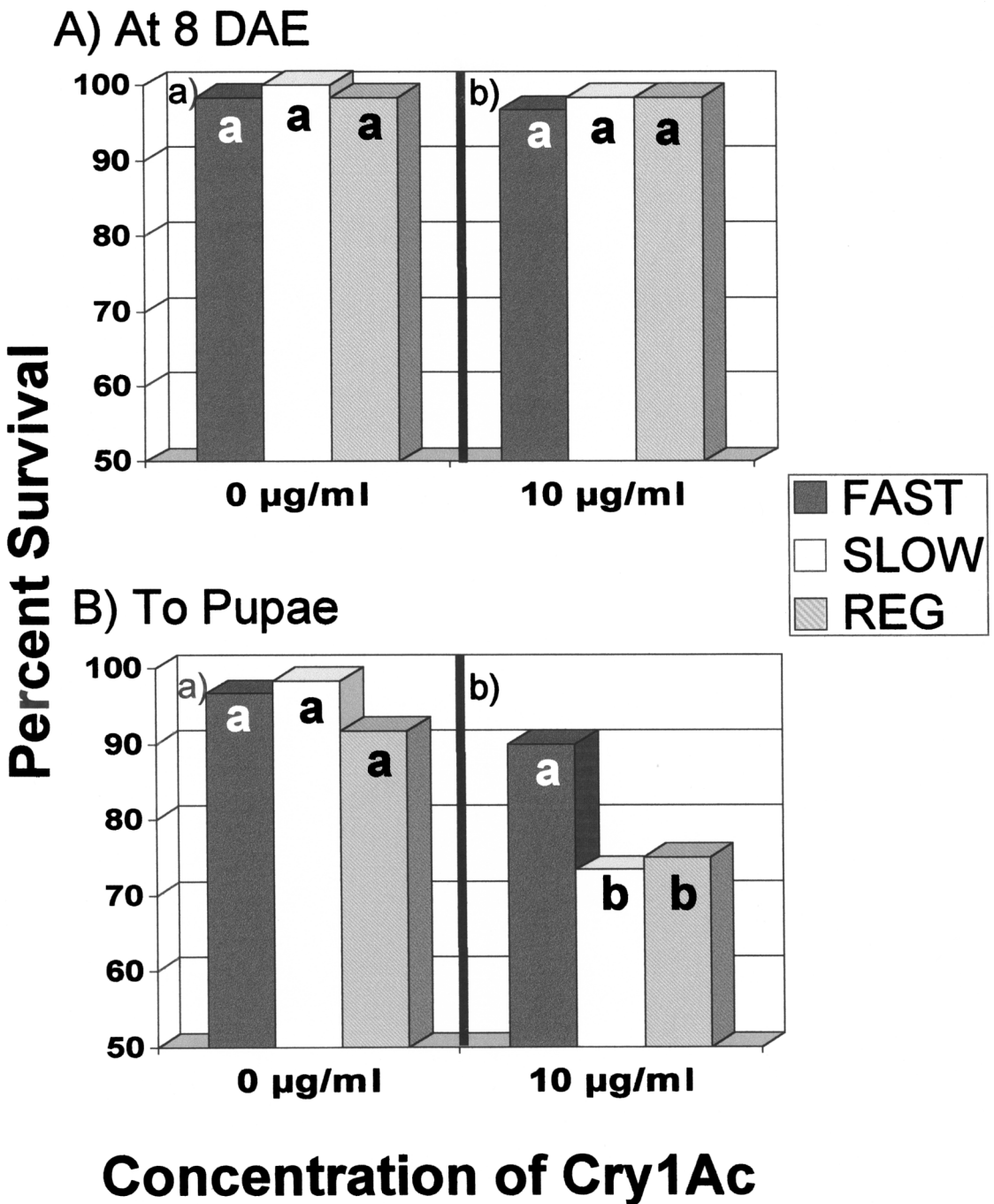


Fig. 3. (A) Mean larval survival of fall armyworms at 8 days after exposure (DAE) and (B) to pupae when fed: (a) non-Cry1Ac diet (0 µg/ml) or (b) Cry1Ac diet (10 µg/ml). FAST and SLOW colonies = previous generation pupated at 15-16 DAE and 19-20 DAE, respectively; REG colony = previous generation reared on non-Cry1Ac diet. Columns separated by dose with the same letter are not significantly different ( $\alpha = 0.05$ ) from one another (likelihood ratio chi-square analysis using PROC FREQ; SAS Institute 1998).

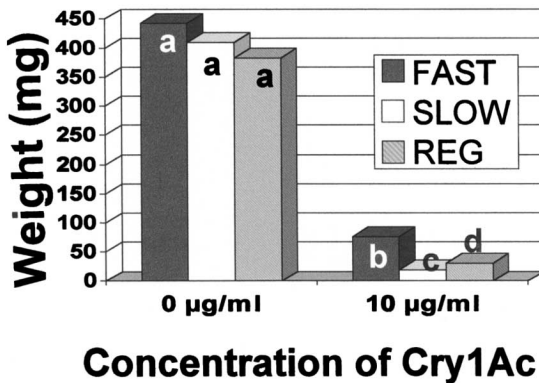


Fig. 4. Mean larval weights at 8 days after exposure (DAE) for fall armyworms fed non-Cry1Ac diet (0 µg/ml) or Cry1Ac diet (10 µg/ml). FAST and SLOW colonies = previous generation pupated at 15 DAE and 19-20 DAE, respectively; REG colony = previous generation reared on non-Cry1Ac diet. Columns with the same letter are not significantly different ( $\alpha = 0.05$ ) from one another (REML-ANOVA; PROC MIXED; Littell et al. 1996).

weight and time to pupation for larvae feeding on non-Cry1Ac diet, there was no evidence to suggest any fitness or vigor differences existed among the FAST and SLOW colonies ( $P > 0.05$ ), although the REG colony took significantly longer to pupate than either the FAST or SLOW colonies ( $P > 0.05$ ).

The assumption that sub-lethal effects from a single generation of exposure of fall armyworms to transgenic Bt cotton has a negative impact on fitness or vigor in the subsequent generation seems to be inaccurate. Although some studies

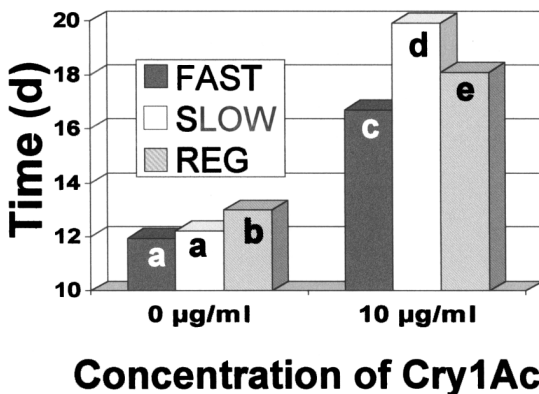


Fig. 5. Mean time to pupation for fall armyworms fed non-Cry1Ac diet (0 µg/ml) or Cry1Ac diet (10 µg/ml). FAST and SLOW colonies = previous generation pupated at 15 DAE and 19-20 DAE, respectively; REG colony = previous generation reared on non-Cry1Ac diet. Columns with the same letter are not significantly different ( $\alpha = 0.05$ ) from one another (REML-ANOVA; PROC MIXED; Littell et al. 1996).

have suggested that negative maternal effects can be transmitted by *H. zea* parents feeding on transgenic Bt cotton to their offspring (Lambert et al. 1998), no indications of this occurred with fall armyworms. Studies have further shown that reduced application rates of foliar insecticides can be used for *H. zea* on transgenic Bt cotton compared to conventional cotton, possible due to reduced vigor of larvae feeding on Cry1Ac (Brickle et al. 1999). Our data suggests that not all lepidopterous pests of cotton that are intrinsically tolerant to Cry1Ac may be controlled identically on transgenic Bt cotton. Because more than one generation of fall armyworms can attack transgenic Bt cotton in one season, future work will be needed to determine if larvae are more tolerant to Cry1Ac in later generations compared to previous generations in naturally occurring populations.

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## COMBINING EXCLUSION TECHNIQUES AND LARVAL DEATH-RATE ANALYSES TO EVALUATE MORTALITY FACTORS OF *SPODOPTERA EXIGUA* (LEPIDOPTERA: NOCTUIDAE) IN COTTON

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### ABSTRACT

By combining pesticide exclusion and cage exclusion techniques, the efficacy of natural enemies in reducing populations of *Spodoptera exigua* (Hübner), the beet armyworm, larvae was effectively demonstrated. Larval collections added information about parasitism and disease, and when combined with data from insecticide treatments, demonstrated that differences in *S. exigua* population densities usually were due to the action of predators. Death-rate analyses demonstrated that much mortality due to parasitism was contemporaneous with death from predation. When predator populations were not reduced by insecticides, most indispensable natural mortality was due to predation. When predators were eliminated, and *S. exigua* populations reached outbreak levels, most larvae died from disease in 1989 and from parasitism in 1990.

Key Words: Contemporaneous mortality, beet armyworm, predation, *Cotesia*

### RESUMEN

Al combinar técnicas de exclusión de pesticida y jaula, la eficacia de enemigos naturales para reducir poblaciones de *Spodoptera exigua* (Hübner) fueron demostradas efectivamente. Colectas de larvas añadieron información sobre parasitismo y enfermedad, y al ser combinadas con datos de tratamientos con insecticida, demostraron que diferencias en densidad de población de *S. exigua* usualmente fueron debidas a la acción de predadores. Analices de índices de muerte demostraron que gran parte de la mortalidad debido a parasitismo fue contemporánea con muerte por predación. Cuando poblaciones de predadores no fueron reducidas por insecticidas, la mortalidad natural más indispensable fue debida a predación. Cuando los predadores fueron eliminados, y poblaciones de *S. exigua* alcanzaron niveles epidémicos, la mayoría de las larvas murieron por enfermedad en 1989 y de parasitismo en 1990.

Evaluating the impact of natural enemies is a critical part of understanding pest population dynamics and of developing IPM systems (Luck et al. 1988; Sterling et al. 1989). One of two major approaches is usually taken to study biological control by indigenous agents in agricultural systems. In the first, exclusion of natural enemies with insecticides, cages, or other techniques is used to free pest populations from the action of the natural enemies. Each exclusion method has biases associated with it, but these biases may be at least partially overcome by combining methods (Luck et al. 1988). Densities of pest populations are then compared to densities of populations that are exposed to biological control agents (Luck et al. 1988). These studies have been used effectively to demonstrate that insecticide applications can disrupt natural enemy populations and lead to secondary pest outbreaks. However, insecticides also may affect insect populations by

direct stimulation of fecundity (hormoligosis) or by indirect stimulation of fecundity (trophobiosis) (Risch 1987; Kerns & Gaylor 1993b). Thus, without further evidence, mechanisms of outbreak induction may be unclear.

The second of the common approaches to studying biological control in agroecosystems uses one or more of several techniques (reviewed in Luck et al. 1988) to identify natural enemies that attack a pest. The importance of each natural enemy is then ranked by the proportion of a host population that is parasitized, diseased or preyed upon at various time intervals. Once mortality agents are identified, numerical and functional responses of agents, or groups of agents, to changes in pest densities may be determined. Effects of natural enemies identified by these techniques may then be incorporated into pest models. However, Van Driesche (1983) and Van Driesche et al. (1991) explained why this ap-

proach is inadequate for explaining host mortality over a generation. Also, evidence that a particular natural enemy or group of enemies can reduce, or even regulate, a pest population is not adequate evidence that the enemies do reduce the population. Different natural enemies may respond differently to temperature, prey availability or density (Sterling et al. 1989). Thus, any of several agents may be capable of reducing a pest population under specific conditions. Also, the action of one natural enemy may be masked by the actions of contemporaneous mortality factors. Contemporaneous mortality factors are two or more factors that attack a host more or less simultaneously, although death ultimately may be due to a single factor (Royama 1981). For example, an individual insect may be infected by a disease, which would ultimately kill it, but also be parasitized by a parasitoid.

The influences of contemporaneous mortality factors on insect population dynamics may be determined with life-tables (Royama 1981; Gould et al. 1990). Death-rate analysis may be used to estimate mortality rates due to contemporaneous mortality factors as one step in the construction of life tables (Bellows et al. 1992). Marginal attack rates are attack rates by individual mortality agents that would occur in the absence of other mortality agents acting on the same host (Royama 1981). Marginal attack rates, which may be calculated from observed death rates (Royama 1981; Gould et al. 1990), are particularly appropriate in death-rate analysis when the action of one or more mortality agents is difficult to detect (Bellows et al. 1992).

Many of these techniques and concepts, which are commonly used in population and community ecology, are relevant to IPM. However, for reasons that are primarily based on the historical separation between applied and basic research (Levins & Wilson 1980), these techniques have been used little by agricultural scientists.

Life tables alone can not be used to document the efficacy of natural enemies (Luck et al. 1988). However, an effective method of assessing the role of natural enemies in host population dynamics is to contrast life-tables for experimentally manipulated populations, in which one population lacks specific natural enemies and the other population is attacked by the enemies (Bellows et al. 1992).

*Spodoptera exigua* (Hübner), the beet armyworm, is an induced pest of many crops throughout the world. Historically, *S. exigua* outbreaks in cotton have been common in the western United States, but serious outbreaks also occur in the Southeast. Until recent years, these outbreaks in cotton have been difficult to control with insecticides and may result in complete destruction of some fields (Smith 1989). However, new insecticides such as spinosad (Tracer, Dow Agrosciences,

Indianapolis, IN) are relatively effective against *S. exigua* larvae (Halcomb et al. 1998), although somewhat expensive.

*S. exigua* is attacked by several predators and parasitoids (Eveleens et al. 1973; Pearson 1982; Oatman et al. 1983; Alvarado-Rodriguez 1987) and by protozoan, fungal and viral diseases (Smits 1987). Eveleens et al. (1973) and Hogg & Gutierrez (1980) concluded that in California cotton, *S. exigua* populations are normally held below economic injury levels primarily by predators feeding on eggs and small larvae. Parasitoids and disease apparently were less important as natural mortality agents of non-outbreak populations (Pearson 1982). A nuclear polyhedrosis virus (NPV) was the most important pathogen in *S. exigua* populations attacking tomatoes in Mexico (Alvarado-Rodriguez 1987) or cotton in California (Pearson 1982). In 1988, *S. exigua* outbreaks on cotton in Alabama were eventually controlled primarily by a naturally occurring NPV epizootic (Smith et al. 1989). In fields where the epizootic did not develop, rates of parasitism by a braconid, *Cotesia marginiventris* (Cresson), ranged from 23-43% (M. J. G., unpublished data). In Georgia and northern Florida, *C. marginiventris* parasitized 46% of the *S. exigua* larvae collected from cotton (Ruberson et al. 1993). More recently, Stewart et al. (1996) presented circumstantial evidence that *S. exigua* problem often coincide to areas where intensive insecticide applications are made for other pests, such as during intensive boll weevil eradication, *Anthonomus grandis grandis* Boheman, efforts.

Thus, the importance of biotic mortality factors on *S. exigua* outbreaks has been established. However, for reasons outlined in Van Driesche et al. (1991), none of these studies adequately determined effects of mortality factors on life stages over a *S. exigua* generation or determined effects of contemporaneous mortality factors on population dynamics. In this study, we combined insecticide and cage exclusion techniques with larval collections and death rate analyses to quantify sources of *S. exigua* mortality in cotton.

#### MATERIALS AND METHODS

In 1989 and 1990, 'DPL90' cotton was planted in  $\approx 0.3$  ha plots at the Wiregrass Substation of the Alabama Agricultural Experiment Station in Henry County, AL. In 1989, untreated plots were not possible because the area was within an active boll weevil eradication program. Our treatments were applications of malathion (Cythion 46.2% RTU, American Cyanamid Company, Princeton, NJ; applied at 1.4 kg [AI]/ha) or methyl parathion 6 EC (formerly marketed by Cheminova, Wayne, NJ; applied at 0.6 kg [AI]/ha), each applied to four main plots in a randomized complete block design. Both insecticides are effective against boll weevil



and most predators and parasitoids but not against *S. exigua*. Insecticides were applied with ground equipment every four to eight days.

Boll weevils were less numerous in 1990, and malathion applications were mandated by the eradication program only on 3 and 6 July. In 1990, main plots were four insecticide treatments: (1)  $\lambda$ -cyhalothrin (Karate 1E, Zeneca Ag Products, Wilmington, DE) applied alone at 0.028 kg (AI)/ha, (2) diflubenzuron (Dimilin 25 W [wetable], Uniroyal Chemical Company, Middlebury, CT) + a crop oil (Super Savol, Leffingwell, Brea, CA) (0.036 kg [AI] + 0.16 kg/ha), (3) a combination treatment of  $\lambda$ -cyhalothrin + diflubenzuron + crop oil (0.028 kg [AI] + 0.036 kg [AI] + 0.16 kg/ha) and (4) no insecticide. Four replicates of each insecticide treatment were used.

Diflubenzuron was applied to treatments 2 and 3 on 29 June, 20 and 27 July and 17 August.  $\lambda$ -cyhalothrin was applied to treatment 1 at 3- to 10-d intervals from 29 June to 24 August and to treatment 3 from 3 to 24 August.  $\lambda$ -cyhalothrin was tank mixed with the diflubenzuron on 29 June, 20 July and 17 August in treatment 3.  $\lambda$ -cyhalothrin is not effective against *S. exigua*, but it reduces populations of most predatory arthropods (Smith et al. 1993). Diflubenzuron may be effective against some *S. exigua* populations (Coudriet & Seay 1979; Ruberson et al. 1993; Smith et al. 1993) and is relatively non-damaging to populations of beneficial arthropods (Keever et al. 1977; Deakle & Bradley 1982).

Larval *S. exigua* and predator populations were estimated during both years with 1 to 3 drop-cloth samples, each sampling 1.8 row-m, per plot taken one or two times weekly. In 1989, weekly mean numbers of *S. exigua* were estimated. Larvae were not separated by size. In 1990, mean numbers were estimated for *S. exigua* in each of three size classes: small (first and second stadia), medium (third stadium) and large (fourth and fifth stadia). Effects of insecticide treatments on weekly mean numbers of predators and *S. exigua* larvae were compared by analysis of variance and, when significant main effects were found, means were separated with Fisher's LSD using the PROC GLM procedure (SAS Institute 1988) at  $\alpha = 0.05$ .

The combined effect of predators and parasitoids on *S. exigua* survival was determined using exclusion cages similar to those of Rice & Wilde (1988) and Kerns & Gaylor (1993a). Cages were 2 liter plastic bottles with two  $\approx 13 \times 9$  cm windows covered with cloth mesh. A Velcro closure was used on one window for access to the interior of the cage. In 1989, cages were of three mesh sizes: large (6.4 mm diam. opening), medium (1.5 mm diam. opening), and small (NoSeeum netting, Balson Hercules, Providence, RI). The small mesh excluded all predators and parasitoids. The medium mesh excluded large predators and parasitoids but allowed small predators and parasitoids

access to the *S. exigua* larvae. The large mesh allowed access by most invertebrate predators and parasitoids. Only small and medium mesh cages were used in 1990. Because methods of disease spread in *S. exigua* populations are unknown, proportions dying from disease were assumed to be equal in all cages and in the field.

Each week, one cage with each of the mesh sizes was placed on individual leaves on the periphery of separate plants in a subplot of each main plot. Subplots were 3-m sections of one row of cotton. Subplots were isolated from surrounding cotton by removing all plants from the ends of the subplots for a distance of  $\approx 2$  m on each end and all cotton from the 3-m sections of adjacent rows. An egg mass ( $\approx 25$  eggs) within one day of hatching or  $\approx 25$  newly eclosed larvae were placed into each cage. All eggs or larvae were from a laboratory colony, reared on a meridic diet, established in 1988 from *S. exigua* collected from cotton and periodically infused with wild males. Cages were moved to other leaves within the same subplot when the larvae had consumed most of the original leaf.

In 1990 *S. exigua* populations were established outside cages in separate subplots instead of in large mesh cages. A *S. exigua* egg mass from the laboratory colony or from a natural infestation in the same main plot was placed on the ventral side of a leaf to simulate a natural infestation. It was difficult to find all larvae on large plants in late-season, and large plants provided more food for developing larvae. Therefore, subplots were thinned to 10 contiguous plants in midseason. For late-season releases, subplots were thinned to 3 contiguous plants per subplot. During both years, caged and uncaged larvae were counted 3 to 5 times weekly until all had died or pupated.

Because stadia could not be accurately determined in the field, a developmental rate model (Ali & Gaylor 1992) was used to estimate *S. exigua* stadia on each sample date. Daily maximum and minimum temperatures were measured at a weather station that was within 1 km of the plots. Kerns & Gaylor (1993a) found no differences in temperatures inside or outside cages placed within the cotton canopy.

Larval mortality was estimated for each cohort and each mesh size. Totals, instead of means, were used because of initial differences in numbers of first instar larvae in individual cages. Initial population size for small larvae was arbitrarily set at 1,000. Mortality was estimated by multiplying the proportion of larvae surviving from one stadium to the next by 1,000. Because survivorship curves included only four data points, regression analyses were not performed. Instead, survivorship within cohorts for larvae in cages with different mesh sizes were compared with  $3 \times 4$  contingency tables using the PROC FREQ/CHISQ procedure (SAS Institute 1988) at

df = 6,  $\alpha = 0.05$ . When cage effects were found, differences in survivorship between cages were compared with  $2 \times 4$  contingency tables (df = 3,  $\alpha = 0.05$ ). Differences between cages in survivorship for each stadium (first and second stadia were combined) were compared with  $2 \times 2$  contingency tables (df = 1,  $\alpha = 0.05$ ).

To determine the incidence of disease and parasitism, larvae were collected from each main plot 2-4 times weekly, placed into individual plastic cups containing velvetbean caterpillar diet (Greene et al. 1976) chilled and returned to the laboratory. When available, larvae were collected from drop cloth samples and by visually searching plants in appropriate plots. However, each week 50 to 100 eggs or neonate larvae also were placed on cotton in a third subplot. Larvae were collected from these artificial infestations when natural infestations were low. When sufficient numbers were present, at least five larvae of each of the three size classes (small, medium and large) were collected from each plot on each collection date. Collected larvae were examined daily in the laboratory to determine their fate.

Immature parasitoids attacking *S. exigua* are difficult to identify. Thus, rates of parasitism were based on emergence of adult parasitoids. Pathogens were not isolated and cultured. Instead, estimates of disease incidence were based on visual symptoms. Larvae in which internal organs and integument remained intact, with minimal darkening of the integument after  $\approx 24$  h, were classed as dying of unknown causes. Cadavers classed as "fungal infected" collapsed and were covered with mycelia within a few hours of death. Larvae were judged to have died of "other diseases" if the internal organs liquefied or the integument became fragile within  $\approx 24$  h of the death of the larva. These are symptoms of acute infection by pathogens such as nuclear polythrosis virus (NPV) or by *Bacillus thuringiensis* Berliner (Bt).

The head capsule from the first molt after collection was measured with an ocular micrometer to determine the instar of each larva at the time of collection. Using the developmental rate model for *S. exigua* on cotton (Ali & Gaylor 1992), larval eclosion dates were estimated.

*S. exigua* generations occur at about one-month intervals (Trumble & Baker 1984). The first *S. exigua* infestations normally occur in southern Alabama cotton in July. Hence, the developmental rate model (Ali & Gaylor 1992) was used to place all larvae into July or August cohorts' equivalent to field generations of *S. exigua*. Contingency table analyses were used ( $\alpha = 0.05$ ) to compare effects of insecticide treatments, larval size classes, and cohorts within years on mortality caused by parasitism, disease or unknown causes. When contingency tables were significant, differences between causes were separated with  $2 \times 2$  contingency tables ( $\alpha = 0.05$ ).

Death-rate tables were developed for each cohort each year. Because of the broad-spectrum effects of both insecticide treatments used in 1989, death-rate tables were developed from both treatments combined. Data from untreated and diflubenzuron plots were combined in 1990 because diflubenzuron has little effect on predator populations (Keever et al. 1977; Deakle & Bradley 1982). Separate tables also were constructed for combination ( $\lambda$ -cyhalothrin + diflubenzuron) plots and for  $\lambda$ -cyhalothrin plots.

To construct death-rate tables, the initial population size ( $l_x$ ) for small larvae was set arbitrarily at 1,000 because separate populations were used to determine mortality rates. The numbers of larvae in each size class dying ( $d_x$ ) of parasitism, disease or "unknown" were estimated for each cohort by multiplying the proportion of field-collected larvae dying of each factor by  $l_x$  for the cohort and size class.

Marginal attack rates ( $m$ ) for disease, parasitism and unknown mortality factors were calculated by the formulae of Gould et al. (1990):

$$m_A = v_A / (1 - cm_B)$$

and

$$m_B = \{(c-1)v_A + cv_B + 1 - [(v_A - cv_A - cv_B - 1)^2 - 4cv_B]^{1/2} / 2c\}$$

where  $v_A$  = proportion of hosts dying of factor A;  $v_B$  = proportion of hosts dying of factor B;  $c$  = proportion of hosts dying of factor B, when A and B attack the same individual. In this case,  $c$  was assumed to be 0.5 because outcomes of competition between parasitoids and disease and "unknown" mortality factors are unknown. Marginal attack rates were calculated for each factor by letting  $v_A$  = the proportion of larvae dying of one factor and  $v_B$  = the proportion dying of all other factors except predation.

Estimating mortality due to predation is difficult because hosts that are preyed upon usually disappear from the system. In many life tables, mortality due to diseases and parasitoids is calculated, and predation is assumed to be the residual mortality that is unaccounted for by other factors (Bellows et al. 1992). This technique may underestimate the importance of predation because predation may be contemporaneous with other factors. Alternatively, predation effects may be overestimated if mortality rates due to abiotic, physiological, or unknown factors are high. Thus, an independent measure of deaths due to predation is needed.

In 1989, the boll weevil eradication program made an untreated control impossible. Thus, marginal attack rates for predators were estimated from the cage experiment using the formula from Royama (1981):

$$m_B = (m_{A+B} - m_A) / (1 - m_A)$$

where  $m_B$  = marginal attack rate of predators;  $m_A$  = marginal attack rate by parasitoids, estimated from collected larvae;  $m_{A+B}$  = total mortality due to A and B. However,  $m_B$  in this formula is not independent of  $m_A$ .

Because mortality was estimated from different host populations, marginal attack rates were used to estimate the proportion of larvae of each size class that should have died of each factor if all factors were acting on the same population ( $v$ ). This was done by solving the formula of Gould et al. (1990) for  $v_A$ . Thus,

$$v_A = m_A (1 - cm_B)$$

where  $c$  was assumed to be 0 when factor A was predation because larvae killed by predators would not produce parasitoids or disease symptoms. Values of  $l_{x+1}$  were obtained by subtracting total  $v_x$  from  $l_x$ .

In 1990, predation rates for each *S. exigua* cohort were estimated from differences in population densities in insecticide treatments. Based on larval collections, the most dense *S. exigua* population was assumed to have been reduced by the action of all agents except predators. Less dense populations were attacked by all natural enemies. Because success rates of most predators are unknown, we assumed that attacks by predators were always successful. Contemporaneous attacks that included predation were assumed to always be won by predators because there was no evidence that predators were less successful in attacking diseased or parasitized *S. exigua* than in attacking larvae that were not affected by other mortality factors. Thus, marginal attack rates for predation were equal to observed mortality ( $v$ ) due to predation. To estimate the marginal attack rate for predators, the area under the potential population density curve for a *S. exigua* population unaffected by natural enemies was first estimated by:

$$PP_{i,j} = PO_{i,j} / (1 - v_{tot})$$

where  $PP_{i,j}$  = area under the potential population density curve for the  $i^{\text{th}}$  size class in the  $j^{\text{th}}$  cohort,  $PO$  = the area under the curve for the most dense *S. exigua* population,  $v_{tot}$  = proportion of the population that was calculated to have died of all causes except predation. In all cases, except for large *S. exigua* in the second cohort, the most dense *S. exigua* populations were in the  $\lambda$ -cyhalothrin treatment. In the second cohort, more large larvae were in the  $\lambda$ -cyhalothrin + diflubenzuron treatment than in the other treatments. The marginal attack rate for predation ( $m_{pred}$ ) was estimated by:

$$m_{pred} = (PO_{i,j} - PL_{i,j}) / PP_{i,j}$$

where  $PL$  = the area under the curve for the least dense *S. exigua* population. Competition among

mortality factors in a single population includes predation. Therefore, a modified proportion ( $v'$ ) of larvae of each size class that would have died of each factor if all factors were acting on a single population was calculated by incorporating  $m_{pred}$  into the formula of Gould et al. (1990) for  $m_A$  and solving for  $v'$ . Thus,

$$v'_A = (1 - m_{pred}) (m_A (1 - (0.5 (m_B + m_C))))$$

The total of all  $v'$  for a *S. exigua* size class in a cohort was subtracted from  $l_x$  for the class to get  $l_x$  for the subsequent size class.

Indispensable mortality (IM) is mortality that would not be replaced in the host population by the subsequent action of other mortality factors if the factor under consideration were removed (Bellows et al. 1992). Within each size class and cohort for each year, IM was estimated for each mortality factor by subtracting the number of larvae surviving for the cohort from the number surviving when the effect of the factor was removed.

## RESULTS

### Insecticide Exclusion

In 1989 there were few differences in predator or *S. exigua* population densities due to insecticide treatments (Fig. 1). The first "wild" *S. exigua* egg mass was found on 17 July 1989. Larval *S. exigua* were first counted in beat sheet samples on 9 August, but populations remained sparse until 30 August, when  $3.2 \pm 0.95$  and  $5.5 \pm 0.23$  larvae per 0.9 row-m were found in malathion and methyl parathion-treated plots, respectively. Weekly mean *S. exigua* population densities were not significantly different between treatments ( $F = 0.74, 1.83, 2.57, 0.47$ ;  $df = 1,36, 1,26, 1,3, 1,3$ ; for 9, 16, and 30 August and 6 September, respectively;  $P > 0.05$ ).

Total predator densities declined after 5 July 1989 in plots treated with either insecticide. Predator populations then increased in mid- and late-August in both treatments, but densities were higher in the malathion-treated plots than in the methyl parathion-treated plots on 23 ( $F = 3.72, df = 1,34, P < 0.05$ ) and 30 August ( $F = 1.94, df = 1,35, P < 0.05$ ).

The imported fire ant, *Solenopsis invicta* Buren, was especially abundant in early season, comprising  $\approx 90\%$  of the predators in malathion plots on 5 and 12 July and  $\approx 80\%$  in the methyl parathion plots on 5 July. When total predator populations were least dense (2 August), ants were only  $\approx 20\%$  of the predator populations in both treatments. When the total predator population peaked again (23 August in the malathion plots) fire ants were  $\approx 70\%$  of the population.

Medium sized *S. exigua* were found in the first 1990 samples, during the week of 4 July (Fig. 2). These larvae were from the first of three *S. exigua*

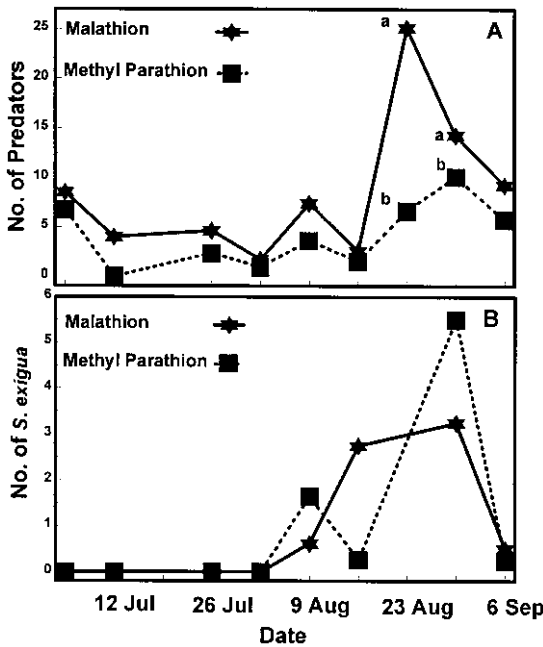


Fig. 1. Mean number of *S. exigua* larvae and predators per 1.8 row-m during 1989. Means on the same date not accompanied by the same letter are significantly different ( $P < 0.05$ ) according to Fisher's LSD (SAS Institute 1988).

generations in 1990. Populations of small larvae of the second generation peaked in mid-July and small larvae of the third generation peaked in mid-August (Fig. 2). Thus, the assignment of larvae to July and August cohorts represented the occurrence of second and third generations in 1990.

$\lambda$ -cyhalothrin treatments affected *S. exigua* and predator population densities in 1990 (Fig. 2). Mean numbers of small *S. exigua* in the July cohort were not different among insecticide treatments ( $F = 2.46, 0.56, 1.81$ ;  $df = 3, 108, 3, 89, 3, 89$ ; for 11, 18, and 25 July, respectively;  $P > 0.05$ ). Although more medium larvae were in the  $\lambda$ -cyhalothrin treatment than in the other plots on 18 July and 8 August ( $F = 4.02, 2.8$ , respectively;  $df = 3, 89$ ;  $P < 0.05$ ), populations in all treatments were small. Beginning on 18 July, total predator populations were lower in the  $\lambda$ -cyhalothrin and combination plots than in untreated and diflubenzuron plots ( $F = 7.60, df = 3, 89, P < 0.05$ ). On 25 July, fewer predators were in plots that had been treated regularly with  $\lambda$ -cyhalothrin (treatment 1) than in any other treatments ( $F = 13.05, df = 3, 89, P < 0.05$ ). Predator population densities in combination plots were intermediate between those in  $\lambda$ -cyhalothrin-treated plots and those in plots that were not treated with  $\lambda$ -cyhalothrin. When  $\lambda$ -cyhalothrin was applied to combination plots at frequent intervals beginning on 3 August,

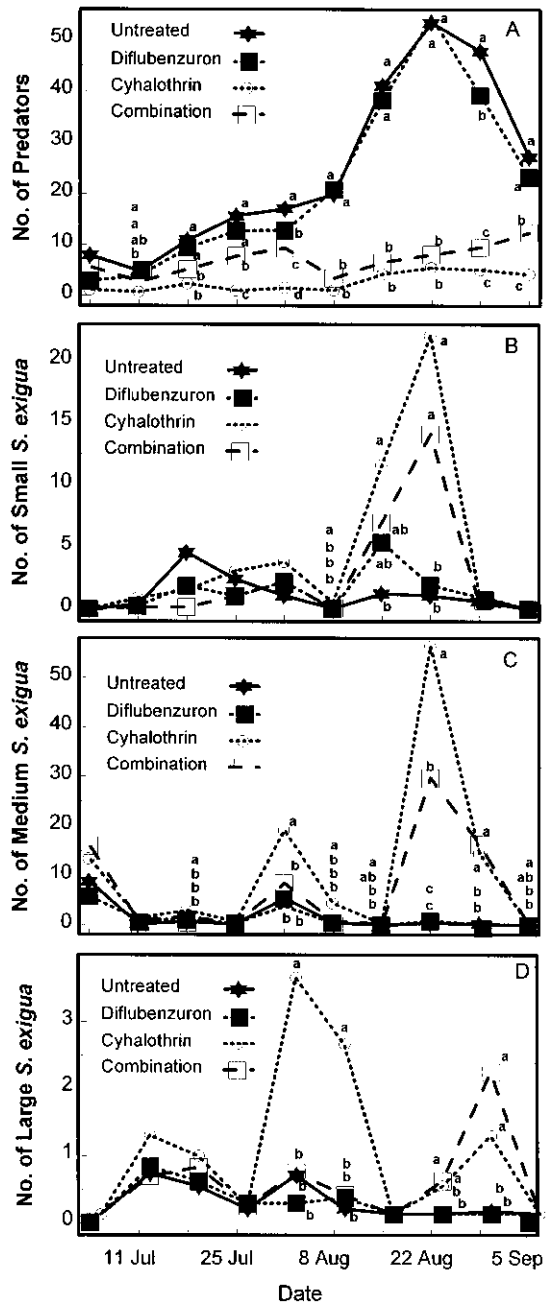


Fig. 2. Mean number of (A) small, (B) medium, and (C) large *S. exigua* larvae and (D) predators per 1.8 row-m during 1990. Means on the same date not accompanied by the same letter are significantly different ( $P < 0.05$ ) according to Fisher's LSD (SAS Institute 1988).

predator populations were quickly reduced. From 8 August until the end of August, predator populations were not different in the  $\lambda$ -cyhalothrin and combination plots, and they were lower than in plots that were not treated with  $\lambda$ -cyhalothrin

( $F = 27.75, 19.15, 67.64$  and  $34.58$  for 8, 15, 22 and 29 August, respectively;  $df = 3, 89$ ;  $P < 0.05$ ). When *S. exigua* populations peaked (22, 22 and 29 August for small, medium and large larvae, respectively), population densities for each size class were lower in plots with dense predator populations (not treated with  $\lambda$ -cyhalothrin) than in  $\lambda$ -cyhalothrin treated plots ( $F = 6.23, 14.51,$  and  $34.58$  for small, medium and large, respectively;  $df = 3, 89$ ;  $P > 0.05$ ). *S. exigua* population densities were not different between control and in diflubenzuron plots. Thus, in plots with dense predator populations, diflubenzuron did not further reduce *S. exigua* populations. Densities of medium *S. exigua* in combination plots were intermediate between those in  $\lambda$ -cyhalothrin-treated and untreated plots on 22 August. Thus, diflubenzuron reduced these population densities when predator populations were lower.

Ants made up 48-93% of the predator populations during the first week of sampling (4 July 1990). Subsequently, ants were virtually eliminated in plots treated with  $\lambda$ -cyhalothrin at 3- to 10-d intervals. Big-eyed bugs, *Geocoris* spp., were the most abundant predators in these plots. Both ant and big-eyed bug populations increased rapidly in August in all plots, but big-eyed bugs were the most abundant predators by 15 August.  $\lambda$ -cyhalothrin did not reduce big-eyed bug population densities below those in untreated plots, but ants remained less abundant in the  $\lambda$ -cyhalothrin-treated plots ( $F = 8.15, 14.30, 12.78$  and  $7.72$  for 8, 15, 22 and 29 August, respectively;  $df = 3, 89$ ;  $P < 0.05$ ).

Results of the insecticide exclusion experiment provided anecdotal evidence that, at least in 1990, predators controlled *S. exigua* populations. *S. exigua* populations increased dramatically when predator populations were eliminated by frequent applications of  $\lambda$ -cyhalothrin. However, effects of contemporaneous mortality factors (disease and parasitism) were not assessable by this experiment.

#### Cage Exclusion

Results of the cage exclusion experiment were similar to the results of the insecticide exclusion experiment. In both 1989 cohorts, more *S. exigua* survived to the fifth stadium in the total exclusion cages than in the cages that allowed access by predators and parasitoids ( $\chi^2 = 19.642$  and  $58.293$  for July and August, respectively;  $df = 6$ ;  $P < 0.05$ ) (Fig. 3). However, in July, there were no differences in survival for individual stadia. In August, survival was greater in total exclusion cages only during the third stadium, indicating that most of the differences in survival were due to mortality to medium larvae. There were no differences in survival in no exclusion and partial exclusion cages. In the July cohort, survival to the fifth stadium was 38.6 and 13.2% in total and no exclusion

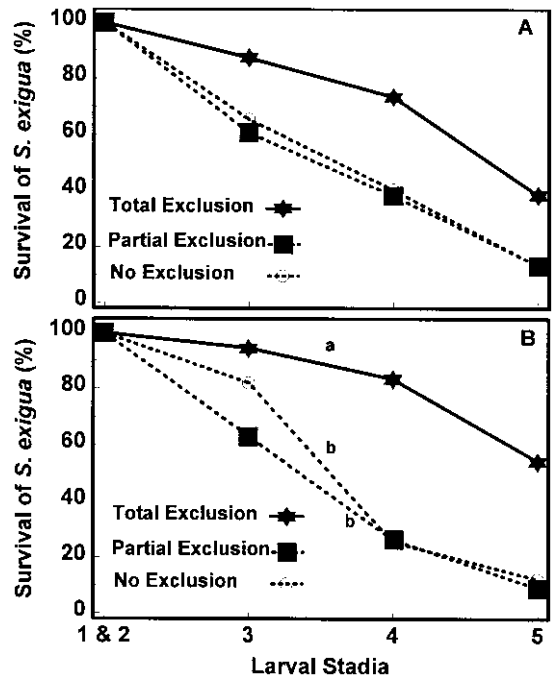


Fig. 3. *S. exigua* larval survival in 1989 inside total, partial and no exclusion cages. (A) July cohort. (B) August cohort. Survival within stadia not accompanied by the same letter are significantly different ( $P < 0.05$ ) according to  $2 \times 2 \chi^2$  tests (SAS Institute 1988).

cages, respectively, and survival was 54.2 and 12.2% in total and partial exclusion cages in the August cohort. Thus, larval survival differed most in the total exclusion cages versus the other cages when predator populations were most dense.

In 1990, in plots with relatively few predators, *S. exigua* survival inside total exclusion cages (Fig. 4) was not different from survival outside cages ( $\chi^2 = 3.313, 6.019$  and  $0.801$  for July cohorts treated and not treated with  $\lambda$ -cyhalothrin and for the treated August cohort, respectively;  $df = 3$ ;  $P > 0.05$ ). In July, survival outside cages in plots not treated regularly with  $\lambda$ -cyhalothrin (Fig. 4A) was intermediate between that inside partial and total exclusion cages. In  $\lambda$ -cyhalothrin-treated plots (i.e., few natural enemies) in July (Fig. 4B) and August (Fig. 4D), there were no differences in *S. exigua* survival inside total exclusion cages versus outside cages, but survival was lower in partial exclusion cages.

More larvae survived in total exclusion cages than outside cages in plots with dense predator populations (Fig. 4C). Thus, exclusion of predators by cages increased *S. exigua* survival when predator populations were dense, but not when predator populations were sparse. Data from the combination plots were included with data from untreated plots in July but were combined with

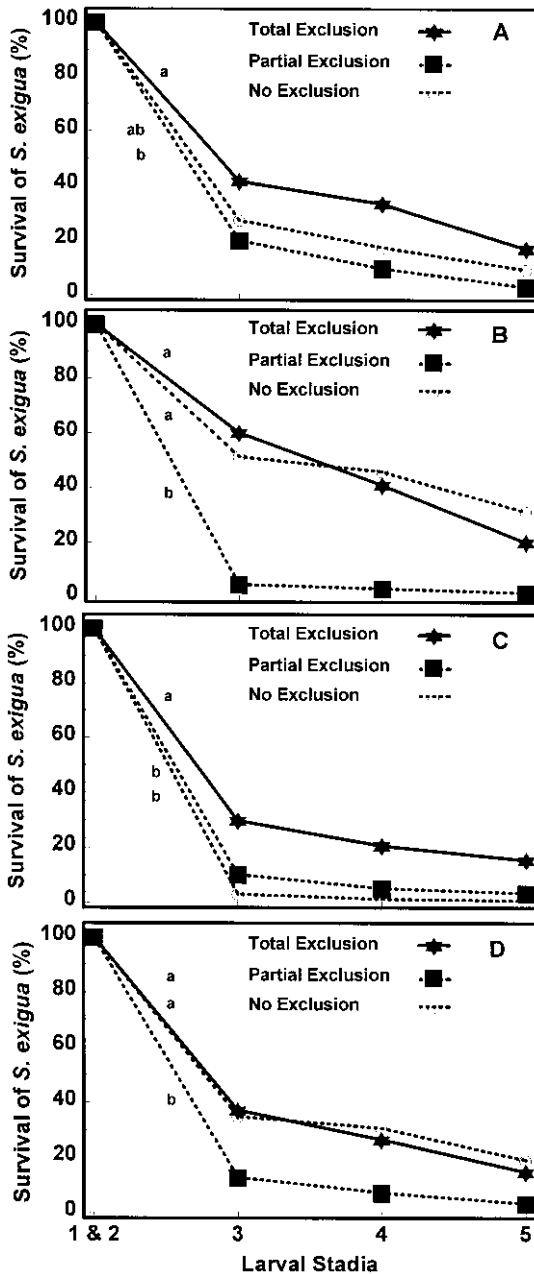


Fig. 4. *S. exigua* larval survival in 1990 outside exclusion cages and inside total and partial exclusion cages. (A) July cohort in cotton not treated with  $\lambda$ -cyhalothrin at 3- to 10-d intervals. (B) July cohort in cotton treated with  $\lambda$ -cyhalothrin. (C) August cohort in cotton not treated with  $\lambda$ -cyhalothrin. (D) August cohort in cotton treated with  $\lambda$ -cyhalothrin. Survival within stadia not accompanied by the same letter are significantly different ( $P < 0.05$ ) according to  $2 \times 2 \chi^2$  tests (SAS Institute 1988).

data from cyhalothrin-treated plots in August because  $\lambda$ -cyhalothrin dramatically reduced natural

enemy populations in August but not in July. In all cohorts in 1990, most *S. exigua* mortality occurred while larvae were small.

Insecticide and cage exclusion methods provided supporting evidence that *S. exigua* populations were controlled by natural enemies acting primarily on small and medium sized larvae. The  $\lambda$ -cyhalothrin-induced outbreak apparently was due to destruction of natural enemy populations. However, neither exclusion method provided information about contemporaneous mortality or the individual impact of different mortality agents attacking incipient versus outbreak populations.

Larval Collection

Prior to 4 August 1989, few *S. exigua* could be collected to determine parasitism or disease incidence. Because survival of laboratory-reared and released larvae was low, only 161 larvae were collected from the July 1989 cohort; 1273 larvae were collected from the August cohort (Table 1).

There were no differences in sources of mortality for different sizes of *S. exigua* larvae in July 1989 ( $\chi^2 = 2.418$ ,  $df = 2$ ,  $P > 0.05$ ). The most common mortality factor affecting larvae in this cohort was "unknown" (16.2%), that may have been partially attributable to handling and to the collected larvae originating from a laboratory colony. The first parasitoids emerged from larvae collected as 3rd instars on 20 July 1989, but little parasitism and no disease were found in this cohort.

Disease was a more important mortality factor in August. The first larva to die from disease was collected during the week of 9 August 1989. It was infected with a fungal pathogen. Rates of disease infection increased as larvae matured. In this cohort, more medium than small larvae ( $\chi^2 = 27.395$ ,  $df = 1$ ,  $P < 0.05$ ) and more large than medium larvae ( $\chi^2 = 14.622$ ,  $df = 1$ ,  $P < 0.05$ ) were diseased. Most diseases were fungal diseases; 3.4, 18.9, and 35.0% of small, medium, and large larvae were infected by fungi, respectively. However, within a single week the infection rate was even greater. On 6 September, 46 and 88% of the medium and large larvae, respectively, produced mycelia. Other diseases killed only 2.1% of the larvae collected from the August cohort.

In August 1989, most parasites emerged from medium larvae (Table 1). Effects on parasitism rates of the area-wide insecticide applications applied in 1989 for the boll weevil are unknown.

In 1990, 860 and 414 larvae were collected for the July and August cohorts, respectively. In both cohorts, insecticide treatments had no statistically significant effect on the incidence of disease ( $\chi^2 = 5.67$  and  $\chi^2 = 4.09$  for July and August, respectively;  $df = 2$ ;  $P > 0.05$ ), parasitism ( $\chi^2 = 2.46$  and  $\chi^2 = 0.24$  for July and August, respectively;  $df = 2$ ;  $P > 0.05$ ), or unknown factors ( $\chi^2 = 3.27$  and  $\chi^2 = 1.27$  for July and August, respectively;  $df = 2$ ;

TABLE 1. SOURCES OF MORTALITY TO THREE SIZES OF *S. EXIGUA* LARVAE COLLECTED FROM COTTON.

| Source     | Percent mortality |         |         |         |          |         |
|------------|-------------------|---------|---------|---------|----------|---------|
|            | July              |         |         | August  |          |         |
|            | Small             | Medium  | Large   | Small   | Medium   | Large   |
| 1989       |                   |         |         |         |          |         |
| Unknown    | 34.8 Aa           | 5.7 Aa  | 10.0 Aa | 22.4 Aa | 11.0 Bb  | 3.5 Cb  |
| Parasitism | 4.4 Aa            | 2.9 Aa  | 5.0 Aa  | 3.7 Bb  | 24.2 Aa  | 4.5 Bb  |
| Disease    | 0.0 Aa            | 0.0 Aa  | 0.0 Aa  | 6.0 Cb  | 21.1 Ba  | 36.7 Aa |
| Total      | 39.2 Aa           | 8.6 Aa  | 15.0 Aa | 32.1 B  | 56.3 A   | 44.7 AB |
| N          | 80                | 35      | 46      | 512     | 408      | 353     |
| 1990       |                   |         |         |         |          |         |
| Unknown    | 47.1 Aa           | 25.4 Ba | 8.1 Cb  | 26.1 Aa | 12.3 Ab  | 17.7 Aa |
| Parasitism | 27.1 Aab          | 29.4 Aa | 14.8 Ba | 23.1 Aa | 28.1 Aa  | 22.4 Aa |
| Disease    | 12.9 Ab           | 9.1 Bb  | 4.0 Cc  | 6.0 Bb  | 15.8 Aab | 17.7 Aa |
| Total      | 87.1 A            | 63.9 B  | 26.9 C  | 55.2 A  | 56.2 A   | 57.8 A  |
| N          | 85                | 330     | 420     | 134     | 114      | 147     |

Means within months and rows followed by the same capital letter are not significantly different according to  $2 \times 2 X^2$  analysis,  $df = 1$ ,  $P < 0.05$ . Means within years and columns followed by the same lower case letter are not significantly different according to  $2 \times 2 X^2$  analysis,  $df = 1$ ,  $P < 0.05$  (SAS Institute 1988).

$P > 0.05$ ). Rates of disease were low throughout 1990, and in July, disease incidence decreased as larvae matured (Table 1). In August, rates of disease increased as larvae matured. "Unknown" causes killed  $\approx 20\%$  of the larvae from both 1990 cohorts. The highest rate of fungal infection (8.8%) was in large larvae from the August cohort.

Parasitism was one of the most common causes of mortality in both cohorts in 1990. In July, higher rates of parasitism were found in small and medium larvae than in large larvae. In August, however, there was no significant difference associated with host size.

Over both seasons, parasitoids emerged from 15% of collected *S. exigua*; 10, 26 and 11% of the large, medium and small larvae, respectively, were parasitized. The most common parasitoid found in both years was *Cotesia marginiventris* (Cresson) (det. P. M. Marsh). This species, which attacks the larvae of at least 21 lepidopteran species (Krombein et al. 1979), emerged from 95% of parasitized *S. exigua* collected as small or medium-sized larvae. *Meteorus rubens* (Nees) (det. P. M. Marsh) emerged from 3% of parasitized medium larvae. The tachinid *Lespesia aletiae* (Riley) (det. N. E. Woodley) emerged from 86% of the parasitized *S. exigua* collected as large larvae. In 1990, the gregarious, external parasitoid, *Euplectrus pathypenae* Howard emerged from three fourth instar *S. exigua*. *E. comstockii* Howard (det. M. E. Schauff), emerged from one fourth instar. The hyperparasitoids *Mesochorus discitergus* (Say) (det. R. W. Carlson) and *Spilochalcis hirtifemora* (Ashmead) (det. E. E. Grissell) also emerged from *S. exigua* collected in 1989.

Because larvae were not dissected to determine rates of parasitism, attack rates could not be estimated directly. However, 98% of *C. marginiventris* emerged from larvae collected as small or medium larvae in 1989 and 1990 combined. Ruberson et al. (1993) also found that *C. marginiventris* oviposited primarily in small and medium sized larvae. In contrast, only 3 of the 103 *L. aletiae* that emerged were from *S. exigua* collected as small or medium larvae. Thus, there was little contemporaneous mortality caused by the two most abundant parasitoids in this study.

Results of this experiment alone might lead to the conclusion that in August 1989, fungal disease of large larvae was the most important *S. exigua* mortality factor. *C. marginiventris* was the most abundant parasitoid both years, but its effects on *S. exigua* population densities is unclear from these data. Contemporaneous mortality, even for parasitism and disease, is not addressed by these results.

#### Mortality Tables

In 1989, different natural enemies were responsible for most mortality in the two *S. exigua* cohorts (Table 2). In July, marginal attack rates (m) and indispensable mortality (IM) from all causes combined were higher for small *S. exigua* than for the other size classes. Thus, death of small larvae appeared to be most important in the decline of this nonoutbreak larval population. However, much of the "unknown" mortality could be removed from the analysis if the high mortality rates for small larvae due to unknown causes were artifacts of

TABLE 2. DEATH RATE ANALYSES FOR *S. EXIGUA* LARVAE IN 1989.

| Mortality factor | Stage         | July  |       |                      |       |       | August |       |         |       |       |       |
|------------------|---------------|-------|-------|----------------------|-------|-------|--------|-------|---------|-------|-------|-------|
|                  |               | $l_x$ | $d_x$ | $m$                  | $v^1$ | IM    | $l_x$  | $d_x$ | $m$     | $v^1$ | IM    |       |
| Unknown          | Small larvae  | 1,000 | 342.8 | 0.357                | 0.287 | 0.148 | 1,000  | 223.8 | 0.237   | 0.204 | 0.027 |       |
| Parasitism       |               |       | 43.5  | 0.053                | 0.036 | 0.004 |        | 36.8  | 0.043   | 0.033 | 0.000 |       |
| Disease          |               |       | 0.0   | 0.000                | 0.000 | 0.000 |        | 59.5  | 0.069   | 0.054 | 0.007 |       |
| Predation        |               |       |       | (219.0) <sup>1</sup> | 0.175 | 0.175 | 0.057  |       | (124.6) | 0.085 | 0.085 | 0.008 |
| Total            |               |       |       | 610.3                | 0.586 | 0.498 | 0.263  |       | 444.7   | 0.434 | 0.377 | 0.051 |
| Unknown          | Medium larvae | 502   | 28.7  | 0.058                | 0.046 | 0.016 | 623    | 68.7  | 0.146   | 0.064 | 0.014 |       |
| Parasitism       |               |       |       | 14.4                 | 0.029 | 0.023 | 0.000  |       | 151.1   | 0.299 | 0.146 | 0.004 |
| Disease          |               |       |       | 0.0                  | 0.000 | 0.000 | 0.000  |       | 131.3   | 0.265 | 0.126 | 0.030 |
| Predation        |               |       |       | (110.5)              | 0.197 | 0.197 | 0.065  |       | (355.2) | 0.387 | 0.387 | 0.112 |
| Total            |               |       |       | 153.6                | 0.284 | 0.265 | 0.096  |       | 706.3   | 1.097 | 0.723 | 0.219 |
| Unknown          | Large larvae  | 369   | 36.9  | 0.103                | 0.085 | 0.031 | 172    | 6.1   | 0.045   | 0.030 | 0.005 |       |
| Parasitism       |               |       |       | 18.4                 | 0.053 | 0.042 | 0.000  |       | 7.7     | 0.057 | 0.039 | 0.001 |
| Disease          |               |       |       | 0.0                  | 0.000 | 0.000 | 0.000  |       | 63.3    | 0.386 | 0.322 | 0.054 |
| Predation        |               |       |       | (73.2)               | 0.154 | 0.154 | 0.048  |       | (29.8)  | 0.123 | 0.123 | 0.012 |
| Total            |               |       |       | 128.5                | 0.309 | 0.281 | 0.104  |       | 106.9   | 0.611 | 0.514 | 0.088 |
|                  | Pupae         | 265   |       |                      |       |       | 84     |       |         |       |       |       |

<sup>1</sup>Observed mortality ( $d_x$ ) due to predation was from caged data and includes predation and parasitism.



their being from a laboratory colony and of handling. Marginal probabilities for total mortality of each size class would then be nearly equal. Alternatively, if much "unknown" mortality was due to insecticides, it should be included in the analysis.

Marginal attack rates and IM for predation were similar for all *S. exigua* size classes, indicating that predators attacked different sizes equally. Indispensable mortality due to predation on all size classes combined and "unknown" mortality to all sizes were similar (Table 3). Parasitism and disease were of little importance to the July cohort, and 26.5% of larvae hatching in July survived to pupation (Table 2).

When *S. exigua* reached outbreak levels in August 1989, larval mortality for the generation increased dramatically (Table 2). Only 8.4% of the August cohort pupated. The highest levels of  $m$  and IM were for medium larvae. The parasitoid *C. marginiventris* parasitized  $\approx 30\%$  of the medium larvae in the August cohort. However, because much of the parasitism was contemporaneous with other mortality factors, only 15% of medium larvae from a single population would have produced parasitoids ( $v' = 0.146$ ). Indispensable mortality due to predation on medium larvae was much higher than IM for any other mortality factor affecting this size class (Table 2). Thus, predation was most responsible for reducing the proportion of this cohort that reached the most damaging developmental stage. Marginal probabilities of attack on medium larvae due to predation and of large larvae due to disease were about equal. However, disease of large larvae affected a smaller portion of the cohort than did predation on medium larvae. Thus, IM for predation on medium larvae was higher than IM for disease of large larvae. Parasitism caused little indispensable mortality to this generation. When all sizes were combined, IM for disease was higher than for any other factor (Table 3).

Because levels of disease, parasitism and unknown mortality were not different across insecticide treatments, mean percentages of mortality due to these factors were used to construct death-rate analyses in 1990 (Table 4). In July, in plots that had not been treated regularly with  $\lambda$ -cyhalothrin (untreated, diflubenzuron and combination), *S. exigua* suffered relatively high rates of

predation on medium and large larvae. About one-third of the large *S. exigua* in these plots were attacked by predators ( $m = 0.343$  and  $0.313$  in control and combination plots, respectively). Predation caused more indispensable mortality than any other factor attacking large larvae.

Nevertheless, because of high rates of parasitism of small and medium larvae (Table 4), parasitism was most responsible for reducing the density of this population before it reached the large larvae stage. When mortality to all sizes was combined (Table 5), parasitism caused more indispensable mortality to the July 1990 cohort than did predation or disease. In all insecticide treatments in both 1990 cohorts,  $m$  values for parasitoids were greater for small and medium larvae than for large larvae (Table 4). Thus, *C. marginiventris*, which caused almost all parasitism in small and medium larvae, appeared to be the most important parasitoid.

In August 1990, contemporaneous mortality was high in plots where predators were abundant (untreated and diflubenzuron). For example, when predation was precluded by collecting larvae, 28% of medium larvae were parasitized (Table 1). Thus, marginal attack rates for parasitism were high (Table 4). However, predation rates also were high in these plots. Predators attacked  $\approx 45\%$  of the medium larvae (Table 4). Because of contemporaneous mortality, only 15% of larvae in these plots would have died from parasitism ( $v' = 0.152$ ). In these plots, predation caused more indispensable mortality than any other factor in August (Table 5). Rates of predation were low (Table 4) where predator populations were virtually eliminated ( $\lambda$ -cyhalothrin treatment and in the combination plots in August), and parasitism was the most important mortality factor.

Natural mortality was greater for each cohort of *S. exigua* larvae when predators were present than when they were eliminated. Pupal  $l_x$  values were less in plots with dense predator populations than in plots with reduced predator populations. However, even in the 1990 plots with few predators (combination plots in August and both cohorts of  $\lambda$ -cyhalothrin-treated), larval mortality was greater than in 1989 (Table 2). These differences apparently were due to higher marginal attack rates by "unknown" factors and by parasitoids.

TABLE 3. TOTAL INDISPENSABLE MORTALITY<sup>1</sup> TO LARVAE IN 1989.

| Mortality factor | July  | August |
|------------------|-------|--------|
| Unknown          | 0.223 | 0.053  |
| Parasitism       | 0.005 | 0.006  |
| Disease          | 0.000 | 0.118  |
| Predation        | 0.208 | 0.086  |

<sup>1</sup>Indispensable mortality was calculated from larval death rates by subtracting the number entering the pupal stage when the mortality factor was included from the number entering the pupal stage when the factor was not included.

TABLE 4. DEATH-RATE ANALYSIS FOR *S. EXIGUA* LARVAE IN COTTON UNDER THREE INSECTICIDE REGIMES IN 1990.

| Stage                                  | Mortality factor | July  |       |       |       |       | August |       |       |       |       |
|--|------------------|-------|-------|-------|-------|-------|--------|-------|-------|-------|-------|
|  |                  | $l_x$ | $d_x$ | $m$   | $v'$  | IM    | $l_x$  | $d_x$ | $m$   | $v'$  | IM    |
| Untreated and diflubenzuron alone      |                  |       |       |       |       |       |        |       |       |       |       |
| Small larvae                           | Unknown          | 1,000 | 471.0 | 0.675 | 0.434 | 0.037 | 1,000  | 261.0 | 0.316 | 0.175 | 0.011 |
|  | Parasitism       |       | 271.0 | 0.440 | 0.234 | 0.018 |        | 231.0 | 0.284 | 0.155 | 0.009 |
|  | Disease          |       | 129.0 | 0.221 | 0.094 | 0.005 |        | 60.0  | 0.081 | 0.038 | 0.002 |
|  | Predation        |       | 38.0  | 0.038 | 0.038 | 0.001 |        | 321.0 | 0.321 | 0.032 | 0.009 |
|  | Total            |       | 909.0 | 1.374 | 0.800 | 0.077 |        | 873.0 | 1.001 | 0.689 | 0.053 |
| Medium larvae                          | Unknown          | 200   | 50.7  | 0.330 | 0.130 | 0.012 | 311    | 38.2  | 0.162 | 0.065 | 0.005 |
|  | Parasitism       |       | 58.7  | 0.373 | 0.151 | 0.014 |        | 87.3  | 0.338 | 0.152 | 0.011 |
|  | Disease          |       | 18.2  | 0.129 | 0.044 | 0.004 |        | 49.1  | 0.204 | 0.084 | 0.006 |
|  | Predation        |       | 95.0  | 0.476 | 0.476 | 0.018 |        | 140.2 | 0.451 | 0.451 | 0.016 |
|  | Total            |       | 222.6 | 1.308 | 0.800 | 0.077 |        | 314.8 | 1.155 | 0.751 | 0.058 |
| Large larvae                           | Unknown          | 40    | 3.2   | 0.090 | 0.053 | 0.002 | 77     | 13.7  | 0.229 | 0.096 | 0.007 |
|  | Parasitism       |       | 5.9   | 0.158 | 0.097 | 0.004 |        | 17.3  | 0.282 | 0.122 | 0.009 |
|  | Disease          |       | 1.6   | 0.045 | 0.026 | 0.001 |        | 13.7  | 0.229 | 0.096 | 0.007 |
|  | Predation        |       | 13.7  | 0.343 | 0.343 | 0.010 |        | 33.8  | 0.438 | 0.438 | 0.015 |
|  | Total            |       | 24.4  | 0.637 | 0.519 | 0.021 |        | 78.5  | 1.178 | 0.752 | 0.058 |
| Pupae                                  |                  | 19    |       |       |       |       | 19     |       |       |       |       |
| $\lambda$ -cyhalothrin + diflubenzuron |                  |       |       |       |       |       |        |       |       |       |       |
| Small larvae                           | Unknown          | 1,000 | 471.0 | 0.675 | 0.405 | 0.052 | 1,000  | 261.0 | 0.316 | 0.220 | 0.036 |
|  | Parasitism       |       | 271.0 | 0.440 | 0.218 | 0.026 |        | 231.0 | 0.284 | 0.194 | 0.032 |
|  | Disease          |       | 129.0 | 0.221 | 0.080 | 0.007 |        | 60.0  | 0.081 | 0.048 | 0.007 |
|  | Predation        |       | 102.0 | 0.102 | 0.102 | 0.003 |        | 147.0 | 0.147 | 0.147 | 0.011 |
|  | Total            |       | 973.0 | 1.438 | 0.814 | 0.120 |        | 699.0 | 0.827 | 0.610 | 0.102 |
| Medium larvae                          | Unknown          | 186   | 47.3  | 0.330 | 0.189 | 0.017 | 390    | 48.0  | 0.162 | 0.098 | 0.016 |
|  | Parasitism       |       | 54.8  | 0.373 | 0.220 | 0.020 |        | 109.7 | 0.338 | 0.231 | 0.038 |
|  | Disease          |       | 17.0  | 0.129 | 0.064 | 0.005 |        | 61.7  | 0.204 | 0.128 | 0.021 |
|  | Predation        |       | 44.0  | 0.236 | 0.236 | 0.009 |        | 64.8  | 0.166 | 0.166 | 0.013 |
|  | Total            |       | 163.1 | 1.068 | 0.708 | 0.067 |        | 284.2 | 0.870 | 0.622 | 0.107 |
| Large larvae                           | Unknown          | 54    | 4.4   | 0.090 | 0.055 | 0.003 | 147    | 26.1  | 0.229 | 0.170 | 0.023 |
|  | Parasitism       |       | 8.0   | 0.158 | 0.101 | 0.006 |        | 33.0  | 0.282 | 0.218 | 0.030 |
|  | Disease          |       | 2.2   | 0.045 | 0.027 | 0.002 |        | 26.1  | 0.229 | 0.170 | 0.023 |

TABLE 4. (CONTINUED) DEATH-RATE ANALYSIS FOR *S. EXIGUA* LARVAE IN COTTON UNDER THREE INSECTICIDE REGIMES IN 1990.

| Stage         | Mortality factor | July  |       |       |       |                              | August |       |       |       |       |
|---------------|------------------|-------|-------|-------|-------|------------------------------|--------|-------|-------|-------|-------|
|               |                  | $l_x$ | $d_x$ | $m$   | $v'$  | IM                           | $l_x$  | $d_x$ | $m$   | $v'$  | IM    |
| Pupae         | Predation        |       | 17.0  | 0.313 | 0.313 | 0.013                        |        | 0.0   | 0.000 | 0.000 | 0.000 |
|               | Total            |       | 31.6  | 0.607 | 0.497 | 0.027                        |        | 85.2  | 0.740 | 0.558 | 0.082 |
|               |                  | 27    |       |       |       |                              | 65     |       |       |       |       |
|               |                  |       |       |       |       | $\lambda$ -cyhalothrin alone |        |       |       |       |       |
| Small larvae  | Unknown          | 1,000 | 471.0 | 0.675 | 0.451 | 0.110                        | 1,000  | 261.0 | 0.316 | 0.258 | 0.033 |
|               | Parasitism       |       | 271.0 | 0.440 | 0.243 | 0.054                        |        | 231.0 | 0.284 | 0.228 | 0.029 |
|               | Disease          |       | 129.0 | 0.221 | 0.098 | 0.014                        |        | 60.0  | 0.081 | 0.056 | 0.007 |
|               | Predation        |       | 0.0   | 0.000 | 0.000 | 0.000                        |        | 0.0   | 0.000 | 0.000 | 0.000 |
|               | Total            |       |       | 871.0 | 1.336 | 0.792                        | 0.221  |       | 552.0 | 0.680 | 0.542 |
| Medium larvae | Unknown          | 208   | 52.8  | 0.330 | 0.247 | 0.035                        | 458    | 56.3  | 0.162 | 0.118 | 0.015 |
|               | Parasitism       |       | 61.0  | 0.373 | 0.288 | 0.042                        |        | 128.6 | 0.338 | 0.276 | 0.035 |
|               | Disease          |       | 18.9  | 0.129 | 0.083 | 0.011                        |        | 72.3  | 0.204 | 0.153 | 0.019 |
|               | Predation        |       | 0.0   | 0.000 | 0.000 | 0.000                        |        | 0.0   | 0.000 | 0.000 | 0.000 |
|               | Total            |       |       | 132.6 | 0.832 | 0.618                        | 0.094  |       | 257.2 | 0.704 | 0.547 |
| Large larvae  | Unknown          | 79    | 6.4   | 0.090 | 0.081 | 0.006                        | 207    | 36.7  | 0.229 | 0.112 | 0.022 |
|               | Parasitism       |       | 11.7  | 0.158 | 0.148 | 0.012                        |        | 46.4  | 0.282 | 0.143 | 0.028 |
|               | Disease          |       | 3.2   | 0.045 | 0.040 | 0.003                        |        | 36.7  | 0.229 | 0.112 | 0.022 |
|               | Predation        |       | 0.0   | 0.000 | 0.000 | 0.000                        |        | 71.1  | 0.343 | 0.343 | 0.032 |
|               | Total            |       |       | 21.3  | 0.294 | 0.268                        | 0.021  |       | 190.9 | 1.083 | 0.710 |
| Pupae         |                  | 58    |       |       |       |                              | 60     |       |       |       |       |

TABLE 5. TOTAL INDISPENSABLE MORTALITY<sup>1</sup> TO *S. EXIGUA* LARVAE DUE TO FOUR MORTALITY FACTORS UNDER THREE INSECTICIDE REGIMES IN 1990.

| Mortality factor | Untreated and diflubenzuron |        | $\lambda$ -cyhalothrin + diflubenzuron |        | $\lambda$ -cyhalothrin only |        |
|------------------|-----------------------------|--------|--|--------|-----------------------------|--------|
|                  | July                        | August | July                                   | August | July                        | August |
| Unknown          | 0.080                       | 0.031  | 0.114                                  | 0.104  | 0.242                       | 0.097  |
| Parasitism       | 0.057                       | 0.047  | 0.082                                  | 0.159  | 0.172                       | 0.147  |
| Disease          | 0.011                       | 0.019  | 0.015                                  | 0.064  | 0.032                       | 0.059  |
| Predation        | 0.039                       | 0.073  | 0.031                                  | 0.027  | 0.000                       | 0.032  |

<sup>1</sup>Indispensable mortality was calculated from larval death rates by subtracting the number entering the pupal stage when the mortality factor was included from the number entering the pupal stage when the factor was not included.

## DISCUSSION

This study demonstrated that, as Luck et al. (1988) suggested, biases associated with the use of a single natural enemy exclusion method can be at least partially overcome by combining methods. For example, if our experiment had included only exclusion cages (no  $\lambda$ -cyhalothrin treatments), effects of natural enemies would have been apparent in August because of higher survival when *S. exigua* were protected from natural enemies by the cages (Fig 4C). In July, however, cage effects also reduced *S. exigua* survival. Evidence included greater mortality in partial exclusion cages than in total exclusion cages, coupled with similar survival in total exclusion cages and with no predator exclusion (Fig. 4A). The insecticide exclusion experiment showed that  $\lambda$ -cyhalothrin reduced predator populations and that *S. exigua* was abundant in treated plots. However, this experiment did not eliminate reduced parasitism or disease incidence, trophobiosis or hormoligosis as mechanisms of outbreak induction. Combining cage exclusion with insecticide exclusion demonstrated that, in plots treated with insecticides, the increase in *S. exigua* populations resulted from fewer natural enemies. Because rates of disease, parasitism and "unknown" mortality of collected larvae were not different across insecticide treatments, differences in *S. exigua* population densities (Fig. 2) between insecticide treatments could be attributed to differences in predation.

The death rate analyses for experimentally manipulated populations effectively demonstrated which mortality factors were important in both insecticide-treated and untreated cotton. This analysis also showed that different factors were important for mortality in outbreak and nonoutbreak populations.

Estimating predation rates based on exclusion cages with the formula of Royama (1981), which subtracts effects of parasitism from combined effects of parasitism and predation, suffers some of the shortcomings of the common practice of assigning unexplained mortality to predation. If rates of parasitism are low, as in 1989, estimates

of predation should be relatively accurate. In 1990, observed mortality due to parasitism of collected larvae usually was at least equal to estimates of mortality due to a combination of parasitism and predation in exclusion cages. Thus, if estimates of mortality due to combined factors are accurate and parasitism is common, predation may be seriously underestimated.

As in California cotton (Eveleens et al. 1973; Hogg & Gutierrez 1980), when predator populations were not disrupted with insecticides, *S. exigua* populations were held below outbreak densities primarily by polyphagous predators. In California, adult and immature *Geocoris pallens* Stalh, *Orius tristicolor* (White), *Nabis americanus* Carayon and immature *Chrysopa carnea* Stephens were important predators of *S. exigua* eggs and newly eclosed larvae (Eveleens et al. 1973). Many of the same, or closely related species, are common in Alabama cotton (Gaylor & Gilliland 1976; Fleischer et al. 1985). Fire ants also have been reported to be effective *S. exigua* predators in Alabama (Cobb 1973). All of these polyphagous predators, except adult *Nabis* spp., are capable of entering cages covered by the medium mesh, and may have contributed to natural control of *S. exigua*. However, only fire ants were more abundant in plots with few *S. exigua* larvae than in plots with dense larval populations. Thus, circumstantial evidence indicates that fire ants were key *S. exigua* predators in plots that were not treated with  $\lambda$ -cyhalothrin.

When populations of predators were not reduced by insecticides, as in 1990, the parasitoid, *C. marginiventris*, was relatively unimportant as a larval mortality agent. The species was important in reducing populations of small and medium *S. exigua* in both insecticide-treated and untreated plots in July 1990. Despite high levels of parasitism by *C. marginiventris* in August 1990, the parasitoid was not responsible for the sparse *S. exigua* populations in cotton that had dense predator populations. When predators were present, they attacked both parasitized and unparasitized *S. exigua*, and little damage to cotton occurred. Parasitism was insufficient to prevent

an economically damaging *S. exigua* population from occurring in plots with few predators. Despite the presence of the parasitoid, defoliation of cotton treated with  $\lambda$ -cyhalothrin was severe in August. Thus, the presence of a sufficient predator population was necessary to prevent an outbreak of *S. exigua*. This does not imply that parasitoids were unimportant in regulating populations of *S. exigua*. Our treatments did not affect parasitoid attack rates. If parasitism had been reduced by insecticide applications, we would have expected to observe even more dramatic increases in beet armyworm larval populations.

In California, egg predation (39%) also was important to *S. exigua* population regulation (Hogg & Gutierrez 1980). We did not estimate egg mortality, but it probably was not important in  $\lambda$ -cyhalothrin-treated plots in 1990 because predator populations were sparse. If 39% egg predation were added to our death-rate analyses for plots with predators,  $I_x$  for small larvae would be reduced to 610. However, indispensable mortality for egg predation in plots with predators would be only 0.007 for each cohort in control plots and 0.010 in combination plots in July 1990. In 1989,  $I_M$  for eggs would have been 0.103 and 0.003 for July and August, respectively. Thus, 39% egg predation would have been important only in July 1989. Instead of determining which factors regulate densities of an entire host generation, pest managers may want to know which mortality agents in an agroecosystem reduce the pest population density before it reaches a damaging stage or before it enters sites where it is protected from natural mortality agents or pesticides. Small and medium size *S. exigua* cause less damage to cotton than do large larvae. Therefore, a primary objective of some cotton IPM programs is to establish conditions that favor mortality by natural enemies to *S. exigua* eggs and small and medium size larvae, so that economically damaging numbers do not survive to the large size class.

Survival past the larval stage is important only if survivors contribute to a subsequent generation that causes economic injury. Thus, mortality to large larvae of the July *S. exigua* generation is important only if survivors contribute substantially to outbreaks in August. Mortality to pupae could be important, but pupal mortality apparently is negligible (Hogg & Gutierrez 1980). The occurrence of each *S. exigua* generation is associated with peaks in adult flight activity (Trumble & Baker 1984). The role of immigration in *S. exigua* outbreaks in August is unknown but may be important. Thus, the relative importance of the July *S. exigua* generation and of immigration in August to outbreaks in August is unclear.

The August *S. exigua* generation does not contribute to subsequent damage to cotton. Most cotton in Alabama is not susceptible to *S. exigua* damage after August, and *S. exigua* apparently

does not readily overwinter in the state. Thus, natural mortality to early developmental stages of the August generation should be more important to the pest manager than is mortality to the entire generation. Diseases of large larvae were most important in the decline of the *S. exigua* outbreak in August 1989. However, natural mortality due to predation on early developmental stages may have been more important than mortality to later stages if the goal was to avoid a damaging outbreak.

Unlike most population ecologists, pest managers often are confronted with identifying natural control agents in an agroecosystem that has been modified by insecticides. Despite the risks of insecticide-induced outbreaks of *S. exigua*, pyrethroids usually are applied in southern Alabama cotton for control of *Helicoverpa zea* (Boddie) and *Heliothis virescens* (F.). Consequently, the pest manager also may want to know which mortality factors might be manipulated to reduce the density of a pest population that has reached damaging levels as a result of insecticide applications. Adult and pupal *C. marginiventris* are susceptible to pyrethroids (Ruberson et al. 1993), but during 1990, rates of parasitism by this species were not different in treated and untreated cotton. When a broad-spectrum insecticide must be applied to control other pests, parasitism and disease may be the most important natural mortality factors remaining. If techniques can be developed for augmenting populations of *C. marginiventris* or diseases, these natural enemies might have potential in applied biological control programs.

Levins & Wilson (1980) listed several reasons for the lack of application of ecological theory to agroecosystems. A primary reason for this situation is the different perspectives of the basic ecologist and the pest manager. However, questions of interest to both the ecologist and the pest manager can be addressed by combining experimental methods that are commonly used in studying natural control in agroecosystems with death-rate analyses or with life tables.

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## OCCURRENCE OF ENTOMOPATHOGENS OF *SPODOPTERA FRUGIPERDA* (LEPIDOPTERA: NOCTUIDAE) IN THE MEXICAN STATES OF MICHOACÁN, COLIMA, JALISCO AND TAMAULIPAS

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### ABSTRACT

Fall armyworm, *Spodoptera frugiperda* (J. E. Smith) (FAW) larvae and soil samples were collected from corn and sorghum fields in the Mexican states of Michoacán, Colima, and Jalisco during August 1998. Additional FAW larvae were collected from a sorghum field in Tamaulipas, Mexico in September. A total of 2219 FAW larvae from 20 locations and 76 soil samples from 19 locations were examined for indigenous FAW biological control agents. Four species of entomopathogenic fungi representing two classes, Zygomycetes (Entomophthorales) and Hyphomycetes (*Beauveria bassiana*, *Nomuraea rileyi*, and *Hirsutiella* sp.) were recovered from 43 (1.94%) of FAW larvae. An unidentified microsporidian was collected from 32 (1.44%) of FAW larvae, 29 from Colima, 2 from Jalisco, and 1 from Michoacán. Forty nine larvae (2.21%) parasitized by mermithid nematodes were collected in the state of Colima. Two (0.09%) larvae infected with ascovirus were collected in Tamaulipas. Three species of Hyphomycetes (*Paecilomyces fumosoroesus*, *B. bassiana*, and *Metarhizium anisopliae*) were isolated from soil samples using *Galleria mellonella* larval traps. Entomopathogenic nematodes (*Steinernema* sp. and *Heterorhabditis* sp.) were recovered from soil samples from 5 of 19 localities using *Galleria mellonella* larval traps. *Bacillus thuringiensis* was isolated from soil samples from 12 locations. The most widely distributed microbial control agent on FAW larvae in the Western Coast of Mexico was the fungus *N. rileyi*, and from soil were the bacterium *B. thuringiensis* and steinernematid nematodes. The microsporidian was found predominantly in Colima and the mermithid nematodes only in Colima. Thus, Colima had the highest total percent mortality (9.67%) due to fungi, microsporidia and mermithids.

Key Words: Fall armyworm, biological control, maize, *Nomuraea rileyi*, mermithid nematode, microsporidia

### RESUMEN

Larvas de gusano cogollero, *Spodoptera frugiperda* (J. E. Smith) (FAW) y muestras de suelos fueron colectadas de campos cultivados de maíz y sorgo, en los estados mexicanos de Michoacán, Colima y Jalisco, durante Agosto de 1998. Más larvas de FAW fueron colectadas de sorgo en Tamaulipas, México en Septiembre. Un total de 2219 larvas de FAW provenientes de 20 localidades y 76 muestras de suelos de 19 localidades fueron examinadas en búsqueda de agentes locales de control biológico de FAW. Cuatro especies de hongos entomopatógenos de dos clases, Zygomycetes (Entomophthorales) e Hyphomycetes (*Beauveria bassiana*, *Nomuraea rileyi*, e *Hirsutiella* sp.) fueron recuperados de 43 (1.94%) de las larvas. Un microsporidio no identificado fue colectado de 32 (1.44%) de las larvas, 29 de Colima, dos de Jalisco, y uno de Michoacán. Cuarenta y nueve larvas (2.21%) parasitadas por nematodos mermithidos fueron colectadas de Colima. Dos larvas (0.09%) infectadas con ascovirus fueron colectadas de Tamaulipas. Tres especies de Hyphomycetes (*Paecilomyces fumosoroesus*, *B. bassiana*, y *Metarhizium anisopliae*) se aislaron de muestras de suelos usando las trampas de larvas de *Galleria mellonella*. Nematodos entomopatógenos (*Steinernema* sp. y *Heterorhabditis* sp.) se recuperaron de muestras de suelos en 5 de 19 localidades usando las trampas de larvas de *G. mellonella*. La bacteria *Bacillus thuringiensis* fue aislada de muestras de suelos de 12 localidades. El agente de control microbiano más ampliamente distribuido sobre larvas de FAW en la Costa Occidental de México fue el hongo *N. rileyi* y del suelo, *B. thuringiensis* y los nematodos entomopatógenos. El microsporidio fue encontrado predominante-

mente en Colima y los nematodos mermitidos sólo en Colima. Así, Colima tuvo el porcentaje de mortalidad más alto (9.67%) debido a hongos, microsporidios y mermitidos.

Corn or maize, *Zea mays* L., is one of the major sources of animal and human foods in the Americas and is one of the most valuable field crops in the U.S.A. It is attacked by a variety of insect pests including, one of the more destructive of these pests, the fall armyworm (FAW), *Spodoptera frugiperda* (J. E. Smith). The FAW causes damage in all plant growth stages, often limiting production due to severe damage to, or complete destruction of, whorl-stage plants (Wiseman et al. 1967, 1996). The use of microbial control is a potentially valuable alternative to chemical pesticides with their high cost, possible pest resurgence, development of resistance, and environmental contamination. The strategies for using pathogens in biological control of insect pests are determined primarily by the interactions among pathogens, insect host, and environment, including the plant to be protected (Hamm 1984). Thus, a first step to develop a microbial control program is the knowledge of the occurrence of insect pathogens, in order to utilize them as a component of an integrated pest management scheme.

The FAW is reported to be susceptible to viruses, fungi, protozoa, bacteria, and nematodes (Steinhaus & Marsh 1962; Gardner & Fuxa 1980; Fuxa 1982; Hughes et al. 1984; Agudelo-Silva 1986; Remillet & Silvain 1988; Richter & Fuxa 1990; Raulston et al. 1992; Molina-Ochoa et al. 1996), but their occurrence and distribution may vary with their habitat. Geographical location and agricultural practices, as well as pesticide use, may have an impact on the occurrence of natural control agents in the host population or in the soils (Fargues et al. 1992; Rogers & Marti 1992; Sosa-Gomez & Moscardi 1994; Vanninen 1996; Mietkiewski et al. 1997).

There is an increased interest in developing biological control methods for FAW in Mexico, but its natural enemy complex (particularly pathogens) is poorly known. More than three decades ago a parasitic nematode (Mermithidae) was reported to infest 21-53% of FAW larvae from Cotaxtla, Veracruz, Mexico (Alcocer and Méndez 1965). More recently, Lezama-Gutiérrez et al. (1996) assessed the virulence of some isolates of *Metarhizium anisopliae* (Metch.) Sor., *Beauveria bassiana* (Bals.) Wuill., and *Nomuraea rileyi* (F.) Samson, obtained from FAW larvae collected in the state of Colima, Mexico, against eggs and neonates of FAW. However, no detailed studies have been conducted on the occurrence of FAW pathogens from Colima, Michoacán, Jalisco, or Tamaulipas, Mexico, although data from other countries suggest that many entomopathogenic fungi and nematodes are ubiquitous inhabitants of the soil (Chan-

der et al. 1997). This paper reports the natural occurrence of entomopathogens and nematodes on FAW larvae and in the soil of corn and grain sorghum fields from the states of Colima, Jalisco, Michoacán, and Tamaulipas, Mexico.

## MATERIALS AND METHODS

### *Isolation of Pathogens from FAW Larvae*

During August of 1998 collections of FAW larvae were made from whorl-stage corn and grain sorghum fields in the states of Colima, Jalisco, and Michoacán, and 1 collection from fruiting corn in Jalisco. A single collection of FAW larvae was made from whorl-stage grain sorghum in Tamaulipas in September. Concurrently, four soil samples were obtained from each location in the first three states. Locations 12 and 18 comprise combinations of collections from adjacent fields of whorl-stage maize and sorghum. Sample size ranged from 25 to 300 FAW larvae per field. The number collected is corrected by subtracting the number that died from injury or unknown causes during the first days after collection. Collection data and percent infection by pathogens and nematodes is presented in Table 1. Larval mortality due to insect parasitoids is reported elsewhere (Molina-Ochoa et al., in press).

The larvae were placed individually in 30 cc plastic cups with pinto bean diet (Burton & Perkins 1989) and held in the laboratory to record the larvae infected by entomopathogens and mermithid nematodes. Mermithid nematodes that emerged from larvae were collected and placed in 70% alcohol. Larvae showing signs of virus infection or infection by microsporidia were examined microscopically for occlusion bodies or spores. Microsporidian and virus infected tissues from field-collected insects were fixed in a modified Karnovsky's fixative, postfixed in OsO<sub>4</sub>, and embedded in epoxy resin; sections were cut, stained and examined as described by Styer et al. (1987). The ascovirus was examined by negative-stain electron microscopy using methods described by Hamm et al. (1992). Ascovirus was diagnosed by the presence of vesicles in stunted larvae and transmitted to other larvae using a cactus spine (Hamm et al. 1986). Final identification was made by electron microscopy (Federici et al. 1991).

Dead larvae showing signs of fungus infection were placed in a plastic Petri dish (60 × 10 mm) lined with filter paper moistened with sterile distilled water, until the fungus sporulated on the insect surface. *Nomuraea rileyi* was isolated from dead larvae on medium composed of 200 ml of



TABLE 1. LOCATION, DATE, CROP, SAMPLE SIZE, AND TOTAL PERCENTAGE FALL ARMYWORM LARVAE INFECTED BY PATHOGENS AND MERMITHID NEMATODES COLLECTED FROM CORN (C) OR SORGHUM (S) IN THE MEXICAN STATES OF MICHOACÁN (M), COLIMA (C), JALISCO (J), AND TAMAULIPAS (T).

| Code | Location           | Date     | Crop     | No. coll. | Percentage infected |
|------|--------------------|----------|----------|-----------|---------------------|
| M 1  | Jazmin             | 08/07/98 | C        | 25        | 8.0                 |
| M 2  | El Batillero       | 08/07/98 | C        | 26        | 0.0                 |
| M 3  | La Sidra           | 08/07/98 | C        | 84        | 2.4                 |
| M 4  | La Sidra           | 08/08/98 | C        | 89        | 9.0                 |
| M 5  | El Hueso           | 08/08/98 | C        | 102       | 2.0                 |
| M 6  | Carreras           | 08/08/98 | C        | 109       | 0.9                 |
| C 7  | Mezcales           | 08/12/98 | C        | 143       | 3.5                 |
| C 8  | Los Clomos         | 08/12/98 | C        | 84        | 14.3                |
| C 9  | Cerro Colorado     | 08/13/98 | C        | 121       | 9.2                 |
| C 10 | El Bordo           | 08/13/98 | C        | 114       | 17.5                |
| C 11 | Crucero de Tepames | 08/13/98 | C        | 89        | 10.1                |
| C 12 | Peña Blanca        | 08/13/98 | C & S    | 219       | 13.2                |
| C 13 | El Narajito        | 08/14/98 | C        | 171       | 6.4                 |
| J 14 | Los Pozos          | 08/19/98 | S        | 81        | 7.4                 |
| J 15 | Apastepe           | 08/19/98 | C        | 89        | 4.5                 |
| J 16 | Los Depositos      | 08/19/98 | C        | 89        | 3.4                 |
| J 17 | Sayula             | 08/19/98 | C        | 89        | 9.0                 |
| J 18 | Sayula             | 08/20/98 | C & S    | 177       | 4.5                 |
| J 19 | Sayula             | 08/20/98 | C (ears) | 18        | 16.7                |
| T 20 | El Mante           | 09/22/98 | S        | 300       | 0.3                 |

"V8" vegetable juice, 3 g CaCO<sub>3</sub>, 5 g glucose, 2 g yeast extract, 15 g agar, and 800 ml distilled water (Fargues & Rodriguez-Rueda 1980). Other fungal species were grown on Sabouraud-Dextrose agar enriched with 1% (w/v) yeast extract (SDAY), with 500 ppm chloramphenicol (Lezama-Gutiérrez et al. 1996) except the entomophthorales which were not isolated.

#### Isolation of Entomopathogens from Soil

Four soil samples, from corn or sorghum fields, were collected from each of 19 locations from the states of Michoacán, Colima, and Jalisco. In every location approximately 2 kg of soil was collected from four points a few meters apart by digging to a depth of 10-15 cm with a small spade. These subsamples were combined to form a sample. The soil samples were put in plastic bags and taken to the laboratory and stored at 25°C until processing. The storage time ranged from a few days to three weeks. For processing, the soil was thoroughly mixed and passed through a 0.4 mm mesh sieve to break or separate any coarse lumps of soil or litter.

In order to isolate the entomopathogenic fungi or nematodes, larvae of laboratory-reared *Galleria mellonella* (L.) were used as bait (Chandler et al. 1997; Bedding and Akhurst 1975). Four groups of sieved soil from each location were placed in plastic pots and five last instar bait larvae were released into each pot. Pots were incubated at room temperature (25°C) in the dark for 10 days (Zimmermann 1986; Woodring & Kaya 1988).

Dead, intact larvae were removed and surface-sterilized in 1% sodium hypochlorite for 3 min, then washed three times in sterile distilled water and placed on damp filter paper within a sealed Petri dish 5.5 cm diameter, and incubated at 25°C for 12 days (Chandler et al. 1997). Entomopathogenic fungi were isolated from the bait larvae using SDAY, with 500 ppm of chloramphenicol (Lezama-Gutiérrez et al. 1996). The fungi were identified by microscopic inspection of morphological characteristics *in situ* or after isolation in SDAY, according to the criteria by Brady (1979) and Samson et al. (1988). Nematodes were separated to genera by identifying coloration of dead bait larvae according to Woodring and Kaya (1988).

To isolate *Bacillus thuringiensis* Berliner from the soil, 1 g samples from each location were placed in 50 ml of sterile distilled water in bioassay tubes, mixed for 3 min, and heated to 80°C for 10 min. After heating, 100 µl, of each sample, was placed on nutrient agar in four Petri dishes. Petri dishes were incubated at 30°C for 72 h, and colonies were microscopically examined after fixation and staining with methylene blue cotton. The presence of protein crystals was utilized as identification criteria of *B. thuringiensis*, according to Chaufaux et al. (1997).

#### RESULTS

Out of 2219 FAW larvae collected from 20 locations, the percentage infected by pathogens and mermithid nematodes ranged from 0 to 17.5 (Table

1), 77 larvae (3.47%) were killed by pathogens and 49 (2.21%) by mermithid nematodes. Four species of entomopathogenic fungi, represented by three Hyphomycetes, *Beauveria bassiana* (Bals.) Vuill., *Nomuraea rileyi* (F.) Samson, and *Hirsutella* sp., and one Zygomycete, Entomophthorales, were recovered (Table 2). *Nomuraea rileyi* was the most abundant and widely distributed fungus attacking FAW larvae in the three Western Mexican States and accounted for most of the pathogen-induced mortality of FAW larvae collected in Michoacán and Jalisco (Table 2). Two larvae infected with *B. bassiana* and a single larva infected by a member of the Entomophthorales were collected in Jalisco. A single larva infected with *Hirsutella* sp. was collected from Colima.

Mermithid infected FAW larvae were found only in the state of Colima and were more numerous than fungus infected larvae at 3 of the 7 locations.

Microsporidia infected larvae were found predominately in Colima and were more numerous than fungus infected larvae at 4 of the 7 locations in Colima (Table 2). Collections from Michoacán and Jalisco had 1 and 2 microsporidia infected larvae, respectively. All mortality appeared to be due to the same unidentified microsporidian which formed clumps of numerous thick-walled spores with no apparent sporophorous vesicle (Fig. 1A). The infected larvae showed no obvious symptoms prior to death, but after death were often dry and fragile, resembling the ash of a cigarette.

Four of five collections with rates of infection greater than ten percent were from the state of Colima (Table 1) and can be attributed to the higher rates of infection by mermithid nematodes and microsporidia in Colima than in the other states (Table 2). The only other collection with more than ten percent infection (J19) was a small collection of larvae from ears of corn in Jalisco which was entirely due to *Nomuraea rileyi*.

Two ascovirus infected larvae from Tamaulipas were the only virus infected larvae identified. The ascovirus (Fig. 1 B & C) resembled the ascovirus collected from FAW in Georgia and Florida, forming vesicles in the fat body but not in the epidermis or tracheal epithelium (Hamm et al. 1998).

#### Entomopathogens from Soil

The most numerous and widely distributed entomopathogen isolated from soil samples was *B. thuringiensis* which was isolated from 4 of 6 locations in Michoacán, 7 of 7 locations in Colima and 1 of 6 locations in Jalisco.

Three species of entomopathogenic fungi were recovered from soil samples. *Metarhizium anisopliae* was recovered from 5 locations, 2 of 7 from Colima and 3 of 6 from Jalisco. *Beauveria bassiana* was recovered from 4 locations, 1 from Michoacán, 1 from Colima, and 2 from Jalisco. *Paecilomyces fumosoroseus* was recovered from a single soil sample from Colima.

TABLE 2. PERCENTAGE OF FALL ARMYWORM LARVAE INFECTED BY VARIOUS PATHOGENS AT EACH LOCATION.

| Code* | <i>N. r.</i> | <i>Ent.</i> | <i>Hir.</i> | <i>B. b.</i> | Mer. | Mic. | Asc. |
|-------|--------------|-------------|-------------|--------------|------|------|------|
| M 1   | 8.0          | 0           | 0           | 0            | 0    | 0    | 0    |
| M 2   | 0            | 0           | 0           | 0            | 0    | 0    | 0    |
| M 3   | 2.4          | 0           | 0           | 0            | 0    | 0    | 0    |
| M 4   | 9.0          | 0           | 0           | 0            | 0    | 0    | 0    |
| M 5   | 1.0          | 0           | 0           | 0            | 0    | 1.0  | 0    |
| M 6   | 0.9          | 0           | 0           | 0            | 0    | 0    | 0    |
| C 7   | 2.1          | 0           | 0           | 0            | 1.4  | 0    | 0    |
| C 8   | 9.5          | 0           | 0           | 0            | 2.4  | 2.4  | 0    |
| C 9   | 0.8          | 0           | 0           | 0            | 4.1  | 4.1  | 0    |
| C 10  | 0            | 0           | 0           | 0            | 14.9 | 2.6  | 0    |
| C 11  | 0            | 0           | 0           | 0            | 0    | 10.1 | 0    |
| C 12  | 1.4          | 0           | 0           | 0            | 9.1  | 2.7  | 0    |
| C 13  | 1.8          | 0           | 0.6         | 0            | 1.8  | 2.3  | 0    |
| J 14  | 6.2          | 0           | 0           | 1.2          | 0    | 0    | 0    |
| J 15  | 3.4          | 0           | 0           | 0            | 0    | 1.1  | 0    |
| J 16  | 2.2          | 0           | 0           | 0            | 0    | 1.1  | 0    |
| J 17  | 6.7          | 1.1         | 0           | 1.1          | 0    | 0    | 0    |
| J 18  | 4.5          | 0           | 0           | 0            | 0    | 0    | 0    |
| J 19  | 16.7         | 0           | 0           | 0            | 0    | 0    | 0    |
| T 20  | 0            | 0           | 0           | 0            | 0    | 0    | 0.7  |

\*Locations are described in Table 1.

*N. r.* = *Nomuraea rileyi*, *Ent.* = *Entomophthora* sp., *Hir.* = *Hirsutella* sp., *B. b.* = *Beauveria bassiana*, Mer. = Mermithid nematode, Mic. = Microsporidia, Asc. = Ascovirus.

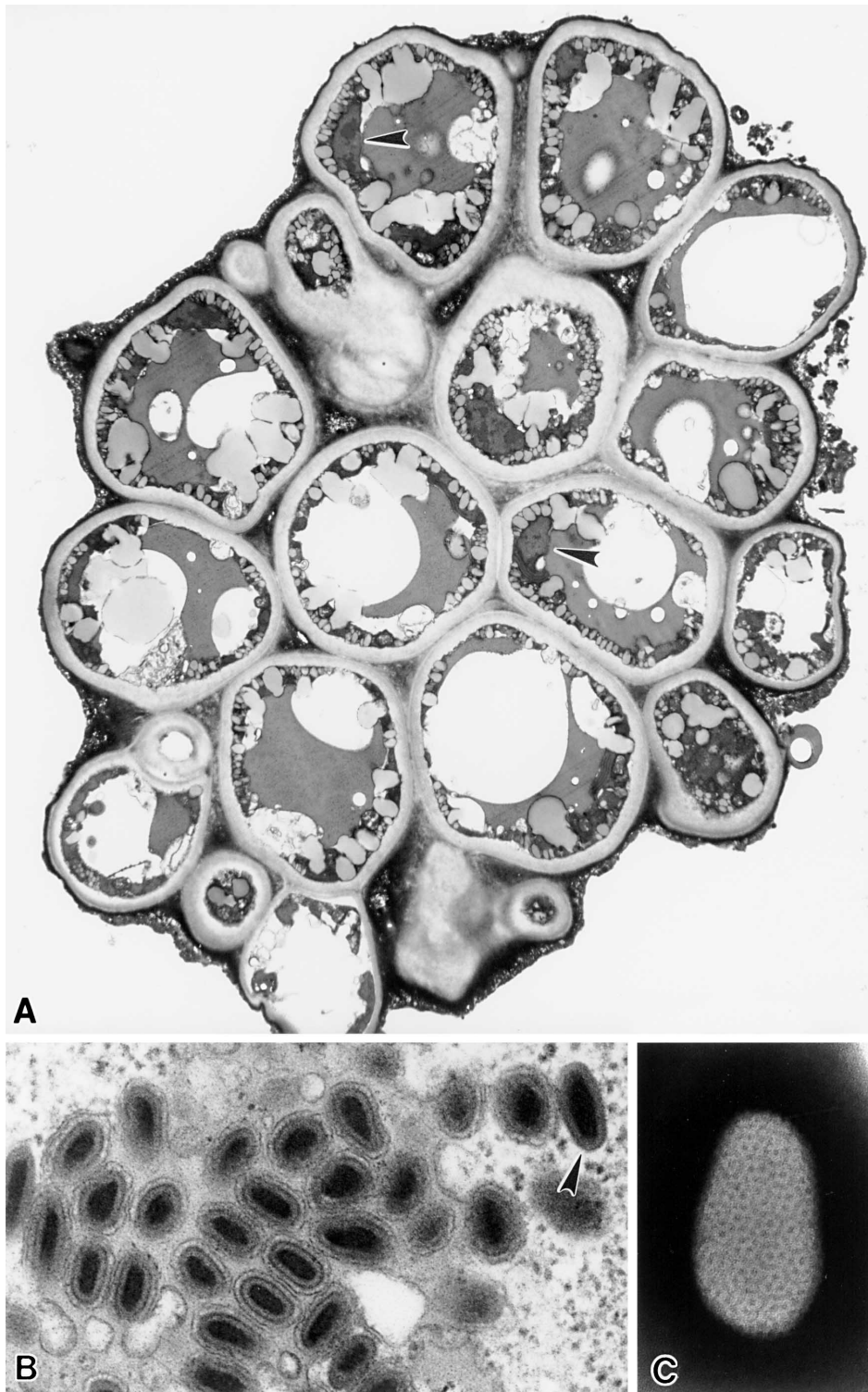


Fig. 1. A) Cluster of microsporidia spores from fall armyworm larva collected in Colima, Mexico, 6,200 $\times$ , arrows pointing to nuclei. B & C) Ascovirus from fall armyworm larva collected in Tamaulipas, Mexico: B, Enlargement of viral inclusion body showing unenveloped virus (arrowhead) and enveloped virions 58,000 $\times$ . C, negative stain of virion showing characteristic surface of envelop 150,000 $\times$ .

Two genera of entomogenous nematodes were collected from soil samples. Steinernematid nematodes were collected from 3 of 4 locations in Michoacán and 1 location in Colima; heterorhabditid nematodes were collected from only one location in Colima.

#### DISCUSSION

*Nomuraea rileyi* is recognized as an important pathogen of many insect pests, especially lepidopteran larvae (Ignoffo 1981; Carruthers & Soper 1987), and has been reported infecting FAW in Puerto Rico, Colombia, Honduras, Mexico, and U.S.A. (Gardner & Fuxa 1980; Ignoffo 1981; Wheeler et al. 1989; Sánchez-Peña 1990; Pantoja & Fuxa 1992). *Entomophthora aulicae* was reported infecting FAW and other noctuid larvae in sorghum in Georgia (Hamm 1980). *Erynia radicans* was reported infecting FAW in Venezuela (Agudelo-Silva 1986).

*Spodoptera frugiperda* (= *Laphygma frugiperda*) has been reported to be infected by *Nosema laphygmae* Weiser, *Nosema trichoplusiae* Tanabe and Tamashiro and *Vairimorpha necatrix* (Kramer) (Bulla & Cheng 1977; Gardner & Fuxa 1980). *Nosema laphygmae* was described from larvae, pupae and adults from the vicinity of Caracas, Venezuela (Weiser 1959). *Vairimorpha necatrix* was reported naturally infecting FAW by Patel and Habib (1988). *Nosema trichoplusiae* was demonstrated to infect FAW in the laboratory. Unidentified microsporidia were reported from FAW in Louisiana by Fuxa (1982), in Venezuela by Agudelo-Silva (1986), and in Puerto Rico by Pantoja & Fuxa (1992). We did not find *Nosema* or *Vairimorpha* in our collections; the unidentified microsporidian that we found was obviously not in either of those genera based on the arrangement of spores. We were unable to infect FAW larvae using dried spores that were a few weeks old and thus were unable to study the developmental stages of the microsporidian.

The ascovirus isolated from FAW larvae collected in Tamaulipas is the first report of an ascovirus from Mexico. While the ascovirus can cause significant mortality in FAW (Hamm et al. 1986) it interferes with development of braconid parasitoids (Hamm et al. 1985). Although baculoviruses, nuclear polyhedrosis virus and granulosis virus, have been reported infecting FAW in many areas, they were not found in this survey. Fuxa (1982) reported that in Louisiana, NPV was more prevalent in fall armyworms infesting pastures than in those infesting corn or sorghum until mid July or early August, but the eventual infection rates were similar. He suggested that the lag in virus in corn and sorghum could be because rain and other physical agents cannot move the NPV from the soil reservoir to vegetation as easily as in grass. Also, the faster growth of corn

or sorghum may produce more uncontaminated leaf surface and larvae may not move from plant to plant as readily as in pastures.

Mermithid nematodes have been reported infecting FAW in various parts of its range but have not been studied extensively. Valincente (1986) reported FAW attacked by mermithid nematodes in Brazil. Pair et al. (1986) reported an unidentified mermithid attacking FAW in South Carolina. Wheeler et al. (1989) reported *Hexameris* sp. from FAW in Honduras where it made up 23% of the natural enemy complex of FAW on corn. *Hexameris* has been reported to cause 8-100% FAW mortality in Mexico (Alcocer & Méndez 1965) and over 50% FAW mortality in Nicaragua (Van Huis 1981).

*Steinernema riobravis* is an important control agent for prepupae and pupae of FAW and corn earworm, *Helicoverpa zea* (Boddie), in cornfields of the Lower Rio Grande Valley (Raulston et al. 1992; Cabanillas et al. 1994) where the nematode appears to be naturally selected for the subtropical semi-arid environment.

The distribution of the various entomopathogens indicates a potential for increasing biological control by moving some of the pathogens and nematodes from one area to another. Additional research is needed on the identification and biology of the microsporidian and the mermithid nematode to determine their potential for biological control. Also, additional research is needed to determine the species and strains of the entomopathogenic nematodes, Steinernematidae and Heterorhabditidae, isolated from the soil and their potential for biological control of fall armyworm.

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## A SURVEY OF FALL ARMYWORM (LEPIDOPTERA: NOCTUIDAE) PARASITOIDS IN THE MEXICAN STATES OF MICHOACÁN, COLIMA, JALISCO, AND TAMAULIPAS

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### ABSTRACT

Fall armyworm larvae, *Spodoptera frugiperda* (J. E. Smith) were collected from whorl stage corn or sorghum in the states of Michoacán, Colima, and Jalisco in August, and Tamaulipas, Mexico in September 1998. Eleven species of hymenopteran parasitoids were recovered representing 3 families: Ichneumonidae (*Ophion flavidus* Brulle, *Campoletis flavicincta* Ashmead, and *Pristomerus spinator* F.); Braconidae (*Aleiodes laphygmae* Viereck, *Cotesia marginiventris* Cresson, *Meteorus laphygmae* Viereck, *Meteorus* sp., *Chelonus insularis* Cresson, *Chelonus* sp. probably *cautus* Cresson, and *Chelonus* sp.); and Eulophidae (*Euplectrus platyhyphenae* Howard). The overall rate of parasitism was 11.3%, based on 2219 larvae collected. The highest rate of parasitism from a single collection was 26.5%, representing 6 species of parasitoids in Michoacán. The next highest rate of parasitism, 23%, was by a single species, *C. flavicincta*, in Michoacán. The most widely distributed species was *P. spinator*, occurring in 12 collections from 3 states. *Chelonus* sp. was collected from all four states in only 6 collections. The greater diversity of parasitoids and higher rates of parasitism in Michoacán may be related to the more diverse habitat with more forests, orchards, and pastures near the cornfields in that state.

Key Words: parasitoids, *Spodoptera frugiperda*, *Ophion*, *Campoletis*, *Pristomerus*, *Aleiodes*, *Cotesia*, *Meteorus*, *Chelonus*, *Euplectrus*, maize, corn, sorghum, Mexico

### RESUMEN

Larvas de gusano cogollero, *Spodoptera frugiperda* (J. E. Smith) fueron colectadas de maíz y sorgo para grano en etapa de verticilio en los estados de Michoacán, Colima y Jalisco durante Agosto, y en Tamaulipas, México durante Septiembre de 1998. Once especies de parasitoides himenópteros se recuperaron y representaron a 3 familias: Ichneumonidae (*Ophion flavidus* Brulle, *Campoletis flavicincta* Ashmead, y *Pristomerus spinator* F.); Braconidae (*Aleiodes laphygmae* Viereck, *Meteorus* sp., *Chelonus insularis* Cresson, *Chelonus* sp. probablemente *cautus* Cresson, y *Chelonus* sp.); y Eulophidae (*Euplectrus platyhyphenae* Howard). La proporción general de parasitismo fue de 11.3%, basada en 2219 larvas colectadas. La proporción más alta de parasitismo proveniente de una colecta simple fue de 26.5%, representando 6 especies de parasitoides en Michoacán. La siguiente proporción más alta, 23%, fue para una especie simple, *C. flavicincta*, en Michoacán. La especie distribuida más ampliamente fue *P. spinator*, presentándose en 12 colectas hechas en 3 estados. *Chelonus* sp. se colectó en los cuatro estados sólo en 6 colectas. La diversidad más grande de parasitoides y proporciones más altas de parasitismo en Michoacán pueden estar relacionadas con los hábitat más diversos con más bosques, huertas y pastizales cerca de los maizales en ese estado.

The fall armyworm, *Spodoptera frugiperda* (J. E. Smith), is an important economic pest of corn, sorghum, grasses, and occasionally other crops in North America, Central America and parts of South America (Luginbill 1928; Vickery 1929; Mitchell 1979; Andrews 1980, 1988). The fall armyworm lacks the ability to diapause during cold weather and thus spreads northward from over-

wintering habitats each year (Luginbill 1928). The seasonal migration of fall armyworm from Southern Florida and the Lower Rio Grande Valley has been studied by Westbrook and Sparks (1986), Pair et al. (1986, 1991), and Pair & Westbrook (1995). The potential for migration from other areas along the Mexican Gulf Coast was discussed by Raulston et al. (1986).

Early workers recognized the value of parasitoids in reducing larval populations of fall armyworm (Luginbill 1928; Vickery 1929). Andrews (1988) reviewed the Latin American research on fall armyworm including its parasitoids, and Ashley et al. (1989) reviewed the literature on fall armyworm. Biological control of this pest is desirable because of increasing economic and environmental concerns which have resulted in surveys of parasitoids and other natural enemies in different parts of its range (Hogg et al. 1982; Pair et al. 1986; Gross & Pair 1986; Castro & Pitre 1989). Ashley (1979) listed 53 species of fall armyworm parasitoids from 43 genera and 10 families and suggested that importations from Central and South America into Florida and Texas may significantly reduce overwintering populations. He also suggested that the number of parasitoids unique to either North or South America is indicative of the need for more larval collections so as to establish whether differences actually are present or are simply a function of inadequate records. Ashley (1986) reported the highest parasitism levels of fall armyworm were found for corn and that *Chelonus insularis* Cresson had the highest parasitism rates of all the parasitoids for North and Central America. Pair et al. (1986) found that the highest rates of parasitism were in overwintering areas of Mexico-Texas and south Florida and confirmed that *C. insularis* was the most common parasitoid. They presented evidence for differential distribution of some parasitoids as indicated by their native scarcity or abundance in defined

geographical areas. Riggin et al. (1993) concluded that the most efficient biological control programs for fall armyworm will be ones that use and amplify several parasitoid species rather than programs that rely on an individual natural enemy.

Biological differences (developmental rates, reproductive compatibility and susceptibility to insecticides) between populations of fall armyworm collected from corn in different areas of Mexico suggested some geographical isolation of populations (Lopez-Edwards et al. 1999). Therefore, we surveyed the natural enemies of fall armyworm in west-central and northeastern Mexico in an effort to find new parasitoids and to add to the knowledge of the distribution of known parasitoids of this pest.

#### MATERIALS AND METHODS

During August 1998, collections of fall armyworm larvae were made from whorl-stage corn and sorghum in the states of Michoacán, Colima, and Jalisco, and 1 collection was made from fruiting corn in Jalisco. A later collection was made from whorl-stage sorghum in Tamaulipas in September. No efforts were made to collect eggs or pupae. The larvae were placed individually in 30 cc plastic cups with pinto bean diet (Burton 1969) and held in the laboratory for emergence of parasitoids. Adult parasitoids were submitted to the USDA Systematic Entomology Laboratory, Beltsville, MD for identification. The dates and locations of collections are presented in Table 1.

TABLE 1. LOCATION, DATE, CROP, SAMPLE SIZE, AND TOTAL PERCENTAGE FALL ARMYWORM LARVAE PARASITIZED BY HYMENOPTERA, COLLECTED FROM CORN (C) OR SORGHUM (S) IN THE MEXICAN STATES OF MICHOACÁN (M), COLIMA (C), JALISCO (J), AND TAMAULIPAS (T).

| Code | Location           | Date     | Crop     | No. coll. | Percentage parasitized |
|------|--------------------|----------|----------|-----------|------------------------|
| M 1  | Jazmin             | 08/07/98 | C        | 25        | 4.0                    |
| M 2  | El Batillero       | 08/07/98 | C        | 26        | 23.0                   |
| M 3  | La Sidra           | 08/07/98 | C        | 84        | 6.0                    |
| M 4  | La Sidra           | 08/08/98 | C        | 89        | 9.0                    |
| M 5  | El Hueso           | 08/08/98 | C        | 102       | 26.5                   |
| M 6  | Carreras           | 08/08/98 | C        | 109       | 14.7                   |
| C 7  | Mezcales           | 08/12/98 | C        | 143       | 7.0                    |
| C 8  | Los Clomos         | 08/12/98 | C        | 84        | 2.4                    |
| C 9  | Cerro Colorado     | 08/13/98 | C        | 121       | 3.3                    |
| C 10 | El Bordo           | 08/13/98 | C        | 114       | 11.4                   |
| C 11 | Crucero de Tapames | 08/13/98 | C        | 89        | 0                      |
| C 12 | Peña Blanca        | 08/13/98 | C & S    | 219       | 15.5                   |
| C 13 | El Narajito        | 08/14/98 | C        | 171       | 7.6                    |
| J 14 | Los Pozos          | 08/19/98 | C        | 81        | 8.6                    |
| J 15 | Apastepe           | 08/19/98 | C        | 89        | 7.9                    |
| J 16 | Los Depositos      | 08/19/98 | C        | 89        | 4.5                    |
| J 17 | Sayula             | 08/19/98 | C        | 89        | 2.2                    |
| J 18 | Sayula             | 08/20/98 | C & S    | 177       | 18.6                   |
| J 19 | Sayula             | 08/20/98 | C (ears) | 18        | 0                      |
| T 20 | El Mante           | 09/22/98 | C        | 300       | 19.7                   |



Collection size ranged from 25 to 300 fall armyworm larvae. Collections 12 and 18 comprise combinations of samples from adjacent fields of whorl-stage corn and sorghum. The number collected was corrected by subtracting the number that died from injury or unknown causes during the first few days after collection before calculating percent parasitism. Mortality due to pathogens and nematodes will be reported elsewhere.

#### RESULTS AND DISCUSSION

Out of 2219 fall armyworm larvae collected, 251 produced parasitoids, for a parasitism rate of 11.3%. This represented 11 species from 3 families of Hymenoptera: 7 Braconidae, 3 Ichneumonidae, and 1 Eulophidae. Only 2 of 20 collections produced no parasitoids: a collection from whorl-stage corn at C11 in Colima and a collection from ears of corn at J19 in Jalisco. The two highest rates of parasitism were found in Michoacán at M5 and M2 with 26.5 and 23% parasitism, respectively (Table 1). M5, C12, and C13, had the highest number of parasitoid species, 6 (Tables 2 and 3). In contrast, M2 had only a single species of parasitoid and represented the highest rate of parasitism by a particular species, 23%. The next most diverse collections of parasitoids were from M3 and M6 in Michoacán with 5 species each (Tables 2 and 3).

*Pristomerus spinator* F. was the most widely distributed parasitoid, being found in 12 collections from 3 states (Table 3). At nearly 13%, it showed the second highest rate of parasitism. *P. spinator* was listed as a parasite of fall armyworm from Nicaragua, Brazil, and Cuba by Andrews (1988). *P. spinator* was reported from Quintana Roo and Tamaulipas, México, by Carrillo (1980) and Pair et al. (1986) respectively. While this was the most widely distributed parasitoid in our collections, we did not find it in the single collection from Tamaulipas.

*Campoletis flavicincta* Ashmead was found in 8 collections from 2 states, occurring most abundantly in Michoacán in 5 of 6 collections and also had the highest rate of parasitism, 23% (Table 3). However, it was not found in collections from Colima and Tamaulipas. *C. flavicincta* was listed from several states of the U.S.A. and from Uruguay by Ashley (1979), and from Nicaragua, Uruguay, and Brazil by Andrews (1988).

*Ophion flavidus* Brulle was found in one collection in Michoacán and two collections from Colima, with the highest rate of parasitism (5%) in Colima. *O. flavidus* was listed from the U.S.A. by Ashley (1986) and from Honduras and Brazil by Andrews (1988).

*Chelonus insularis* was found in 9 collections from 3 states but always at less than 5%. Luginbill (1928) and Vickery (1929) indicated that *C. in-*

TABLE 2. PERCENTAGE OF FALL ARMYWORM LARVAE PARASITIZED BY EACH SPECIES OF BRACONIDAE AT EACH LOCATION.

| Code* | Braconidae   |              |              |               |              |              |               |
|-------|--------------|--------------|--------------|---------------|--------------|--------------|---------------|
|       | <i>A. l.</i> | <i>C. i.</i> | <i>C. c.</i> | <i>C. sp.</i> | <i>C. m.</i> | <i>M. l.</i> | <i>M. sp.</i> |
| M 1   | 0            | 0            | 0            | 0             | 0            | 0            | 0             |
| M 2   | 0            | 0            | 0            | 0             | 0            | 0            | 0             |
| M 3   | 0            | 0            | 0            | 0             | 0            | 0            | 1.2           |
| M 4   | 0            | 0            | 0            | 2.2           | 0            | 0            | 3.4           |
| M 5   | 0            | 1.0          | 1.0          | 0             | 2.0          | 0            | 2.9           |
| M 6   | 0            | 0.9          | 2.8          | 0             | 0            | 0            | 0             |
| C 7   | 0            | 2.1          | 3.5          | 0             | 0            | 0.7          | 0             |
| C 8   | 0            | 0            | 0            | 1.2           | 1.2          | 0            | 0             |
| C 9   | 0            | 0.8          | 0            | 0             | 0            | 0            | 0             |
| C 10  | 0            | 0.9          | 3.5          | 0             | 0.9          | 0            | 0             |
| C 11  | 0            | 0            | 0            | 0             | 0            | 0            | 0             |
| C 12  | 0            | 0            | 10.6         | 2.9           | 0            | 1.9          | 1.0           |
| C 13  | 0            | 2.3          | 0.6          | 1.2           | 0            | 0.6          | 0             |
| J 14  | 0            | 3.7          | 0            | 0             | 0            | 0            | 0             |
| J 15  | 0            | 0            | 0            | 0             | 0            | 0            | 6.7           |
| J 16  | 0            | 1.1          | 0            | 0             | 0            | 0            | 3.4           |
| J 17  | 0            | 2.2          | 0            | 0             | 0            | 0            | 0             |
| J 18  | 0            | 1.1          | 0            | 9.6           | 0            | 0            | 0             |
| J 19  | 0            | 0            | 0            | 0             | 0            | 0            | 0             |
| T 20  | 0.3          | 0            | 0            | 0.7           | 0            | 10.3         | 0             |

\*Locations are described in Table 1. *A. l.* = *Aleoides laphygmae*, *C. i.* = *Chelonus insularis*, *C. c.* = *Chelonus* sp. prob. *cautus*, *C. sp.* = *Chelonus* sp., *C. m.* = *Cotesia marginiventris*, *M. l.* = *Meteorus laphygmae*.

TABLE 3. PERCENTAGE OF FALL ARMYWORM LARVAE PARASITIZED BY EACH SPECIES OF ICHNEUMONIDAE AND EULOPHIDAE AT EACH LOCATION.

| Code* | Ichneumonidae |              |              | Eulophidae   |
|-------|---------------|--------------|--------------|--------------|
|       | <i>C. f.</i>  | <i>O. f.</i> | <i>P. s.</i> | <i>E. p.</i> |
| M 1   | 0             | 0            | 4.0          | 0            |
| M 2   | 23.1          | 0            | 0            | 0            |
| M 3   | 1.2           | 1.2          | 1.2          | 1.2          |
| M 4   | 1.1           | 0            | 2.2          | 0            |
| M 5   | 6.9           | 0            | 12.7         | 0            |
| M 6   | 1.8           | 0            | 8.2          | 0.9          |
| C 7   | 0             | 0            | 0.7          | 0            |
| C 8   | 0             | 0            | 0            | 0            |
| C 9   | 0             | 0            | 1.7          | 0            |
| C 10  | 0             | 0            | 6.1          | 0            |
| C 11  | 0             | 0            | 0            | 0            |
| C 12  | 0             | 4.8          | 8.7          | 0            |
| C 13  | 0             | 0.6          | 2.3          | 0            |
| J 14  | 1.2           | 0            | 3.7          | 0            |
| J 15  | 1.1           | 0            | 0            | 0            |
| J 16  | 0             | 0            | 0            | 0            |
| J 17  | 0             | 0            | 0            | 0            |
| J 18  | 2.3           | 0            | 4.5          | 0            |
| J 19  | 0             | 0            | 0            | 0            |
| T 20  | 0             | 0            | 0            | 8.3          |

\*Locations are described in Table 1. *C. f.* = *Camponotus flavicincta*, *O. f.* = *Ophion flavidus*, *P. s.* = *Pristomerus spinator*, *E. p.* = *Euplectrus platyphenae*.

*sularis* was important in controlling fall armyworm populations in its overwintering habitats of Florida and Southern Texas. Pair et al. (1986) confirmed the importance of *C. insularis* in these areas but found that it was of secondary importance elsewhere. *C. insularis* has been reported as an important parasite of fall armyworm in Latin America (Andrews 1988). Ashley (1986) reported that the Braconidae had the greatest impact on fall armyworm populations with *C. insularis* having the highest parasitism rates in Central and North America.

*Chelonus* sp. was found in 6 collections, representing all 4 states, and *Chelonus* sp. probably *cautus* was found in 6 collections, representing only 2 states (Table 2). The taxonomy of the genus *Chelonus* needs more study in Mexico (P. M. Marsh, pers. comm.). In our collections, *C. insularis* was most common in Colima and was not found in the single collection from Tamaulipas. When all *Chelonus* sp. are considered, they were found in 14 locations, representing all 4 states. While most of the collections had less than 5% parasitism by any member of the genus *Chelonus*, one collection in Colima had 11% parasitism by *Chelonus* sp. probably *cautus* and one collection from Jalisco had 10% parasitism by *Chelonus* sp. Thus, *Chelonus* spp. appear to be highly important as natural enemies of fall armyworm in Mexico.

*Meteorus laphygmae* Viereck was found in 4 collections, representing 2 states. It occurred at

only 1 to 2% in the three collections in Colima, but was the most abundant in Tamaulipas with 10%. *Meteorus* sp. was found in 6 collections, representing 3 states, with its highest rate of 7% in Jalisco. Ashley (1986) listed the genus *Meteorus* only from the continental U.S.A., stating that *M. laphygmae* in Texas had its greatest impact on fall armyworm feeding on grass. Andrews (1988) lists *M. laphygmae* from Surinam, Venezuela, and Colombia. Pair et al. (1986) list *Meteorus autographae* Musebeck from Mexico as well as several southern states of the U.S.A.

*Aleiodes laphygmae* Viereck (formerly *Rogas laphygmae*) was the only parasitoid limited to a single collection, in Tamaulipas. Ashley (1986) reported that *R. laphygmae* appeared to be confined to the continental U. S. and that the highest parasitism rates occurred in fall armyworm feeding on grass. Andrews (1988) listed *R. laphygmae* from Nicaragua.

*Cotesia marginiventris* Cresson (formerly *Apanteles marginiventris*) was found in 4 collections, representing only 2 states at rates of 2% or lower. *C. marginiventris* has often been reported as a parasitoid of fall armyworm in the U.S.A. (Ashley 1986). Andrews (1988) listed *A. marginiventris* from Lesser Antilles, Surinam, Venezuela, Brazil, and Nicaragua. Ashley (1986) reported that *C. marginiventris* appeared to have its greatest impact on fall armyworm feeding on grass. However, under experimental conditions of whorl-stage corn

infested with newly hatched fall armyworm, *C. marginiventris* can produce rates of parasitism up to 40% in Georgia (Hamm et al. 1994).

*Euplectrus platyhyphenae* Howard was the only member of the family Eulophidae collected. *E. platyhyphenae* was most abundant in Tamaulipas (8%), but occurred at very low levels in 2 collections from Michoacán. Ashley (1986) lists *E. platyhyphenae* from Lesser Antilles, Cuba, Barbados, Trinidad, Venezuela, and Colombia. Montoya-Burgos (1980) reported natural parasitism by *Euplectrus* sp. against second instar fall armyworm of about 15% in corn from Veracruz.

Due to technological advances in mass rearing, Lewis and Nordlund (1980) suggested *C. insularis* and *C. marginiventris* as candidates for "rear and release" approaches using either: (1) release throughout the overwintering zone, (2) early-season colonization, or (3) therapeutic release on target crops.

We did not sample eggs or pupae and therefore did not find any egg (except for *Chelonus* which is an egg-larval parasitoid) or pupal parasitoids. The rare incidence of tachinid parasitoids was probably due to the low incidence of large host larvae in our collections (Rohlf's & Mack 1985). *Archytas marmoratus* (Townsend) is an important parasitoid of both fall armyworm and corn earworm, *Helicoverpa zea* (Boddie), in whorl-stage corn in the U.S.A (Gross & Pair 1991). Pair et al. (1986) reported that *A. marmoratus* was the primary parasitoid attacking medium and large fall armyworm larvae in whorl-stage corn throughout the southern states during the spring of 1981-83. *Archytas* spp. have been reported attacking *S. frugiperda* in several areas in Latin America (Andrews 1988). The low incidence of *O. flavidus* may also have been influenced by the collection of mostly small larvae (Rohlf's & Mack 1985). Gross & Pair (1991) state that *O. flavidus* parasitized 4th, 5th, and 6th instar armyworm with equal success, but were minimally successful in completing development on late 6th instars. Although *O. flavidus* does not kill the host until the late prepupal or pupal stage, Rohlf's and Mack (1983) found that parasitized larvae consumed 17 to 22% less artificial diet than unparasitized larvae.

Results of this survey suggest a need for more taxonomic studies of parasitic hymenoptera in Mexico, especially for *Meteorus* sp., *Chelonus* sp., and *Chelonus* sp. prob. *cautus*. All of the determined species, except *Pristomerus spinator*, have ranges which extend into the U.S.A. However, they may not be evenly distributed throughout Mexico. This study was only a partial survey of these areas of Mexico within a defined time frame, during the rainy season when most corn is grown. A thorough survey would require sampling all developmental stages of fall armyworm to evaluate the importance of parasitoids that de-

velop in the egg, pupal, and adult stages throughout the growing season for corn and sorghum over several years to determine if the differences seen in this study were due to location, the developmental stage of the host crop, or the developmental stage of the fall armyworm larvae. Additional ecological studies are needed to determine where and how the fall armyworm and its various natural enemies survive the dry season when few crops are available.

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## EVALUATION OF YIELDGARD TRANSGENIC RESISTANCE FOR CONTROL OF FALL ARMYWORM AND CORN EARWORM (LEPIDOPTERA: NOCTUIDAE) ON CORN

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### ABSTRACT

Fall armyworm, *Spodoptera frugiperda* (J. E. Smith), and corn earworm, *Helicoverpa zea* Boddie, perennially cause leaf and ear damage to corn in the southeastern USA. Development of transgenic hybrids expressing insecticidal endotoxin from *Bacillus thuringiensis* (Bt) offers a new approach to managing these insects in field corn. Transgenic Bt hybrids with either the Bt11 or MON810 event, collectively known as YieldGard Technology, were evaluated for control fall armyworm and corn earworm in southern Georgia during 1998, which coincided with a severe outbreak of fall armyworm. YieldGard Bt resistance consistently reduced whorl infestation and damage to low levels and also reduced ear infestations and larval numbers per ear. However, larval establishment did occur on many ears of resistant plants, but once established in ears, larvae of both species developed more slowly and caused much less kernel damage on resistant than susceptible plants. We found no relationship between YieldGard Bt resistance and corn grain aflatoxin concentrations. Yield responses were variable with the prevention of yield loss being proportional to the severity of insect damage. These results indicate that YieldGard resistance is effective in preventing significant losses to field corn by fall armyworm and corn earworm. Further, evaluation under a variety of growing conditions and insect infestation levels is needed to clearly assess the value of YieldGard technology to corn growers in the Southeast.

Key Words: Plant resistance, *Spodoptera frugiperda*, *Helicoverpa zea*, Transgenic crops, Bt resistance

### RESUMEN

*Spodoptera frugiperda* (J.E. Smith), y *Helicoverpa zea* Boddie, perennemente causan daño de hoja y mazorca al maíz en el sureste de los Estados Unidos de América. El desarrollo de híbridos transgénicos que expresan la endotoxina insecticida de *Bacillus thuringiensis* (Bt) ofrece una nueva practica para controlar estos insectos en campos de maíz. Híbridos transgénicos Bt, ya sea con evento Bt11 o MON810, colectivamente conocidos como tecnología YieldGard, fueron evaluados para control de *S. frugiperda* y *H. zea* en el sur de Georgia durante el 1998, lo cual coincidió con una epidemia severa de *S. frugiperda*. Resistencia YieldGard Bt consistentemente redujo infestación de cogollo y daños a niveles bajos y también redujo infestación de mazorca y el numero de larvas por mazorca. Sin embargo, establecimiento larval si ocurrió en numerosas mazorcas de plantas resistentes, pero una vez establecidas el la mazorca, larvas de ambas especies se desarrollaron mas lentamente y causaron mucho menos daño de grano en plantas resistentes que en las susceptibles. No encontramos ninguna relación entre resistencia YieldGard Bt y concentraciones de aflatoxina en granos de maíz. Producción de cosechas fueron variables con la prevención de perdida de producción siendo proporcional a la severidad del daño por insecto. Estos resultados indican que resistencia YieldGard es efectiva en prevenir perdidas significativas de maíz de campo por *S. frugiperda* y *H. zea*.

Fall armyworm, *Spodoptera frugiperda* (J. E. Smith), and corn earworm, *Helicoverpa zea* Boddie, are the most important insect pests of corn in the southeastern U.S. Both insects infest whorl stage plants causing leaf injury, but more impor-

tantly they infest ears causing direct loss of grain. Insecticidal control is difficult and generally not cost effective in field corn. Typically, early planting times are recommended in the Southeast partly to avoid damaging levels of both insects

which often occur later in the season. Germplasm with moderate levels of leaf feeding resistance to fall armyworm has been released (Williams et al. 1997, 1998). High levels of resistance to fall armyworm and corn earworm in silks of the tropical corn 'Zapalote Chico' also have been identified (Wiseman & Windstrom 1986). However, these natural sources of plant resistance have not been effectively deployed.

Transgenic corn hybrids expressing the insecticidal protein Cry1Ab from *Bacillus thuringiensis* (Bt) var. *kurstaki* were originally developed to control European corn borer, *Ostrinia nubilalis* (Hübner) and offer the potential for reducing losses by fall armyworm and corn earworm. Several events of transgenic Bt corn have been developed with different modes of toxin expression (Ostlie et al. 1997). The most promising events are Bt11 (Novartis Seeds) and MON810 (Monsanto Co.) where endotoxin is expressed in vegetative and reproductive structures throughout the season (Armstrong et al. 1995; Williams et al. 1997). Hybrids containing either of these events are collectively referred to as having 'YieldGard Technology' (Ostlie et al. 1997). Laboratory feeding trials and small controlled field trials have shown that hybrids containing the Bt11 event reduce fall armyworm and corn earworm growth and survival (Williams et al. 1997, 1998). YieldGard resistance also is very effective against Southwestern corn borer (Archer et al. 2000; Williams et al. 1998), but this insect either does not occur or is not economically important in the coastal plain region of the southeastern U.S.

Because of concerns about the potential for corn earworm to develop virulence to Bt technology in transgenic cotton, YieldGard transgenic corn was not commercially deployed in the Southeast until 1998. In a series of studies at five locations in Alabama in 1998 with corn planted at the recommended time and 1 and 2 months later, DeLamar et al. (1998a-e) demonstrated that YieldGard resistance (events MON810 and Bt11) prevented whorl damage, kernel damage, and yield loss by lepidopterans, primarily fall armyworm and corn earworm, in later plantings at all locations. YieldGard resistance generally did not improve the performance of corn planted at recommended times, because these plantings generally escaped severe lepidopteran damage. YieldGard resistance has not been evaluated in the field under natural infestations of fall armyworm or corn earworm in Georgia. Furthermore, lepidopteran infestations of ears have been linked with increased levels of fungal infection and contamination of grain by mycotoxins such as aflatoxin produced by *Aspergillus flavus* (e.g., Windstrom 1979; McMillian 1983; McMillian et al. 1985; Smith & Riley 1992). Effective reduction in lepidopteran ear infestations with transgenic Bt resistance may also help reduce aflatoxin contamination of grain (Williams et al. 1998).

Our objective was to evaluate the effect of both YieldGard Technology events on fall armyworm and corn earworm infestations and damage and on grain aflatoxin contamination of field corn. Trials were conducted in southern Georgia during the summer of 1998 and coincided with a severe outbreak of fall armyworm.

#### MATERIALS AND METHODS

Trials were conducted on a Greenville sandy loam soil at the Univ. of Georgia Southwest Branch Experiment Station near Plains, and on a Tifton sandy loam soil at the Attapulgus Research Center near Attapulgus and the Coastal Plain Experiment Station near Tifton. The study area at each location was fertilized, chisel plowed or subsoiled twice, and disk harrowed. Before disking 440 kg/ha of 3-18-9 (N-P-K) granular fertilizer was applied and an additional 112 kg of nitrogen was applied as ammonium nitrate about 20 d after planting. Seed at all locations was planted with an air-planter at the rate of 66,700 plants per ha. Pendimethalin (Prowl) at 0.71 L/ha and atrazine (Aatrex) at 0.57 L/ha were applied to control weeds. No other pesticides were applied. Natural rainfall was supplemented by irrigating weekly with 6 cm/ha of water as needed at all locations.

The experimental design within each planting date and location was a split plot design with whole plots being brand (manufacturer) and split plots being hybrid pairs within manufacturer. At Attapulgus a single planting occurred on 23 April 1998. Planting dates were conducted as separate side-by-side trials with two dates at Tifton (13 and 23 April 1998) and three dates at Plains (14 April, 12 May and 3 June 1998). Hybrid pairs were a Bt hybrid and a non-Bt isoline or near isoline hybrid. Susceptible and Bt-resistant hybrid pairs at Plains were Pioneer Brand 3223 and 31B13 (Bt), Golden Harvest (Monsanto) 2530 and 2530Bt, Dekalb DK 591 and DK 591BtY, and Novartis N79-P4 and N79L3 (Bt). Pairs at Tifton were Pioneer Brand 3223 and 31B13 (Bt), Pioneer Brand 3394 and 33V08 (Bt), and Novartis N79-P4 and N79L3 (Bt). Pairs at Attapulgus were Pioneer Brand 3223 and 31B13 (Bt), Dekalb DK 591 and DK 591BtY, and Novartis N79-P4 and N79L3 (Bt). Whole plots were arranged in a randomized complete block design with four replications at Attapulgus and the first and second planting dates at Plains and 3 replications at both plantings at Tifton and the third date at Plains. Plots measured 15.2 m by 6 rows (76-cm rows) at Plains, 21.3 m by 4 rows (91-cm rows) at Attapulgus and 9.1 m by 4 rows (91-cm rows) at Tifton.

Whorl defoliation was assessed by rating 30 plants (all plants at Tifton) in the two center rows per plot about 6 wk after planting at the 8-10 leaf stage for each planting date. Plants were rated for damage using a 0-9 scale (Davis et al. 1992)

where 0 is no damage and 9 is whorl and furl almost completely defoliated. The damage scale is not linear with ratings of  $\geq 4$  indicating substantially more damage than ratings of  $\leq 3$ . Twenty to 30 larvae were collected for species identification from infested whorls in rows at the edge of plots. Ear damage of 12 ears per plot was assessed by counting the number of live larvae and larval feeding cavities and measuring the total length of all feeding cavities for each ear (Windstrom 1967). Ear infestations were sampled twice at Attapulugus with live larvae in the first sample being identified to species and categorized as small, medium or large and final ear damage being assessed at the second sample.

The two center rows of each plot were harvested with a Hege two-row corn combine on August 12, September 1, and September 12 for the three respective planting dates at Plains and 20 August at Tifton. At Attapulugus, plots (all 4 rows) were harvested on 10 August using a John Deere 4420 combine with a 4-row corn head modified for small plot harvesting. Grain yields were adjusted to 15.5% moisture content. A 2-kg subsample of grain was collected from each plot in trials at Plains and Attapulugus for determination of grain aflatoxin levels. Kernels were ground to pass a 20 mesh screen, well mixed, and 100 g subsample were extracted. Aflatoxin contamination was determined using the Vicam immunoaffinity column method (Truckness et al. 1991) and is reported as total aflatoxin ( $B_1 + B_2 + G_1 + G_2$ ) in parts per billion of seed.

Results were analyzed within each planting date and location with an analysis of variance for a split plot design. Before analysis, percentage data were transformed by square-root arcsine, and numeric data were transformed by  $\log_{10}(x + 1)$ . Significance of main effects for brand (manufacturer) and hybrid resistance (i.e., Bt versus non-Bt) were determined using *F* test at  $P = 0.05$ . Brand  $\times$  hybrid-resistance interactions were not significant ( $P = 0.05$ ) for any parameter. Therefore, only hybrid resistance main effects (i.e., average across all brands) are presented for the combined analyses.

## RESULTS

### Species Composition

Fall armyworm populations reached damaging outbreak levels earlier than normal in 1998 resulting in the worst damage to corn in Georgia in the last decade. Whorl infestations in all trials consisted almost entirely of fall armyworm. Ear infestations at Attapulugus ( $N = 430$  larvae) were 89% fall armyworm and 11% corn earworm. At Tifton, ear infestations were 90% fall armyworm and 10% corn earworm in both plantings ( $N = 178$ ). At Plains, fall armyworms accounted for 11%, 48% and 73% of total live larvae observed in

ears of all hybrids in the first ( $N = 75$ ), second ( $N = 131$ ), and third planting ( $N = 33$ ) dates, respectively, with the balance being corn earworm.

### Whorl Infestations and Damage

Hybrid brand did not significantly ( $P = 0.05$ ) affect the percentage of infested whorls or mean damage rating per plant and per infested plant in any trial. Whorl infestations and damage increased substantially from the first to third plantings at Plains, but were similar between plantings at Tifton (Table 1). YieldGard Bt resistance greatly reduced whorl infestations, whorl damage ratings per plant and whorl damage rating per infested plant at Attapulugus and in all plantings at Plains (Table 1). Whorl infestations and whorl damage ratings also were smaller in resistant than susceptible hybrids in both plantings at Tifton. However whorl infestations were much lower at Tifton than at the other locations, and differences were not significant in either planting.

### Ear Infestations and Damage

Hybrid brand main effects were not significant ( $P = 0.05$ ) in any trial for the percentage of infested ears or mean damage rating per ear and per infested ear. Ear infestations in susceptible hybrids were uniformly high in all trials, but ear damage became progressively more severe in later plantings at Tifton and Plains (Table 2). The percentage of infested ears was reduced in resistant hybrid in all plantings at Tifton and Plains, although 30% to 70% of ears of resistant hybrids were infested. However, at Attapulugus all ears of susceptible hybrids and almost every ear of resistant hybrids were infested. YieldGard resistance significantly reduced the number of larval cavities which is a direct measure of the number of larvae per ear in all trials (Table 2). Furthermore, resistance also greatly reduced the amount of damage per ear and per infested ear in all trials, with the exception of the damage per infested ear in the first planting at Tifton.

Many larvae were present in ears during the first ear sample at Attapulugus. The size distribution of larvae reveals that the majority of fall armyworms and corn earworms were medium sized (i.e., instars 3 and 4) in ears of Bt resistant plants, but most larvae were large sized (i.e., instars 5 and 6) in ears of susceptible plants (Fig. 1).

### Grain Yield and Aflatoxin Levels

Grain yield and aflatoxin level were not significantly different between brands in any trial, except at Attapulugus where both Pioneer brand hybrids yielded more than the other hybrids. This difference in yield presumably is due to differences in agronomic characteristics and not to differences in insect resistance.

TABLE 1. MEAN ( $\pm$ SE) WHORL INFESTATION AND WHORL DAMAGE RATING CAUSED BY FALL ARMYWORM IN SUSCEPTIBLE AND 'YIELDGARD' RESISTANT CORN IN GEORGIA, 1998.

| Location    | Planting date | Bt resistance | Infested whorls (%) | Damage rating <sup>1</sup> per plant | Damage rating <sup>1</sup> per infested plant |
|-------------|---------------|---------------|---------------------|--------------------------------------|---|
| Attapulugus | 23 March      | -             | 83.3 $\pm$ 2.2      | 4.0 $\pm$ 0.3                        | 4.9 $\pm$ 0.2                                 |
|             |               | +             | 13.8 $\pm$ 1.8      | 0.5 $\pm$ 0.1                        | 3.2 $\pm$ 0.4                                 |
|             |               | F             | 675.83***           | 303.82***                            | 16.81**                                       |
| Tifton      | 13 April      | -             | 12.3 $\pm$ 2.2      | 0.23 $\pm$ 0.06                      | 1.77 $\pm$ 0.13                               |
|             |               | +             | 6.9 $\pm$ 1.5       | 0.10 $\pm$ 0.02                      | 1.42 $\pm$ 0.11                               |
|             |               | F             | 2.03 ns             | 2.14 ns                              | 2.09 ns                                       |
|             | 23 April      | -             | 8.8 $\pm$ 0.4       | 0.14 $\pm$ 0.03                      | 1.75 $\pm$ 0.49                               |
|             |               | +             | 4.4 $\pm$ 1.9       | 0.05 $\pm$ 0.02                      | 1.16 $\pm$ 0.06                               |
|             |               | F             | 2.33 ns             | 2.42 ns                              | 1.19 ns                                       |
| Plains      | 14 April      | -             | 23.3 $\pm$ 3.2      | 0.5 $\pm$ 0.1                        | 2.1 $\pm$ 0.2                                 |
|             |               | +             | 9.2 $\pm$ 1.8       | 0.1 $\pm$ 0.1                        | 1.0 $\pm$ 0.2                                 |
|             |               | F             | 19.64**             | 24.56**                              | 19.75**                                       |
|             | 12 May        | -             | 49.4 $\pm$ 5.4      | 2.5 $\pm$ 0.3                        | 5.1 $\pm$ 0.2                                 |
|             |               | +             | 14.1 $\pm$ 3.6      | 0.5 $\pm$ 0.2                        | 3.8 $\pm$ 0.3                                 |
|             |               | F             | 42.79**             | 37.22***                             | 18.35**                                       |
|             | 3 June        | -             | 96.1 $\pm$ 1.4      | 5.4 $\pm$ 0.2                        | 5.6 $\pm$ 0.1                                 |
|             |               | +             | 35.0 $\pm$ 2.6      | 1.1 $\pm$ 0.1                        | 3.4 $\pm$ 0.1                                 |
|             |               | F             | 283.41***           | 1068.67***                           | 45.08***                                      |

ns, \*\*, \*\*\* indicate not significant and significant at  $P = 0.01$  and  $P = 0.001$ , respectively.

<sup>1</sup>Rating scale of Davis et al. (1992) where 0 is no damage and 9 is whorl and furl destroyed.

TABLE 2. MEAN ( $\pm$ SE) EAR INFESTATION, CAVITY NUMBER AND LENGTH CAUSED BY FALL ARMYWORM AND CORN EARWORM IN SUSCEPTIBLE AND 'YIELDGARD' RESISTANT CORN IN GEORGIA, 1998.

| Location    | Planting date | Bt resistance | Infested ears (%) | Larval cavities per ear | Damage rating <sup>1</sup> per ear | Damage rating <sup>1</sup> per infested ear |
|-------------|---------------|---------------|-------------------|-------------------------|------------------------------------|---|
| Attapulugus | 23 March      | -             | 100.0 $\pm$ 0     | 2.4 $\pm$ 0.1           | 8.9 $\pm$ 0.6                      | 8.9 $\pm$ 0.6                               |
|             |               | +             | 96.5 $\pm$ 1.6    | 1.1 $\pm$ 0.1           | 2.7 $\pm$ 0.2                      | 2.8 $\pm$ 0.2                               |
|             |               | F             | 4.48 ns           | 90.89***                | 135.79***                          | 131.21***                                   |
| Tifton      | 13 April      | -             | 81.5 $\pm$ 4.0    | 0.9 $\pm$ 0.1           | 2.3 $\pm$ 0.2                      | 2.7 $\pm$ 0.2                               |
|             |               | +             | 28.7 $\pm$ 7.9    | 0.3 $\pm$ 0.1           | 0.7 $\pm$ 0.2                      | 2.0 $\pm$ 0.3                               |
|             |               | F             | 45.73***          | 67.73***                | 37.82***                           | 2.42 ns                                     |
|             | 23 April      | -             | 93.5 $\pm$ 4.0    | 1.1 $\pm$ 0.1           | 4.0 $\pm$ 0.2                      | 4.3 $\pm$ 0.1                               |
|             |               | +             | 53.7 $\pm$ 4.9    | 0.6 $\pm$ 0.1           | 1.3 $\pm$ 0.4                      | 2.3 $\pm$ 0.6                               |
|             |               | F             | 30.61**           | 34.65***                | 25.40**                            | 15.25**                                     |
| Plains      | 14 April      | -             | 91.0 $\pm$ 2.4    | 1.21 $\pm$ 0.18         | 4.3 $\pm$ 1.2                      | 4.7 $\pm$ 0.1                               |
|             |               | +             | 36.8 $\pm$ 7.2    | 0.38 $\pm$ 0.25         | 0.9 $\pm$ 0.2                      | 2.2 $\pm$ 0.2                               |
|             |               | F             | 63.77**           | 124.22***               | 392.57***                          | 97.81***                                    |
|             | 12 May        | -             | 95.3 $\pm$ 2.0    | 1.43 $\pm$ 0.32         | 5.7 $\pm$ 0.6                      | 5.9 $\pm$ 0.6                               |
|             |               | +             | 68.2 $\pm$ 4.7    | 0.73 $\pm$ 0.22         | 1.9 $\pm$ 0.3                      | 2.7 $\pm$ 0.2                               |
|             |               | F             | 54.89***          | 37.17***                | 37.56***                           | 23.69**                                     |
|             | 3 June        | -             | 94.4 $\pm$ 2.0    | 1.77 $\pm$ 0.34         | 7.5 $\pm$ 0.7                      | 7.9 $\pm$ 0.6                               |
|             |               | +             | 40.7 $\pm$ 6.0    | 0.41 $\pm$ 0.18         | 0.9 $\pm$ 0.1                      | 2.1 $\pm$ 0.2                               |
|             |               | F             | 288.74***         | 142.16***               | 104.88***                          | 104.43***                                   |

ns, \*\*, \*\*\* indicate not significant and significant at  $P = 0.01$  and  $P = 0.001$ , respectively.

<sup>1</sup>Rating scale of Windstrom (1967) where 0 is no damage.



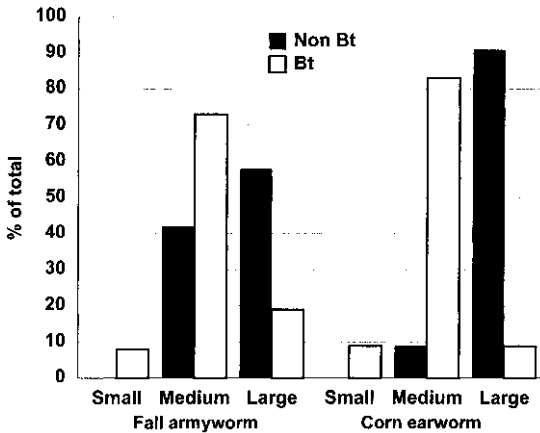


Fig. 1. Size distribution fall armyworm and corn earworm larvae in ears of susceptible and Bt resistant corn hybrids at Attapulgus, GA, 1998.

YieldGard resistance at Attapulgus prevented significant yield ( $F = 113.29$ ;  $df = 1, 9$ ;  $P = 0.0001$ ) losses in all brands of hybrids (Fig. 2). Resistance in this trial prevented an average yield loss of 28% which equaled 2141 kg/ha (=34.1 bu/acre). Grain yields at Tifton were not significantly different between Bt resistant types in either planting (Date 1:  $F = 0.14$ ;  $df = 1, 9$ ;  $P = 0.72$ ; Date 2:  $F = 0.08$ ;  $df = 1, 9$ ;  $P = 0.79$ ).

Grain yields at Plains were low for all planting dates with yields being greatest on the second planting date (Fig. 2). Average grain yields were not significantly ( $F = 0.01$ ;  $df = 1, 12$ ;  $P = 0.98$ ) different between susceptible and resistant hybrids on the first planting date. YieldGard resistance prevented significant grain yield losses of 21.8% during the second planting date ( $F = 15.27$ ;  $df = 1,$

12;  $P = 0.0021$ ) and 74.5% during the third planting date ( $F = 128.41$ ;  $df = 1, 9$ ;  $P = 0.0001$ ).

Grain aflatoxin concentrations were extremely high at Attapulgus and in the first planting at Plains. Aflatoxin progressively declined to low levels with later plantings at Plains (Fig. 3). Grain aflatoxin concentrations were not significantly different between susceptible and Bt-resistant hybrids in any trial (Attapulgus:  $F = 2.58$ ,  $df = 1, 9$ ,  $P = 0.14$ ; Plains PD1:  $F = 0.50$ ,  $df = 1, 12$ ,  $P = 0.50$ ; Plains PD2:  $F = 0.20$ ,  $P = 0.66$ ; Plains PD3:  $F = 0.81$ ,  $df = 1, 9$ ,  $P = 0.40$ ).

DISCUSSION

YieldGard Bt resistance consistently prevented whorl infestation and damage by fall armyworm. Even when larvae established on resistant plants, whorl damage of infested plants was substantially reduced. YieldGard resistance also reduced lepidopteran infestations and the number of larvae in ears, but larval establishment did occur on many ears of resistant plants. Indeed, at Attapulgus, where large numbers of fall armyworm predominated, Bt resistance did not reduce the percentage of infested ears. However, once established in ears, larvae of both species developed more slowly and caused much less kernel damage on ears of resistant than susceptible plants. The lack of significant brand by resistance interactions for any variable measured also verifies that hybrids with the Bt11 and MON810 events were similar in efficacy controlling in whorl and ear infestations for both species.

Despite reports showing an association between lepidopteran damage and aflatoxin contamination of corn grain (e.g., Windstrom 1979; McMillian 1983; McMillian et al. 1985; Smith & Riley 1992), we found that YieldGard Bt resistance

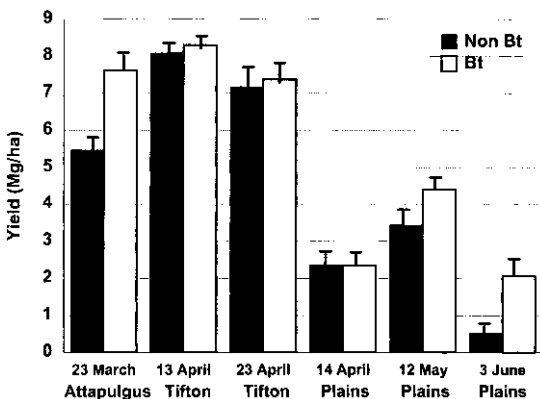


Fig. 2. Grain yield ( $\pm$ SE) of susceptible and Bt resistant corn hybrids planted at three location in southern Georgia in 1998.

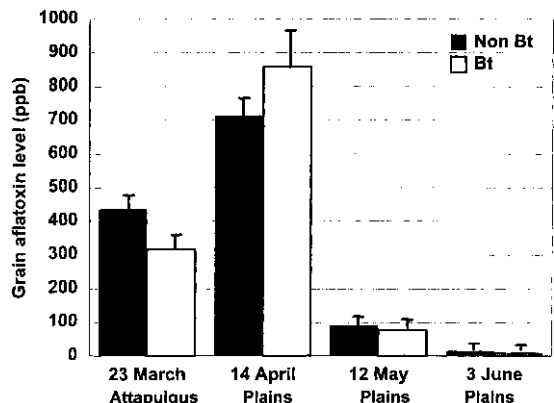


Fig. 3. Grain aflatoxin concentration ( $\pm$ SE) of susceptible and Bt resistant corn hybrids at Attapulgus and three planting times at Plains, GA in 1998.

did not affect aflatoxin concentrations in grain. Although YieldGard Bt resistance prevented most kernel damage, larvae frequently established on resistance ears. Newer events with high levels of toxin expression that virtually prevent larval establishment may be needed to effectively test the hypothesis that lepidopterans infestations enhance aflatoxin contamination of corn grain.

Growing conditions in 1998 were hot and very dry causing most dryland corn production to be destroyed. Yield of irrigated corn also was reduced because of high temperatures during most of the season and especially during pollination and silking. Hybrid yields of plantings at Plains were low and do not permit useful economic comparisons of the value of YieldGard technology. However, grain yields at Attapulugus were typical of this location in 1998. Assuming no large differences in grain quality and at a grain price of \$76.52 per Mg (= \$2.00 per bu), the average yield loss of 2141 kg/ha (= 34.1 bu/acre) produced a \$168.45 per ha (= \$68.20 per acre) gross return from YieldGard technology in this one trial. However, economic benefit must be more extensively evaluated under a variety of growing conditions and insect infestations levels to clearly assess the value of YieldGard technology to corn growers in the Southeast.

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FECUNDITY AND LONGEVITY OF *DIAPETIMORPHA INTROITA* (CRESSON)  
(HYMENOPTERA: ICHNEUMONIDAE) REARED ON ARTIFICIAL DIETS:  
EFFECTS OF A LIPID EXTRACT FROM HOST PUPAE AND CULTURE  
MEDIA CONDITIONED WITH AN INSECT CELL LINE

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ABSTRACT

*Diapetimorpha introita* (Cresson) (Hymenoptera: Ichneumonidae) is a native ectoparasitoid of *Spodoptera* spp. pupae. This parasitoid has been reared in the laboratory on an artificial diet devoid of any insect host components. However, wasps reared on this artificial diet had reduced fecundity. Efforts to increase fecundity included supplementing the diet with cell culture media conditioned with a cell line from ovaries of the fall armyworm, *S. frugiperda*, in one experiment and fortifying the diet with lipids extracted from pupae of *S. frugiperda* in a second experiment. In the first experiment, differences in mean oviposition and mean longevity among females reared on the artificial control diet (*artificial diet*), cell line-supplemented diet (*Sf9Cell*), and natural host (*Host*) were not significant. However, during the first 10 days of oviposition, *Sf9Cell*-reared females oviposited at a rate similar to the *Host*-reared parasitoids and at a rate faster than *artificial-diet* reared females. In the second experiment, females reared on the diet with added host lipid (*host lipid*) laid significantly more eggs than females on the *artificial diet*, however, longevity was not significantly affected by diet treatment. We conclude that total egg production by *D. introita* was improved on artificial diet supplemented with lipids from the natural host but was not increased by the addition of materials produced by an ovarian cell line derived from *S. frugiperda*. Future research efforts should focus on increasing fecundity of wasps reared on the artificial diet by identifying the lipid(s) or lipid-soluble material in the host pupal extract that is responsible for enhancing egg production in *D. introita* females.

Key Words: *Diapetimorpha introita*, *Spodoptera*, parasitoid, artificial diet, fecundity, host lipids, insect cell line

RESUMEN

*Diapetimorpha introita* (Cresson) (Himenóptera: Ichneumonidae) es un ectoparásito nativo en pupas de especies de *Spodoptera*. Este parásito ha sido criado en el laboratorio en una dieta artificial desprovista de componentes de insecto hospedero. Sin embargo, avispas criadas en esta dieta artificial tuvieron fecundidad reducida. Esfuerzos para incrementar la fecundidad incluyeron: suplir la dieta con medio de cultivo de células acondicionadas con una línea de células de ovarios de *S. frugiperda* en un experimento, y fortificando la dieta con lípidos extraídos de pupas de *S. frugiperda* en un segundo experimento. En el primer experimento, diferencias en oviposición promedio y longevidad promedio entre hembras criadas bajo la dieta artificial de control (*artificial diet*), la dieta complementada con línea de células (*Sf9Cell*), y hospedero natural (*Host*) no fueron significantes. Sin embargo, durante los primeros 10 días de oviposición, hembras criadas con *Sf9Cell* ovipositaron a una velocidad similar a los parásitos criados con *Host* y a una velocidad más rápida que hembras criadas con *artificial diet*. En el segundo experimento, hembras criadas con la dieta complementada con lípidos de hospedero (*host lipid*) pusieron significativamente mas huevos que hembras con *artificial diet*, sin embargo, la longevidad no fue afectada significativamente por el tratamiento de dieta. Concluimos que producción total de huevos por *D. introita* fue mejorada por la dieta artificial complementada con lípidos del hospedero natural pero no fue incrementada por la adición de materiales producidos por la línea de células de ovario derivada de *S. frugiperda*. Futuros esfuerzos de estudio deberán enfocarse en incrementar la fecundidad de avispas criadas con la dieta artificial al identificar el lípido (s) o material soluble en lípidos en el extracto pupal de hospedero que es responsable por aumentar la producción de huevos en hembras de *D. introita*.

*Diapetimorpha introita* (Cresson) (Hymenoptera: Ichneumonidae) is a native ectoparasitoid of *Spodoptera* spp. (Pair & Gross 1984) that has been reared in the laboratory on an artificial diet devoid of any insect components (Carpenter & Greany 1998; Greany & Carpenter 1996). Female parasitoids that are reared on this artificial diet are able to search for and parasitize natural hosts in the field (Carpenter and Greany 1998). However, survival rate, fecundity, and weight are less for diet-reared *D. introita* than for host-reared *D. introita*. Also, developmental time is significantly longer for wasps reared on the artificial diet than for wasps reared on host pupae (Carpenter & Greany 1998). Efforts to increase wasp weight and reduce developmental time have included the addition of commercial nutrients, the use of culture media conditioned by insect cell lines, and supplementing the diet with lipid extracts from host pupae (Ferkovich et al. 1999; Ferkovich et al., in press). One of the cell lines, Sf, derived from ovaries of *S. frugiperda* resulted in some improvement in wasp weight (Ferkovich et al. 1999), whereas, the use of a lipid extract from *S. frugiperda* not only enhanced the average weight of the males and females but also reduced their developmental time. Other parameters such as cocoon production or adult emergence were unaltered. Molting hormone titers of diet-reared and host-reared *D. introita* were examined and it was concluded that insufficient ecdysteroid in the hemolymph during metamorphosis may contribute to the lowered emergence in wasps reared on the artificial diet (Gelman et al. 1999).

In view of some of the positive effects on growth and development of *D. introita* with dietary supplements of extracted host lipids and cell line-conditioned media (Ferkovich et al. 1999, Ferkovich et al., in press), we decided to examine their effects on fecundity and longevity of *D. introita* females.

## MATERIALS AND METHODS

### Insect Rearing

Insects used in this study were obtained from laboratory colonies at the Crop Protection and Management Research Unit, Tifton, GA. *D. introita* were reared according to the methods described by Pair (1995), unless noted otherwise. *S. frugiperda* larvae were reared in plastic cups (30 ml) containing meridic diet (Burton 1969) at a photoperiod of 14:10 (L:D) h and temperature of  $28 \pm 1$  and  $25 \pm 1^\circ\text{C}$ , respectively.

### Diet Preparation and Encapsulation of Diet

The original artificial diet (control diet) contained ground beef liver, chicken egg yolk, and the amino acid L-glutamine (Sigma, St. Louis, MO)

and was prepared according to Carpenter and Greany (1998) under aseptic conditions in a clean room as described by Ferkovich et al. (1999). All the ingredients were added to 25 ml of serum-free SF-900 II cell culture medium. The diet was encapsulated in Parafilm® using a diet encapsulation apparatus (Greany & Carpenter 1996). Diet was dispensed at 0.5 ml of diet/dome with 24 domes/sheet. Each diet sheet was covered with a modified (bottomless) Falcon® tissue culture plate (Sigma, St. Louis, MO) so that each dome (one larva/dome) was situated within a well. The entire culture plate was covered with a Plexiglas® plate to prevent escape of the larvae. Diet was changed during larval development four days after the neonates were initially placed on the diet.

### Preparation of Cell line-supplemented Diet

The Sf9 cell line was an embryonic line originally derived from ovaries of the fall armyworm, *S. frugiperda*, and purchased from ATCC, Rockville, MD. The cells were cultured in Grace's medium with 10% fetal bovine serum (FBS), 1% bovine serum albumin (BSA) and 0.33% lactalbumin enzymatic hydrolysate (Sigma, St. Louis, MO). For larger-scale culture of the cell lines, cells were grown in 250 ml magnetic spinner flasks (Bellco Glass, Vineland, NJ) at  $29^\circ\text{C}$  and were grown to densities of  $1.3 \times 10^5$  to  $2 \times 10^5$  cells/ml 10 days post inoculation. For the experiments, 25 ml of cell suspension were centrifuged at  $250 \times g$  for 2 min at room temperature. The resultant cell-conditioned supernatant then was substituted for the SF-900-II medium in preparing the artificial treatment diets. Two cell line control diets were also tested to measure the effects of Grace's culture medium and the additives FBS, 1% BSA and 0.33% lactalbumin enzymatic hydrolysate, additives that were required for optimal cell growth.

### Preparation of Diet With Extracted Host Pupal Lipids

Lipids were extracted using a modified method of Folch et al. (1957) as described by Ferkovich et al. (in press). Briefly, twenty-four 4 day-old pupae of *Spodoptera frugiperda* pupae were homogenized in 12.5 ml of Ringers solution (Ephrussi & Beadle 1936); the homogenate was filtered through glass wool to remove cuticular debris and the filtrate saved. The filtrate was then extracted with a chloroform:methanol (2:1) mixture and the chloroform phase was dried down.

Twenty-five ml of diet were added to the dried chloroform extract and the flask was rotated for 5 min to dissolve the residue. The chloroform extract of freshly homogenized pupae of *S. frugiperda* was added to the artificial diet so that each diet dome contained one pupal equivalent of lipid per *D. introita* larva.

## Treatment Diets

The treatment diets used in this study were as follows: 1) *Host*, *S. frugiperda* pupae; 2) *artificial diet*, original control diet; 3) *host lipid*, original diet containing chloroform-extracted lipids from freshly homogenized *S. frugiperda* pupae (prepared according to the methods described by Ferkovich et al., in press); 4) *Sf9Control<sub>A</sub>*, artificial diet prepared with Grace's cell culture medium, 5) *Sf9Control<sub>B</sub>*, artificial diet prepared with Grace's cell culture medium with % bovine serum albumin, 10% fetal bovine serum and lactalbumin enzymatic hydrolysate; and 6) *Sf9Cell*, diet prepared with S9 cell-conditioned Grace's medium with 1% bovine serum albumin, 10% fetal bovine serum and lactalbumin enzymatic hydrolysate.

## Bioassay

First instar larvae that hatched within a 12 hour period were placed on encapsulated diet domes (one larva/dome) in individual cells of a 24 well plate. Each treatment was replicated four times. The larvae were allowed to feed and de-

velop to adults (described below) at  $29.1 \pm 1^\circ\text{C}$  and 70% RH. Diet was replaced four days after the neonates were initially placed on the diet domes. The third instar larvae were transferred to the new diet domes using a camel hair brush. A 24 well plate containing 24 larvae on a diet constituted one replication. As the adults emerged, they were held individually in plastic portion cups (102 cc) for 24 hrs before they were weighed.

## Oviposition and Longevity Studies

Ten male and ten female wasps from each treatment were randomly selected, weighed 24 h post emergence, and paired in small (480 ml) plastic containers fitted with screened lids. Each container was maintained with a source of honey and water. A plastic cup (30 ml) containing 15 ml of soil in which a *S. frugiperda* larva had pupated was provided for each female wasp as an oviposition site. Cups were replaced daily and the number of eggs laid by each female wasp was recorded. Longevity of male and female wasps was recorded.

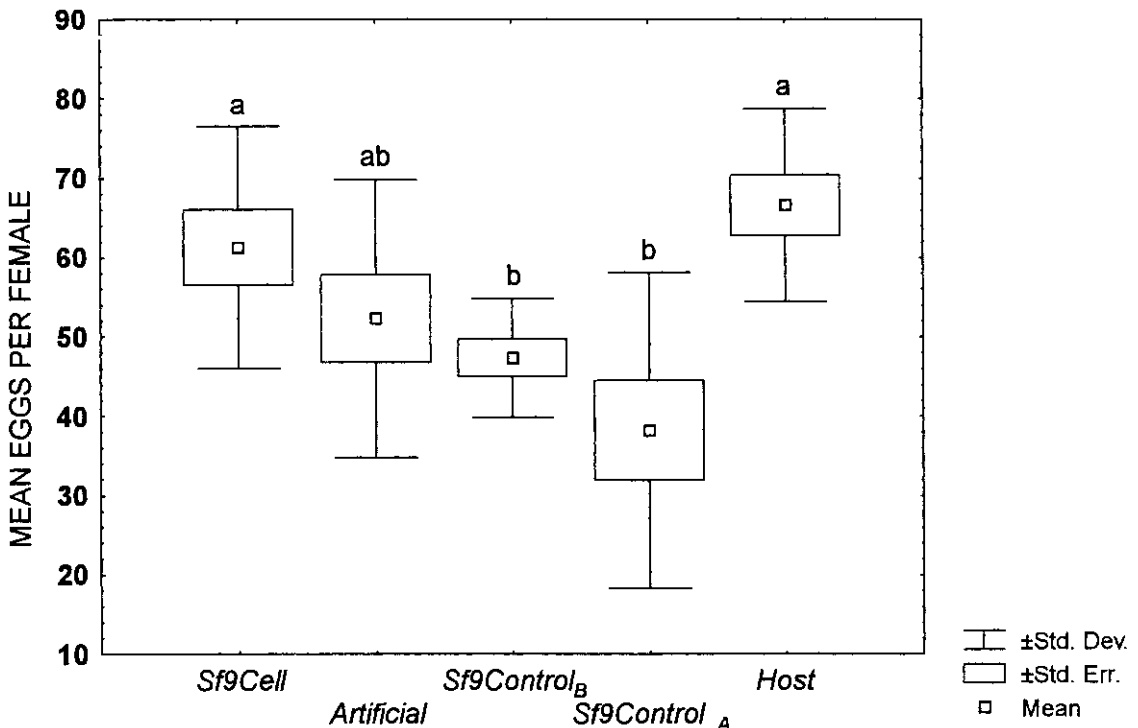


Fig. 1. Comparison of mean oviposition by female *Diapetimorpha introita* reared on: *Host* (*Spodoptera frugiperda* pupae), *artificial diet*, original control diet; *Sf9Control<sub>A</sub>*, artificial diet prepared with Grace's cell culture medium; *Sf9Control<sub>B</sub>*, artificial diet prepared with Grace's cell culture medium with 1% bovine serum albumin, 10% fetal bovine serum and lactalbumin enzymatic hydrolysate; and *Sf9Cell*, diet prepared with S9 cell-conditioned Grace's medium with 1% bovine serum albumin, 10% fetal bovine serum and lactalbumin enzymatic hydrolysate.

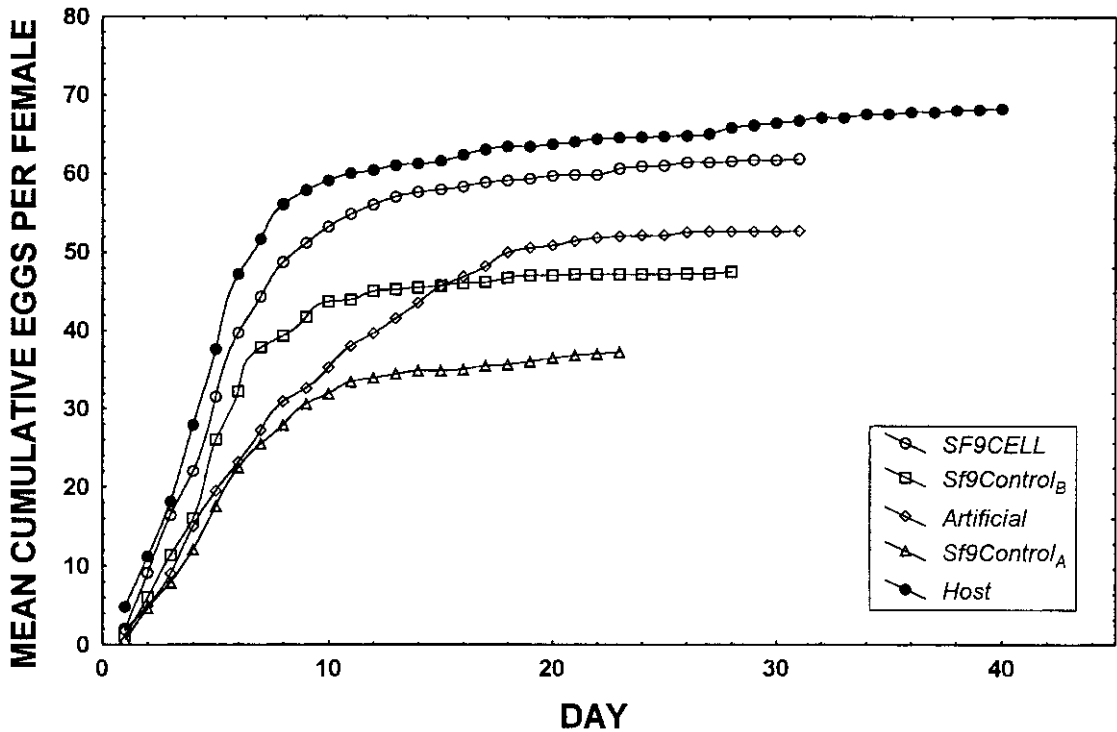


Fig. 2. Rate of oviposition by female *Diapetimorpha introita* reared on: *Host* (*Spodoptera frugiperda* pupae), *artificial diet*, original control diet; *Sf9Control<sub>A</sub>*, artificial diet prepared with Grace's cell culture medium; *Sf9Control<sub>B</sub>*, artificial diet prepared with Grace's cell culture medium with 1% bovine serum albumin, 10% fetal bovine serum and lactalbumin enzymatic hydrolysate; and *Sf9Cell*, diet prepared with S9 cell-conditioned Grace's medium with 1% bovine serum albumin, 10% fetal bovine serum and lactalbumin enzymatic hydrolysate.

#### Statistical Analysis

The treatment means for fecundity and longevity were compared using the *t*-test (Steel & Torrie 1980). Regression analysis (StatSoft 1995) was used to examine the relationship between mean fecundity and female longevity.

### RESULTS

#### Diet Supplementation with Sf9 Cell Line-Conditioned Medium

Mean oviposition of females reared on the *artificial diet*, *Sf9Cell* and *Host* treatments was not significantly different, and only females reared on the *Sf9Cell* and *Host* diet treatments oviposited significantly ( $P < 0.05$ ) more eggs than females reared on the two control diets, *Sf9Control<sub>A</sub>* and *Sf9Control<sub>B</sub>* (Fig. 1). However, during the first ten days females reared on the *Sf9Cell* diet and the *Host* oviposited at a faster rate than females reared on *Sf9Control<sub>A</sub>* diet and *artificial diet* (Fig. 2). When data from all diet treatments were combined, there was a significant ( $P < 0.001$ ,  $R^2 = 0.999$ ) relationship between mean oviposition and

female longevity (Fig. 3). However, there were no significant differences in mean longevity of female wasps among the five treatments (*artificial diet*, 24.4d; *Sf9Cell*, 23.5d; *Host*, 18.8d; *Sf9Control<sub>A</sub>*, 20.1d; and *Sf9Control<sub>B</sub>*, 20.1d).

#### Diet Supplementation with Host Pupal Lipid Extract

Although females developing on the *host lipid* diet and the *artificial diet* demonstrated similar patterns in oviposition (Fig. 4), the mean ( $\pm$  S.D.) number of eggs laid by females reared on the *host lipid* diet ( $46.67 \pm 8.7$ ) was significantly ( $t = 4.39$ ,  $df = 8$ ,  $P = 0.002$ ) more eggs than the number of eggs laid by females reared on the *artificial diet* ( $34.67 \pm 10.5$ ). The difference in longevity of females reared on the two diets was not significant (*artificial diet*,  $16.86 \pm 7.7$  days and *Host Lipid*,  $22.50 \pm 7.7$  days).

### DISCUSSION

The fecundity of females was increased with the addition of lipids from host pupae to the artificial diet. The concentration of lipid added to the diet was one pupal equivalent per parasitoid; this

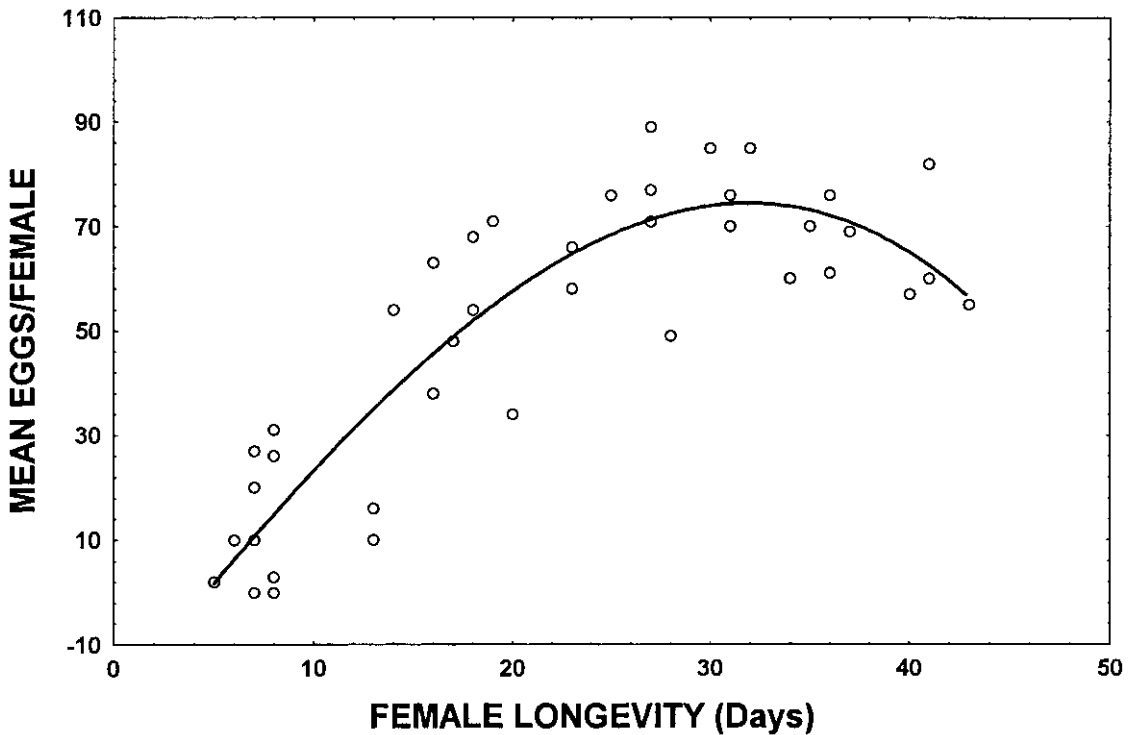


Fig. 3. Relationship between oviposition and longevity for female *Diapetimorpha introita* reared on host pupae (*Spodoptera frugiperda*) and artificial diets ( $y = 21.4 + 4.74x - 0.015x^2 - 0.001x^3$ ,  $R^2 = 0.999$ ,  $P < 0.0001$ ).

concentration was selected because a single parasitoid develops on one host pupa in the wild (Pair 1995). It is possible, however, that fecundity could be improved further with the addition of a higher concentration of host lipid extract. At present, the identity of the bio-active compound active is not known. It could either be a lipid(s) or a lipid-soluble compound(s). If the material is a lipid(s), it would be difficult to speculate as on the identity of the material since the dietary lipid requirements in parasitoids vary with the species. Many species of parasitoids copy the lipid composition of their host (Thompson & Barlow 1972). Others such as *Exeristes roborator* Fab. (Thompson 1977) are able to regulate their fatty acid concentrations in the absence of the dietary lipids. Still others such as *Agria housei* (Shewell) (House 1954) and *Itopectis conquisitro* (Say) (Yazgan 1972) can be reared on a diet without fatty acids but supplementation of fatty acids to the diet improves adult emergence and fecundity. Other parasitoids such as *Pimpla turionellae* (L) require fatty acids in their diet to produce normal looking adults (Yazgan 1981).

Reinecke (1985) stated that all insects have certain lipid dietary requirements, especially the immature stages, however very few of these lipids are essential and only the sterols are universally required. Thus, it is interesting that the lipids

present in the egg yolk component of the artificial diet did not adequately support fecundity of *D. introita*. Egg yolk-based diets have been used to rear a number of parasitoids and predators (Grenier et al. 1994; Nelson 1999). The egg yolk in this artificial diet either lacked the required lipid(s) or contained the needed lipid(s) but not in levels adequate for higher fecundity. Supplementing the diet with host lipids apparently provided a better nutritional balance to the diet, allowing the *D. introita* females to oviposit at a significantly higher rate than females reared on the artificial control diet. Bracken (1969) found that sustained egg production for adults of the parasitoid, *Exeristes comstockii* was dependent on a balance of nutrients in the artificial diet.

Wheeler (1996) indicated that oogenesis is typically a nutrient-limited process and is initiated only if sufficient nourishment is taken for egg production. Nourishment for *D. introita* egg production is apparently acquired during the larval stage because the ovaries of the females are well developed at adult emergence (pers. obs.), and females are able to produce eggs throughout their adult lives by feeding only on honey and water (Pair 1995).

Cell line-conditioned media have been used to improve the growth of two endoparasitoids, *Lysiphlebus fabarum* (Marshall) (Rotundo et al. 1988)

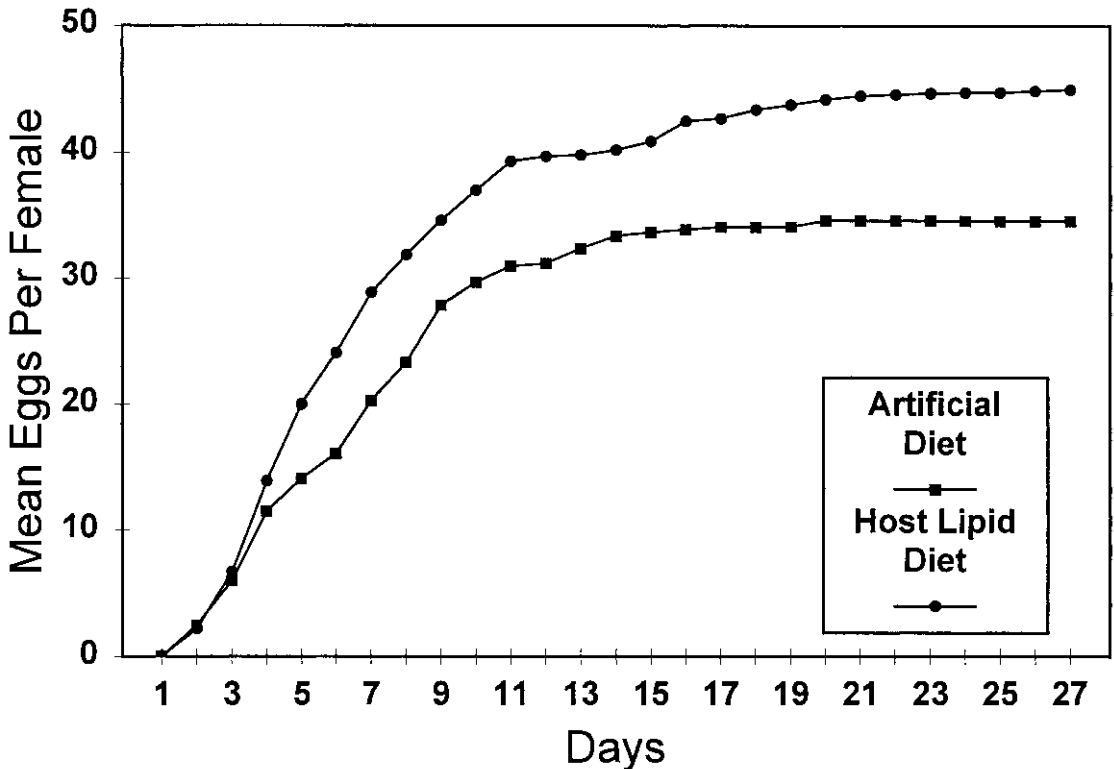


Fig. 4. Rate of oviposition by female *Diapetimorpha introita* reared on: *artificial diet*, original control diet; and *host lipid*, original diet containing chloroform-extracted lipids from freshly homogenized *Spodoptera frugiperda* pupae (prepared according to the methods described by Ferkovich et al., in press).

and *Microplitis croceipes* (Ferkovich et al. 1994), and an egg ectoparasitoid, *Edovum puttleri*, (Hu et al. 1999). However, fecundity could not be assessed in these studies because the insects either did not develop to the adult stage or only low numbers of adults emerged on the cell line-supplemented diets. In this study, we were able to examine the effects of the cell conditioned medium on fecundity because successive generations can be produced on the *artificial diet* (Carpenter & Greany 1998). The positive effect the *Sf9Cell* diet had on the increased rate of oviposition with no accompanying effect on the mean oviposition rate was interesting. It appears that the cell line produced an unknown material that induced the females to deposit their eggs in a pattern that paralleled *Host*-reared females and at a rate faster than *artificial diet*-reared females (Fig. 2).

In view of the positive effects of the pupal lipid extract on fecundity of *D. introita*, we suggest that future research should focus on identifying the fecundity-enhancing material(s) from host pupae so that it can more easily be tested at various concentrations in the diet. Moreover, once the identity of the material is known, it may be possible to obtain it from a commercial source.

#### ACKNOWLEDGMENTS

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## SEASONAL ABUNDANCE OF A PUPAL PARASITOID, *DIAPETIMORPHA INTROITA* (HYMENOPTERA: ICHNEUMONIDAE)

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### ABSTRACT

Seasonal abundance of a pupal parasitoid *Diapetimorpha introita* (Cresson) (Hymenoptera: Ichneumonidae) and the beet armyworm *Spodoptera exigua* (Hübner) (Lepidoptera: Noctuidae) was monitored with pheromone-traps. *D. introita* males were caught in wing traps baited with live females, and beet armyworm males were caught in bucket traps baited with synthetic pheromone. The greatest number of *D. introita* adult males was caught during early autumn, approximately one month after the greatest number of beet armyworm males was caught, and represents the most convenient time during which to conduct trapping experiments.

Key Words: sex-attractant, biological control, population dynamics, beet armyworm, fall armyworm, *Spodoptera*, pheromone-trapping

### RESUMEN

La abundancia estacional del parásito pupal *Diapetimorpha introita* (Cresson) (Himenóptera: Ichneumonidae) y de *Spodoptera exigua* (Hübner) (Lepidóptera: Noctuidae) fue observada con trampas de feromona. Machos de *D. introita* fueron capturados en trampas de ala con señuelo de hembras vivas, y machos *S. exigua* fueron capturados en trampas de cubo con señuelo de feromona sintética. La mayor captura de machos adultos *D. introita* fue alcanzada durante el comienzo del otoño, aproximadamente un mes después de que el mayor número de *S. exigua* fue capturado, y representa el tiempo más conveniente para conducir experimentos de trampas.

When the Boll Weevil Eradication Program was initiated in autumn of 1987 over large portions of the southeastern United States, the beet armyworm *Spodoptera exigua* (Hübner) (Lepidoptera: Noctuidae) emerged as the most important threat to cotton production (Haney et al. 1996). Other hosts of economic importance with which the beet armyworm is associated include tomatoes, corn, alfalfa, onions, asparagus, potatoes, and citrus as well as numerous non-economic species (Hendricks et al. 1995). Prior to 1991, Georgia experienced outbreaks during 1977, 1980, 1981, 1988, and 1990 (Douce & McPherson 1991). Difficult to manage, outbreaks have been correlated with less than normal precipitation and disruptive insecticide use (Chandler & Ruberson 1996). Its threat to cotton production has declined in recent years, presumably the result of declining insecticide applications directed at the boll weevil and increased planting of BT-cotton against the beet armyworm (John Ruberson, Department of Entomology, University of Georgia, pers. comm.).

Historically, the fall armyworm *Spodoptera frugiperda* (J. E. Smith) is an important pest of

corn, sorghum, and coastal Bermuda grass (Metcalf et al. 1951). It was not recognized as an important threat to cotton production. Recently, however, the importance of fall armyworm to cotton production has increased (Riley et al. 1997) because BT-cotton is not as effective against it as beet armyworm (Adamczyk et al. 1998).

*Diapetimorpha introita* (Cresson) (Hymenoptera: Ichneumonidae) is a pupal parasitoid native to southern Georgia, and it may be valuable to the biological control of *Spodoptera* spp. if mass propagated and released against incipient populations (Pair & Gross 1989). It has been reared from fall armyworm pupae collected in the field (Pair & Gross 1989). Although *D. introita* has been reared from laboratory strains of the beet armyworm (Carpenter & Greany 1998), a comprehensive survey of pupae in the field has yet to be completed.

Previous attempts to describe population dynamics of *D. introita* have relied upon measuring frequency of parasitism among fall armyworm pupae in the field (Pair & Gross 1989). As investigation of *D. introita* and its importance to the management of *Spodoptera* spp. proceeds, more convenient methods of monitoring populations in

the field are needed (Jewett & Carpenter 1998). One possible approach to monitoring *D. introita* concerns pheromone-trapping males. That *D. introita* males use a sex-attractant to find females is supported by data from laboratory and field bioassays (Jewett & Carpenter 1998). Significantly more males were caught in traps baited with live females than in traps baited either with nothing or with live males. Experiments with a related pupal parasitoid, *Ichneumon promissorius* (Hymenoptera: Ichneumonidae) (Erichson), have yielded further insights concerning extraction of a sex-attractant from *D. introita* females (Jewett & Carpenter 1999).

A lure formulated with the sex-attractant of *D. introita* for conveniently monitoring its populations in the field is anticipated. Field bioassays had been completed previously during spring and early summer, and number of males caught in traps was generally low (Jewett & Carpenter 1998). A greater population reservoir from which to trap *D. introita* males would accommodate more replications and greater resolution. The present study was undertaken to describe local seasonal abundance of *D. introita* males, and to identify the most convenient time of year during which they may be trapped.

For comparison, local seasonal abundance of the beet armyworm also was considered. Although a comprehensive survey of beet armyworm pupae in the field has not been completed, more *D. introita* were reared from beet armyworm pupae than from fall armyworm pupae (Carpenter & Greany 1998). Relative abundance of the fall armyworm was not considered because it is not resident to southern Georgia, but instead migrates annually from Florida (Pair et al. 1986; Wilson 1934). This disposition may be responsible, in part, for inconsistent trapping results and their lacking reliability as indicators of relative abundance (John Ruberson, Department of Entomology, University of Georgia, pers. comm.). Unable to rely upon results of trapping fall armyworm males, beet armyworm was substituted because it is a potential host that responds well to lures.

#### MATERIALS AND METHODS

Twenty different sites at Gibbs and Bellflower Farms in Tift Co. were monitored for *D. introita* adult males during two years. Traps were established as described by Jewett and Carpenter (1998). Briefly, wing traps were supported 0.5 m above ground and were baited with two live females (either mated or unmated) (Fig. 1). Number of males caught was recorded once a week, and trap bottoms were replaced. Lures also were replaced once a week or when a female had expired.

Sex-attractant of the beet armyworm is (z)-9-tetradecen-1-ol (2.5%), (z)-9, 12-tetradecadien-1-ol acetate (87.2%), and (z)-11-hexadecen-1-ol ace-

tate (10.3%). It has been used previously to trap beet armyworm adult males (Hendricks et al. 1995; Mitchell & Tumlinson 1994), and is as reliable an indicator of seasonal occurrence of beet armyworm adults in the field as live virgin females (Mitchell & Tumlinson 1994). Beet armyworm adult males were monitored at Gibbs and Rigdon Farms in Tift Co., Georgia with traps established as described by Ruberson and Herzog (1997, 1998). Briefly, bucket traps were suspended 1.5 m from the ground and were baited with lures of synthetic pheromone (Great Lakes IPM Inc., Vestaburg, MI). Number of males caught in traps was recorded once a week and lures were replaced biweekly.

Adult males of both insects were monitored between June 1997 and December 1998. Occasionally, data were not recorded from beet armyworm traps during 1998 or from *D. introita* traps during 1997. Data were not recorded from beet armyworm traps during the last two weeks of December in 1997 and the first two weeks of January in 1998. Data were not collected from *D. introita* traps during January 1997.

#### RESULTS AND DISCUSSION

Although pheromone-traps have been used to detect insects at remarkably low population densities, the number caught often does not reflect actual density (Cardé & Elkinton 1984). However, data from trapping programs often are valuable for relative estimates (Evans 1984). Results of trapping *D. introita* adult males in the present study are generally consistent with those of the previous study by Pair and Gross (1989), although some differences do exist. In the present study, greatest number of *D. introita* males was caught during September and early October of both years (Fig. 2). Pair and Gross (1989) also reported that emergence of *D. introita* from fall armyworm pupae was greatest during September and October. During the present study, twenty one males were caught over 2 days in one trap at Gibbs Farm in October, and two traps caught 23 males each during one week in September at Bellflower Farm (Fig. 2).

*D. introita* first appeared during May in the study by Pair and Gross (1989), but in the present study, males first appeared during the end of March (Fig. 2). *D. introita* overwinter in their hosts, and this difference in date of appearance may reflect sensitivity of pheromone-traps to low numbers that have emerged after overwintering. Although Pair and Gross (1989) did not detect *D. introita* adults between mid-July and end of August, small numbers were detected during that time in the present study (Fig. 2).

Greatest number of beet armyworm adult males was caught in traps during August and September. Total number caught exceeded 500 on



Fig. 1. In the field, wing trap baited with *Diapetimorpha introita* (Cresson) (Hymenoptera: Ichneumonidae) females.

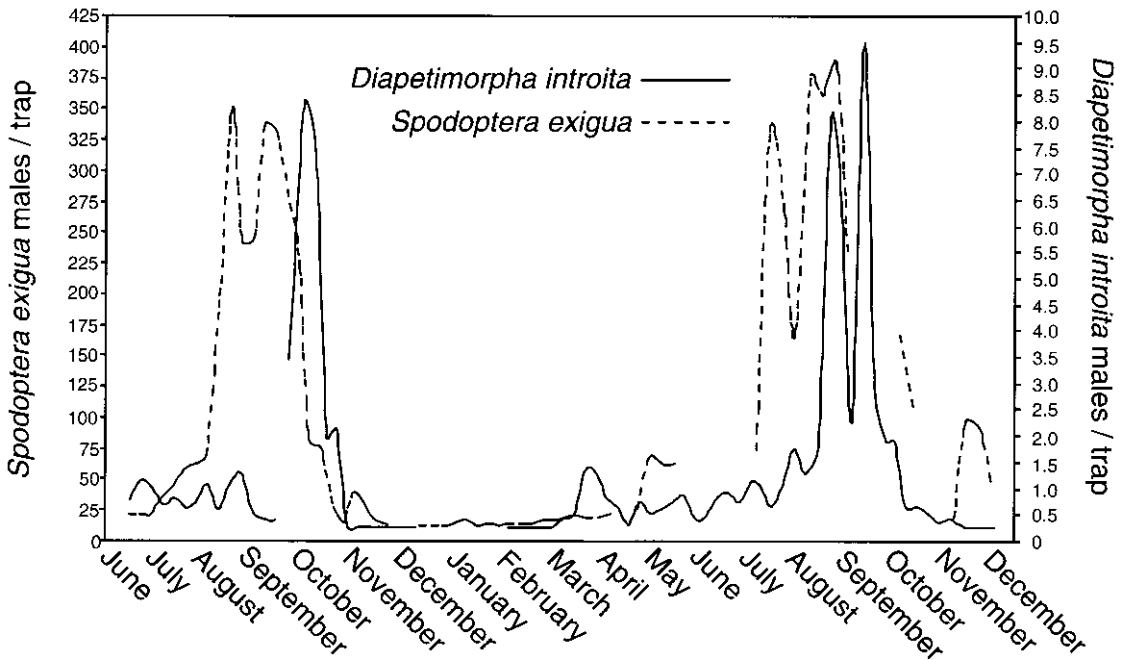


Fig. 2. Average number of *Diapetimorpha introita* (Cresson) (Hymenoptera:Ichneumonidae) males caught in wing traps baited with live females (solid line) and average number of beet armyworm *Spodoptera exigua* (Hübner) (Lepidoptera: Noctuidae) males (intermittent line) caught in bucket traps baited with synthetic pheromone. Breaks in either line represent weeks during which data were not collected.

only 4 dates in 1997, but never exceeded 1,000 as it had in previous years (Ruberson & Herzog 1998). Total number caught in 1998 was comparable to 1997 (Ruberson & Herzog 1997, 1998).

DeBach and Smith (1941) demonstrated that oscillations are inherent to host-parasitoid systems. They predicted that if a parasitoid is host-specific and more successful at finding abundant hosts, then reduction in host numbers would result in decreased number of parasitoids, permitting host numbers to rise (DeBach & Smith 1941). Relative abundance of the beet armyworm and *D. introita* males reflects oscillations predicted by DeBach and Smith (1941) (Fig. 2). Greatest number of *D. introita* males trapped for both years followed the greatest number of beet armyworm males trapped by approximately one month, which is consistent with combined development times of beet armyworm and *D. introita* (Wilson 1934; Pair 1995).

Pair and Gross (1989) concluded that abundance of *D. introita* reflects availability of its host. Although a direct host-parasitoid relationship between the beet armyworm and *D. introita* has not been formally demonstrated in the field, it is supported by their population curves in the present study and the successful rearing of *D. introita* on laboratory strains of the beet armyworm (Carpenter & Greany 1998). Furthermore, the fall army-

worm is migratory from Florida (Pair et al. 1986; Wilson 1934) and the beet armyworm could satisfy any requirement *D. introita* has for an alternate host in southern Georgia.

As investigation of their importance to biological control of *Spodoptera* spp. proceeds, more convenient methods of monitoring *D. introita* in the field are needed. The development of lures for monitoring the relative abundance of males is anticipated, but a need for populations large enough to accommodate the convenient testing of different formulations has been expressed. Results of the present study suggest that the best time during which to test different formulations of lures is during early autumn, approximately one month following greatest relative abundance of beet armyworm adult males.

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COMPARATIVE STUDIES OF THREE POPULATIONS OF THE LADY BEETLE PREDATOR *HIPPODAMIA CONVERGENS* (COLEOPTERA: COCCINELLIDAE)JOHN J. OBRYSKI<sup>1</sup>, ELLIOT S. KRAFSUR<sup>1</sup>, CARLOS E. BOGRAN<sup>2</sup>, LUIS E. GOMEZ<sup>3</sup> AND RONALD E. CAVE<sup>4</sup><sup>1</sup>Department of Entomology, Iowa State University, Ames, IA 50011-3140<sup>2</sup>Texas A & M University, Department of Entomology, College Station, TX 77843<sup>3</sup>a - Calle A, 10-66 Zona 3, Colonia Bella Vista, Quetzaltenango, Guatemala<sup>4</sup>Departamento de Proteccion Vegetal, Escuela Agricola Panamericana, El Zamaorano, Honduras

## ABSTRACT

Allozyme electrophoresis showed much genetic variation in *Hippodamia convergens*, suggesting the possibility of geographic genetic differentiation. Twenty-two of 31 putative allozyme loci resolved on acrylamide gels from *H. convergens* populations were polymorphic (71%). Heterozygosity (diversity) averaged over all loci was  $21.3 \pm 4.2\%$ . However, thirteen polymorphic loci examined in F1 Honduran  $\times$  Iowa hybrids indicated that all alleles were shared in the two populations. In addition, no significant geographic variation was observed in developmental and reproductive responses of *H. convergens* from Iowa, California, and Honduras to aphid prey densities. All inter-population and backcrosses produced fertile eggs. Adult body size of *H. convergens* from Iowa and Honduras was similar. This study indicates that augmentatively released California *H. convergens* could successfully mate with local *H. convergens* populations in Iowa and Honduras.

Key Words: convergent ladybeetle, allozymes, gene diversity, augmentation, biological control

## RESUMEN

Electroforesis de alozima demostró gran variación genética en *Hippodamia convergens*, sugiriendo la posibilidad de diferenciación genética por geografía. Veintidós de 31 lugares de alozimas putativas resueltas en geles de acrilamida de poblaciones de *H. convergens* fueron polimórficos (71%). Heterocigosidad (diversidad) promedio sobre todos los lugares fue  $21.3 \pm 4.2\%$ . Sin embargo, trece lugares polimórficos examinados en híbridos F1 Hondureño  $\times$  Iowa indicaron que todos los alelos fueron compartidos en ambas poblaciones. Adicionalmente, no se observó variación geográfica significativa en respuestas reproductivas y de desarrollo de *H. convergens* de Iowa, California, y Honduras a densidades de presa de áfido. La inter población y cruces de híbridos "backcross" produjeron huevos fértiles. El tamaño del cuerpo adulto de *H. convergens* de Iowa y Honduras fue similar. Este estudio indica que *H. convergens* liberado aumentativamente pudiera aparearse satisfactoriamente con poblaciones locales de *H. convergens* en Iowa y Honduras.

Yearly mass collections and augmentative releases of overwintering adult *Hippodamia convergens* from California are made for aphid suppression, even though evidence for effectiveness is limited (Hagen 1962; Majerus 1994; Hodek & Honek 1996; Obrycki & Kring 1998). Dreistadt & Flint (1996) reported a temporary 3-day decline in aphid densities following release of *H. convergens* adults. Release of *H. convergens* from California may have negative effects on local populations of *H. convergens* because of the distribution of pathogens and parasitoids found in adults (Lipa & Steinhaus 1959; Sluss 1968; O'Neil et al. 1998). An additional concern that has been raised for *Danaus plexippus* L., a species that is also distributed widely by humans (Brower et al. 1995), relates to the idea that

unique local populations with favorable co-adapted genotypes may be compromised by releases of foreign genotypes. Characterization of *H. convergens* intra-specific variation is needed to assess the potential effects of releases of California beetles on local *H. convergens* populations.

One trait that has been examined in *H. convergens* populations from Arizona, Cuzco (Peru), New York, and Oregon, is the thermal requirement for development (Butler & Dickerson 1972; Escalante 1972; Obrycki & Tauber 1982; Miller 1992). Consistency in developmental thresholds of *H. convergens* across geographically separated populations in North America has been reported by Miller (1992). However, earlier studies on *H. convergens* reported differences in thermal responses between populations from Arizona and

New York (Butler & Dickerson 1972; Obrycki & Tauber 1982). A second set of traits that may be used to characterize intra-specific variation are those related to predator responses to prey species, e.g., prey suitability (Tauber et al. 1995).

The objectives of this study were to 1) compare allozyme variation and developmental responses of *H. convergens* from two North American (Iowa and California) and one Central American (Honduras) population to aphid prey, and 2) test for reproductive isolation among *H. convergens* populations from Honduras, Iowa, and California.

## MATERIALS AND METHODS

### Developmental Characteristics

In 1994, *Hippodamia convergens* adults were collected at the Escuela Agrícola Panamericana, Zamorano, Honduras, and in Story and Marion Counties, Iowa. Approximately 50 adult *H. convergens* were sent from Honduras to the USDA-ARS, Beneficial Insects Introduction Research Laboratory, Newark, DE, where they were reared for one generation. First laboratory generation adults were sent to Iowa State University. Six lines, each descended from a different single pair mating, were established from Honduras beetles and 10 lines were established from Iowa beetles; eggs were collected daily. Pairs were kept at 24°C, (L:D) 16:8 and fed pea aphids, *Acyrtosiphon pisum* (Harris), and green peach aphids, *Myzus persicae* (Sulzer) *ad libitum*. Forty larvae from each pair were reared individually on 2-4 *A. pisum* per day at 24°C, (L:D)16:8. Observations on larval survival and developmental stage were made every 24 h.

In 1997, *H. convergens* were collected in Ames, Iowa, and in the Departamento Francisco Morazan, Honduras. Adults from California were provided by Gardens Alive, Lawrenceburg, IN. Five pairs from each population, (California, Iowa, and Honduras) were maintained at 24°C, 16:8 L:D; eggs were collected daily. To examine developmental responses to prey density, 25 first instars from each population were reared on three levels of *A. pisum*: two per day, three per day and >20 per day.

### Inter-population Crosses and Reproductive Responses

In 1994, 30 pairs of second laboratory generation individuals were established using virgin females from Honduran and Iowan populations: 3 pairs were Honduras × Honduras crosses, 3 were Iowa × Iowa crosses, 12 were Iowa female × Honduras male, and 12 were Honduras female × Iowa male. Pairs were fed daily *ad libitum* with *A. pisum* and *M. persicae*. Eggs were collected every day for seven days to determine fecundity (number of eggs laid) and fertility (proportion of fertile

eggs). Following eclosion of at least half of the eggs in each egg mass, the egg masses and newly eclosed larvae were frozen to avoid cannibalism. The number of fertile eggs in each egg mass was estimated by adding eclosed larvae and darkened eggs. Eggs were considered infertile if they were pale yellow and slightly shrunken.

Fifteen larvae from each of the thirty mating pairs were reared individually at 24°C, (L:D)16:8. Twelve adults from each cross were used in a backcross experiment. Twelve pairs were backcrossed to Honduras, [(H×IA) × (H×H)], 12 were backcrossed to Iowa [(H×IA) × (IA×IA)], and 12 were Honduras × Iowa reciprocal crosses [(H×IA) × (H×IA)]. The pairs were fed daily with *A. pisum* and *M. persicae*. Eggs were collected every day for 7 days to determine fecundity and fertility. Observations of larval development and survival were made every 24 h on ten larvae from each pair.

In 1997, F1 adults reared from Iowa, California, and Honduras populations were crossed. Thirty pairs were established: 7 pairs were Iowa females × Honduras males, 7 were Iowa males × Honduras females, 3 were Iowa females × California males, 6 were Iowa males × California females, 6 were California females × Honduras males, and 1 pair was California male × Honduras female. The pairs were fed >20 *A. pisum* per day for 15 days. When egg masses were observed, they were placed in glass vials and held to determine fertility. Larvae were removed from the vials daily to prevent cannibalism.

### Size of adult *H. convergens* from Honduras and Iowa

To compare morphometric characteristics of *H. convergens* adults from Honduras and Iowa, intercrosses, and reciprocal backcrosses, pronotal and elytral length and width were measured by using NIH image software (Macintosh version 1.57). Before measurements were taken, each adult was pinned through the right elytron at exactly the same distance from the head of the pin. Each beetle was photographed using a color video camera (JVC-TK1070U) mounted on a stereo zoom microscope. The filmed images were digitally captured, amplified and measured in millimeters.

### Genetic Diversity Estimates

To estimate gene diversity, beetles from Honduras and Iowa were killed by freezing and stored at -80°C. Reciprocal crosses of Honduras and Iowa beetles provided hybrid progeny; 46 of which were frozen for genetic analysis. Procedures for preparing ladybeetle homogenates for allozyme electrophoresis, histochemical demonstration of putative loci, and statistical methods were those already published (Krafsur et al. 1996a, b).

Voucher specimens of *H. convergens* are deposited in the Iowa State University Insect Collection.



## Data Analysis

In 1994, data obtained from rearing and adult body measurements were summarized by mating pair and means were calculated. Five variables were compared among the populations, crosses and backcrosses, using analysis of variance (PROC GLM, SAS Institute 1985): developmental time, survivorship, fecundity, fertility and adult body size. Means were separated by using a least significance difference test (LSD). Percentage survival and fertility were arcsine transformed [ $\arcsin(\sqrt{\text{proportion}})$ ] before analysis. In 1997, two-way ANOVA was used to compare the effects of aphid prey and population of *H. convergens* on development and survival.

A preliminary analysis revealed that the adult body measurements were correlated to each other. Thus, adult size was compared among groups using an average of the four standardized body measurements. The standardization was done by subtracting the measurement mean among groups from each measurement, and then dividing by the standard deviation among groups.

## RESULTS

## Developmental Responses

Individuals from Honduras required approximately 3-4 more days to complete preimaginal development than individuals from Iowa, Honduras  $\times$  Iowa crosses, and the backcrosses to Honduras and Iowa (Table 1). Similarly, in 1997, Honduran beetles reared on 2*A. pisum* per day required  $27.3 \pm 3.2$  days to complete development, approximately 2-5 days longer than the Iowa and California beetles (Table 2). Developmental time varied with population ( $F = 98.63$ ;  $df = 2,101$ ;  $P = 0.001$ ), sex ( $F = 7.89$ ;  $df = 1,101$ ;  $P = 0.048$ ), diet ( $F = 514.6$ ;  $df = 2,101$ ;  $P = 0.001$ ) and the interaction

between population and diet ( $F = 14.3$ ;  $df = 4,101$ ;  $P = 0.012$ ).

Preimaginal survival of *H. convergens* from Honduras and Iowa and the Honduras - Iowa crosses ranged from 78 to 89% (Table 3). In 1997, survivorship of Iowa, California, and Honduras beetles on three levels of aphid prey was similar; no effect of population ( $F = 5.81$ ;  $df = 2,4$ ;  $P = 0.066$ ) or diet ( $F = 1.88$ ;  $df = 2,4$ ;  $P = 0.265$ ) was observed (Table 2). For Iowa and Honduran *H. convergens*, survivorship increased with higher levels of *A. pisum* per day, but this was not observed for the California beetles (Table 2).

## Reproductive Responses

No differences were observed among groups in fertility and fecundity, however, large variation within groups was observed in both fecundity and fertility (Table 4). Fertility ranged from 70-76% and 79-94% for the Honduran and Iowan populations, respectively. Fertility ranged from 4-93% for the Honduras  $\times$  Iowa crosses, 66-96% for the backcrosses to Honduras, and 34-97% for backcrosses to Iowa. In 1997, all 30 crosses among California, Iowa, and Honduras *H. convergens* produced similar numbers of fertile eggs (Gomez 1998).

Size of Adult *Hippodamia convergens*

The average pronotal width and length of Honduras and Iowa *H. convergens* was 2.68 and 1.27 mm, respectively (Table 5). The average elytral width and length among these groups was 2.16 and 4.98 mm, respectively (Table 5). Significant differences were observed among original populations, crosses and backcrosses in the standardized female body size ( $F = 7.96$ ;  $df = 4, 51$ ;  $P = 0.001$ ) but not in the standardized male body size ( $F = 0.42$ ;  $df = 4, 45$ ;  $P = 0.79$ ).

TABLE 1. DEVELOPMENTAL TIME (DAYS;  $X \pm SE$ ) OF *HIPPODAMIA CONVERGENS* FROM HONDURAS, HONDURAS  $\times$  IOWA CROSSES AND RECIPROCAL BACKCROSSES; REARED ON 2-3 APHIDS PER DAY, 24°C, L:D 16:8.

|                 | N <sup>c</sup> | Days $\pm$ SE <sup>a, b</sup> |               |               |               |                  |                 |                  |
|-----------------|----------------|-------------------------------|---------------|---------------|---------------|------------------|-----------------|------------------|
|                 |                | Egg                           | Instar I      | Instar II     | Instar III    | Instar IV        | Pupa            | Egg-Adult        |
| Honduras        | 89 (6)         | 3.9 $\pm$ 0.2                 | 3.7 $\pm$ 0.5 | 2.6 $\pm$ 0.3 | 2.9 $\pm$ 0.5 | 6.9 $\pm$ 0.5 a  | 7.0 $\pm$ 0.8 a | 26.9 $\pm$ 1.4 a |
| Iowa            | 236 (8)        | 3.5 $\pm$ 0.2                 | 3.2 $\pm$ 0.1 | 2.1 $\pm$ 0.1 | 2.5 $\pm$ 0.1 | 6.8 $\pm$ 0.3 a  | 5.3 $\pm$ 0.1 b | 23.1 $\pm$ 0.6 b |
| Hon $\times$ IA | 424 (36)       | 3.6 $\pm$ 0.1                 | 3.2 $\pm$ 0.1 | 2.4 $\pm$ 0.1 | 2.6 $\pm$ 0.1 | 5.9 $\pm$ 0.1 b  | 5.6 $\pm$ 0.1 b | 23.4 $\pm$ 0.2 b |
| F1 $\times$ Hon | 160 (12)       | 3.5 $\pm$ 0.2                 | 3.6 $\pm$ 0.3 | 2.6 $\pm$ 0.2 | 2.8 $\pm$ 0.1 | 6.2 $\pm$ 0.2 ab | 5.6 $\pm$ 0.3 b | 24.3 $\pm$ 0.4 b |
| F1 $\times$ IA  | 87 (9)         | 3.7 $\pm$ 0.1                 | 3.4 $\pm$ 0.3 | 2.3 $\pm$ 0.2 | 2.9 $\pm$ 0.2 | 6.1 $\pm$ 0.3 b  | 5.4 $\pm$ 0.1 b | 23.7 $\pm$ 0.4 b |
|                 | (F; df)        | (1.2; 4, 64)                  | (1.4; 4, 64)  | (1.1; 4, 64)  | (1.5; 4, 64)  | (3.9; 4, 64)     | (3.8; 4, 64)    | (6.9; 4, 64)     |
|                 | (P)            | (0.304)                       | (0.246)       | (0.344)       | (0.210)       | (0.007)          | (0.008)         | (0.001)          |

<sup>a</sup>Values represent means of mating pairs.

<sup>b</sup>Means followed by the same letter in a column are not statistically different ( $P > 0.05$ ).

<sup>c</sup>Number of individuals (number of pairs).

TABLE 2. DEVELOPMENTAL TIME, SURVIVAL, AND ADULT CHARACTERISTICS OF THREE POPULATIONS OF *HIPPODAMIA CONVERGENS* REARED ON THREE LEVELS OF *ACYRTHOSIPHON PISUM*; 24°C; 16:8 L:D; 1997.

| Population | <i>A. pisum</i> /day | Dev. time<br>Days; X ± SD | Survival <sup>a</sup><br>% | Sex ratio<br>F: M | Female<br>weight (mg) | Male<br>weight (mg) |
|------------|----------------------|---------------------------|----------------------------|-------------------|-----------------------|---------------------|
| Honduras   | 2                    | 27.3 ± 3.2                | 52 [13]                    | 5:8               | 8.7 ± 2.3             | 8.0 ± 0.4           |
|            | 3                    | 23.4 ± 2.0                | 56 [14]                    | 8:6               | 8.5 ± 1.6             | 8.9 ± 0.9           |
|            | >20                  | 16.8 ± 1.0                | 84 [21]                    | 15:6              | 24.8 ± 3.6            | 19.3 ± 2.6          |
| Iowa       | 2                    | 24.6 ± 1.2                | 48 [12]                    | 3:9               | 8.2 ± 0.7             | 6.9 ± 0.4           |
|            | 3                    | 21.4 ± 2.2                | 56 [14]                    | 5:9               | 10.1 ± 0.6            | 8.3 ± 0.6           |
|            | >20                  | 16.9 ± 0.6                | 72 [18]                    | 11:7              | 21.7 ± 2.5            | 17.9 ± 1.5          |
| California | 2                    | 21.7 ± 1.1                | 28 [7]                     | 3:4               | 8.4 ± 1.5             | 8.3 ± 0.8           |
|            | 3                    | 19.5 ± 1.1                | 44 [11]                    | 6:5               | 10.7 ± 1.3            | 9.1 ± 1.1           |
|            | >20                  | 15.7 ± 1.0                | 28 [7]                     | 5:2               | 25.6 ± 3.0            | 21.4 ± 4.0          |

<sup>a</sup>Numbers in square parentheses = number of *H. convergens* that completed development; 25 first instars started on each aphid diet.

In 1997, sex ( $F = 23$ ;  $df = 1,101$ ;  $P = 0.009$ ), levels of aphid prey provided to the larvae ( $F = 412.3$ ;  $df = 2,101$ ;  $P = 0.001$ ) and the interaction between diet and sex ( $F = 7.83$ ;  $df = 3,101$ ;  $P = 0.001$ ) affected weight of adult *H. convergens*, but no differences among populations were observed ( $F = 4.89$ ;  $df = 2,101$ ;  $P = 0.084$ ) (Table 2). A positive correlation was observed between levels of *A. pisum* provided to larvae and adult weight of Iowa ( $R^2 = 0.91$ ), California ( $R^2 = 0.92$ ) and Honduras ( $R^2 = 0.87$ ) *H. convergens*.

#### Genetic Diversity

Of 31 putative allozyme loci resolved on acrylamide gels, 22 were polymorphic (71%). Heterozygosity (diversity) averaged over all loci was  $21.3 \pm 4.2\%$ ; an average  $2.9 \pm 0.3$  alleles per locus was observed (Table 6). The heterozygosity of only polymorphic loci was  $30 \pm 4.8\%$  with  $3.6 \pm 1.3$  alleles. The distribution of single locus heterozygosities (Fig. 1) shows high levels of diversity and is

consistent with the neutral theory of mutations (Nei et al. 1976). Examination of F1 Honduran × Iowa hybrids at 13 polymorphic loci showed no alleles not detected in North American beetles.

#### DISCUSSION

Response to aphid prey levels was similar among populations, even though *A. pisum* has not been reported from Honduras and therefore may not be a common prey species there (Castro 1993). Total developmental time was inversely correlated with the number of aphid prey provided to *H. convergens* larvae. A reduction of more than 4 days in the total developmental time was observed when aphid prey was increased from 3 to > 20 *A. pisum* per day. Similarly, a reduction in total developmental time was observed for the hemipteran predator *Podisus maculiventris* (Say) when fed greater quantities of Mexican bean beetle larvae, *Epilachna varivestis* Mulsant (Legaspi and O'Neil 1994).

TABLE 3. PERCENTAGE SURVIVAL (X ± SE) FOR LIFE STAGES OF *HIPPODAMIA CONVERGENS* FROM HONDURAS, IOWA, HONDURAS × IOWA CROSSES AND RECIPROCAL BACKCROSSES.

|          | Survival (% ± SE) <sup>a</sup> |               |               |               |               |               |
|----------|--------------------------------|---------------|---------------|---------------|---------------|---------------|
|          | Instar I                       | Instar II     | Instar III    | Instar IV     | Pupa          | Preimaginal   |
| Honduras | 90.0 ± 4.5 ab                  | 97.0 ± 1.6    | 96.2 ± 3.3    | 97.2 ± 2.3    | 95.0 ± 5.0    | 78.1 ± 8.1    |
| Iowa     | 98.6 ± 0.7 b                   | 96.9 ± 1.6    | 98.4 ± 0.9    | 97.9 ± 0.9    | 96.6 ± 1.3    | 88.9 ± 2.1    |
| Hon × IA | 95.1 ± 1.4 b                   | 97.1 ± 0.9    | 99.5 ± 0.4    | 97.3 ± 1.0    | 98.9 ± 0.6    | 88.3 ± 1.7    |
| F1 × Hon | 87.7 ± 3.9 a                   | 94.4 ± 3.1    | 98.3 ± 1.7    | 97.6 ± 2.4    | 94.8 ± 2.8    | 76.7 ± 6.4    |
| F1 × IA  | 92.2 ± 2.8 ab                  | 98.8 ± 1.1    | 96.5 ± 2.4    | 99.1 ± 0.9    | 98.6 ± 1.4    | 85.7 ± 2.3    |
| (F; df)  | (2.54; 4, 64)                  | (0.47; 4, 64) | (1.30; 4, 64) | (0.36; 4, 64) | (1.43; 4, 64) | (1.32; 4, 64) |
| (P)      | (0.04)                         | (0.75)        | (0.28)        | (0.83)        | (0.23)        | (0.27)        |

<sup>a</sup>Means followed by the same letter in a column are not statistically different ( $P > 0.05$ ).

TABLE 4. MEAN FECUNDITY AND PERCENTAGE FERTILITY OF *HIPPODAMIA CONVERGENS* EGGS FROM HONDURAS AND IOWA, THEIR OFFSPRING (HONDURAS × IOWA) AND RECIPROCAL BACKCROSSES.

| Group      | N <sup>a</sup> | Fecundity           |      |      | Fertility (%)       |      |      |
|------------|----------------|---------------------|------|------|---------------------|------|------|
|            |                | (X ± SE)            | min. | max. | (X ± SE)            | min. | max  |
| Honduras   | 6              | 119.0 ± 32.8        | 59   | 172  | 73.7 ± 1.5          | 70.1 | 76.3 |
| Iowa       | 8              | 120.7 ± 56.0        | 24   | 218  | 84.7 ± 4.7          | 79.2 | 94.0 |
| Hon × IA   | 35             | 111.8 ± 8.8         | 7    | 225  | 72.0 ± 3.3          | 4.4  | 93.5 |
| F1 × Hon   | 12             | 119.8 ± 10.8        | 23   | 208  | 84.8 ± 0.2          | 66.1 | 95.5 |
| F1 × IA    | 12             | 104.8 ± 10.0        | 31   | 188  | 76.0 ± 3.6          | 34.0 | 97.2 |
| (F; df; P) |                | (0.11; 4, 55; 0.97) |      |      | (1.58; 4, 55; 0.24) |      |      |

<sup>a</sup>Number of females.

Total developmental times of *H. convergens* from Iowa, California, and Honduras fed > 20 *A. pisum* per day were approximately 3 days shorter than *H. convergens* from Arizona, Oregon and New York (20 days) reared at similar temperatures (Miller 1992, Obrycki & Tauber 1982). Hagen and Sluss (1966) showed that developmental time and life span of *H. convergens* from California were influenced by prey species. Thus, these observed differences might be due to the use of different aphid species among studies. Butler and Dickerson (1972) reared *H. convergens* on the cotton aphid, *Aphis gossypii* Glover, and *A. pisum*, whereas Miller (1992) used the Russian wheat aphid, *Diuraphis noxia* (Mordvilko), and the oat-bird cherry aphid, *Rhopalosiphum padi* (L). Differences in developmental time between beetles fed > 20 *A. pisum* per day in our study and those reared by Obrycki & Tauber (1982) on *A. pisum* may be due to geographical variation.

The size of *H. convergens* crosses and backcrosses are within the ranges described by Gordon

(1985). Weight of adult *H. convergens* was highly correlated to levels of *A. pisum* provided to larvae. When the prey provided was increased from 3 to >20 *A. pisum* per day, adult weights doubled.

*Hippodamia convergens* from Iowa, Honduras, and California mated and exhibited similar fecundity and fertility. The number of eggs produced per day by *H. convergens* inter-population crosses (14 to 17 eggs per day) was slightly lower than that observed by Hagen & Sluss (1966) for California *H. convergens* (20 eggs per day). The fecundity of *H. convergens* was higher in our study than that observed by Wipperfurth et al. (1987), who fed beetles fewer aphids than they could consume on a daily basis.

High levels of gene diversity have been detected in several species of ladybirds (Krafsur & Obrycki 1996; Krafsur et al. 1992, 1995, 1996a, b, 1997). Of the 11 coccinellid species examined, only one, *Coleomegilla maculata* Degeer, shows evidence of being a species complex (Munyanza & Obrycki 1998; Krafsur & Obrycki 2000). The

TABLE 5. SIZE OF PRONOTUM AND ELYTRA OF ADULT *HIPPODAMIA CONVERGENS* FROM HONDURAS AND IOWA, THEIR OFFSPRING (HONDURAS × IOWA) AND RECIPROCAL BACKCROSSES.

| Group    | Sex (N) | Pronotum (mm; X ± SD) |             | Elytra (mm; X ± SD) |             |
|----------|---------|-----------------------|-------------|---------------------|-------------|
|          |         | Width                 | Length      | Width               | Length      |
| Honduras | F (5)   | 2.88 ± 0.12           | 1.42 ± 0.06 | 2.46 ± 0.18         | 5.30 ± 0.18 |
|          | M (4)   | 2.72 ± 0.12           | 1.28 ± 0.05 | 2.22 ± 0.12         | 4.95 ± 0.30 |
| Iowa     | F (5)   | 2.65 ± 0.20           | 1.34 ± 0.11 | 2.26 ± 0.17         | 5.01 ± 0.27 |
|          | M (5)   | 2.79 ± 0.12           | 1.33 ± 0.03 | 2.34 ± 0.02         | 4.79 ± 0.21 |
| Hon × IA | F (29)  | 2.68 ± 0.22           | 1.26 ± 0.10 | 2.13 ± 0.15         | 5.01 ± 0.45 |
|          | M (30)  | 2.63 ± 0.25           | 1.26 ± 0.13 | 2.08 ± 0.26         | 4.91 ± 0.47 |
| F1 × Hon | F (11)  | 2.64 ± 0.10           | 1.29 ± 0.10 | 2.12 ± 0.16         | 4.89 ± 0.37 |
|          | M (9)   | 2.90 ± 0.12           | 1.31 ± 0.11 | 2.38 ± 0.16         | 5.27 ± 0.27 |
| F1 × IA  | F (9)   | 2.49 ± 0.06           | 1.15 ± 0.11 | 2.04 ± 0.10         | 4.76 ± 0.14 |
|          | M (5)   | 2.74 ± 0.09           | 1.21 ± 0.08 | 2.23 ± 0.10         | 5.06 ± 0.11 |

TABLE 6. GENE DIVERSITY  $H_e$  AT PUTATIVE ALLOZYME LOCI IN *HIPPODAMIA CONVERGENS*.

| Enzyme                                   | Locus        | E.C. number system | Buffer  | Expected heterozygosity <sup>a</sup> $h_e$ |
|--|--------------|--------------------|---------|--|
| Acid phosphatase                         | <i>AcpH</i>  | EC 3.1.3.2         | NAM     | 0.539                                      |
| Aconitase                                | <i>Aco</i>   | EC 4.2.1.3         | OD      | 0.502                                      |
| Aldehyde oxidase                         | <i>Aox</i>   | EC 2.6.1.1.        | NAM     | 0.623                                      |
| Adenylate kinase                         | <i>Adk-1</i> | EC 2.7.4.3.        | NAM     | 0.454                                      |
|  | <i>Adk-2</i> |                    |         | 0  |
| Arginine kinase                          | <i>Argk</i>  | EC 2.7.3.3.        | NAM     | 0  |
| Diaphorase                               | <i>Dia-1</i> | EC 1.6.2.2.        | NAM     | 0  |
|  | <i>Dia-2</i> |                    |         | 0  |
| Esterase                                 | <i>Est</i>   | EC 3.1.1.-         | NAM     |  |
| Fructose biphosphatase                   | <i>Fbp</i>   | EC 3.1.3.11.       | NAM     | 0.237                                      |
| Fumarate hydratase                       | <i>Fum</i>   | EC 4.2.1.2.        | OD      | 0  |
| Glucose-6-phosphate dehydrogenase        | <i>G6pd</i>  | EC 1.1.1.49        | NAM     | 0.418                                      |
| Glyceraldehyde-3-phosphate dehydrogenase | <i>G3pd</i>  | EC 1.2.1.12        | NAM     | 0.036                                      |
| Glycerophosphate dehydrogenase           | <i>Gpd</i>   | EC 1.1.1.8.        | TBE     | 0  |
| Hexokinase                               | <i>Hk</i>    | EC 2.7.1.1.        | TBE     | 0.530                                      |
| Hydroxy acid dehydrogenase               | <i>Had-1</i> | EC 1.1.1.30        | NAM, OD | 0.311                                      |
|  | <i>Had-2</i> |                    |         | 0.234                                      |
| Isocitrate dehydrogenase-1               | <i>Idh-1</i> | EC 1.1.1.42        | NAM     | 0.036                                      |
| Isocitrate dehydrogenase-2               | <i>Idh-2</i> |                    | NAM     | 0.053                                      |
| Malate dehydrogenase                     | <i>Mdh-1</i> | EC 1.1.1.37        | NAM, OD | 0.102                                      |
|  | <i>Mdh-2</i> |                    |         | 0  |
| Malic enzyme                             | <i>Me-1</i>  | EC 1.1.1.40        | OD      | 0  |
| Mannose-6-P-dehydrogenase                | <i>Mpi</i>   | EC 5.3.1.8         | NAM     | 0.114                                      |
| Phosphoglucoisomerase                    | <i>Pgi</i>   | EC 5.3.1.9         | OD      | 0  |
| Phosphoglucomutase                       | <i>Pgm</i>   | EC 5.4.2.2         | NAM, OD | 0.201                                      |
| 6-Phosphogluconate dehydrogenase         | <i>6pgd</i>  | EC 1.1.1.44        | NAM     | 0.297                                      |
| Sorbitol dehydrogenase                   | <i>Sdh</i>   | EC 1.1.1.14        | NAM     | 0.610                                      |
| Superoxide dismutase                     | <i>Sod-1</i> | EC 1.15.1.1        | OD      | 0.018                                      |
|  | <i>Sod-2</i> |                    |         | 0.036                                      |
| Trehalase                                | <i>Tre</i>   | EC 3.2.1.28        | NAM     | 0.683                                      |
| Triose-phosphate isomerase               | <i>Tpi</i>   | EC 5.3.1.1         | NAM     | 0.086                                      |

Mean of polymorphic loci:  $H_e = 0.301$ ; SD = 0.048

Mean of all loci ( $n = 31$ ):  $H_e = 0.213$ ; SD = 0.042

<sup>a</sup>Expected proportions heterozygous when mating is random.

high levels of variation characteristic of ladybirds is indicative of large population sizes and high rates of gene flow, inferences supported by ecological and genetic studies. High rates of gene flow in *H. convergens* argue against the notion that disruption of co-adapted gene complexes (supergenes) will cause local populations to decline dramatically (see discussion in Dobzhansky & Pavlovsky 1960). It remains to be determined if supergenes exist in ladybirds, and if alternative gene arrangements are lethal, or if a lower frequency of supergene carriers somehow causes populations to decline. Colonizing species such as ladybirds must naturally accommodate alternative intervals of local inbreeding and invasion by foreign genotypes throughout much of their evolutionary history. Environmental influences, rather than genetic, most likely explain the failure of deliberate ladybird introductions to become established.

All inter-population *H. convergens* crosses in our studies produced fertile eggs. Thus, no intrinsic reproductive barriers exist among Iowa, Honduras, and California populations. If *H. convergens* from California are augmentatively released in Honduras or Iowa, individuals or their progeny may interbreed. Our results indicate that if *H. convergens* from California do cross with local populations, no detrimental effects in F1 developmental and reproductive parameters in response to one species of aphid prey (*A. pisum*) may occur. Predation of other prey species of local importance may need to be examined (Bogran & Obrycki 1998). However, this does not mean that the F1 *H. convergens* crosses would be well suited to local conditions. For example, photoperiodic responses for diapause induction may be different among California and local populations of *H. convergens*, and this parameter could be altered in the F1 crosses (see Tauber et al. 1997). The observed interbreed-

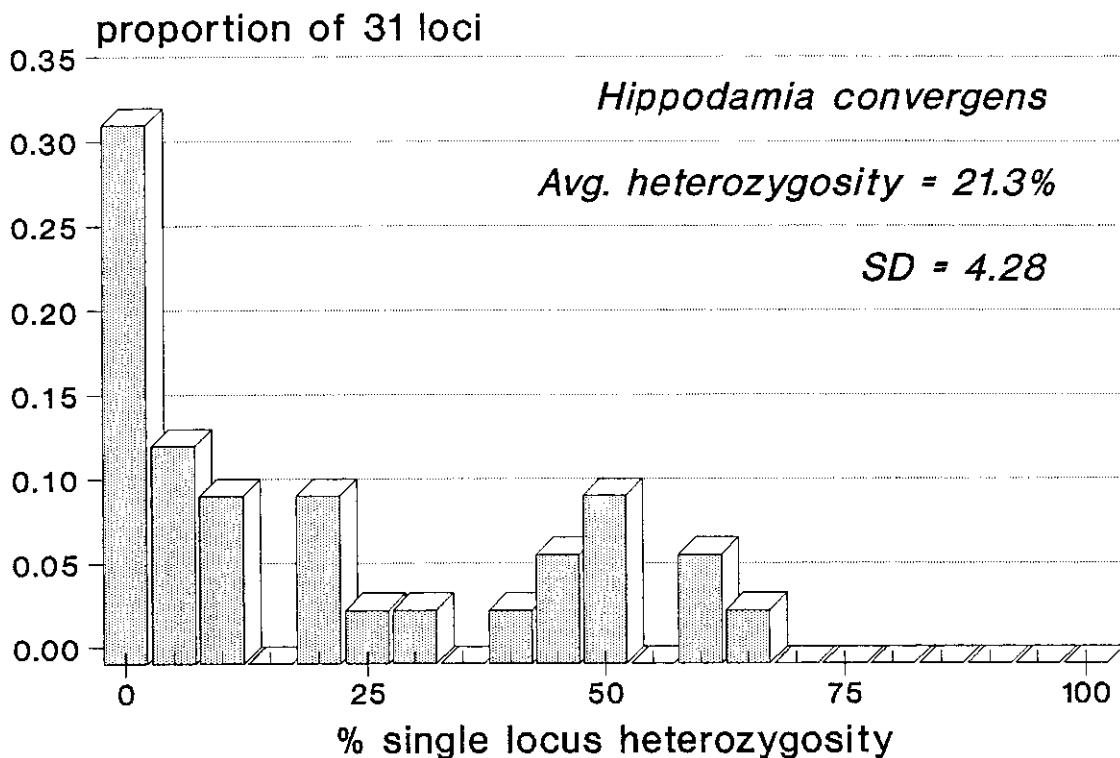


Fig. 1. Single locus heterozygosity ( $h_s$ ) in *Hippodamia convergens*.

ing among California, Honduras, and Iowa beetles, in addition to the presence of parasitoids and pathogens in California adults, combined with the lack of substantial evidence of effectiveness, suggest that the practice of augmentative releases of field collected *H. convergens* needs to be carefully examined for non-target effects.

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## EFFECT OF PARASITOID RELEASE PATTERN ON WHITEFLY (HOMOPTERA: ALEYRODIDAE) CONTROL IN COMMERCIAL POINSETTIA

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### ABSTRACT

Under commercial poinsettia production conditions we compared two patterns of parasitoid release for the aphelinid whitefly parasitoid *Eretmocerus eremicus* Rose and Zolnerowich. We compared the currently used pattern of a fixed weekly release number (3 females per plant per week) to an experimental pattern in which more parasitoids were released early in the crop (wks 1-8), followed by a lower number (wks 9-17), with the seasonal release average still being 3 female parasitoids per plant per week. We further compared the outcome of these two treatments (fixed and variable) to a low release rate (1 parasitoid per pl per wk) of *Encarsia formosa* Gahan, an aphelinid parasitoid widely used for whitefly control in greenhouse crops. In control cages without parasitoid releases, whitefly nymphal densities reached 15-32 live nymphs per leaf, which was 7 to 16-fold greater than the acceptable level at crop harvest. In cages in which parasitoid releases were made, whitefly nymphal densities were suppressed 99.8%, 96.8% and 50.9% by fixed-rate *E. eremicus*, variable-rate *E. eremicus*, and low-rate *E. formosa* treatments, respectively. In greenhouse populations, the final densities of live whitefly nymphs per leaf were significantly higher in the *E. formosa* treatment than the two *E. eremicus* treatments. Releases of low numbers of *E. formosa* provided commercially acceptable control in only one of two greenhouses. There was no difference between the fixed and variable release rate treatments of *E. eremicus*, indicating that whitefly suppression was not increased by concentrating the release of this parasitoid early in the crop.

**Key Words:** *Eretmocerus eremicus*, *Encarsia formosa*, *Bemisia argentifolii*, poinsettia, biological control, variable release rate, augmentative release, evaluation, cost, greenhouses

### RESUMEN

Dos patrones de liberación de parásito para *Eretmocerus eremicus* Rose y Zolnerowich fueron comparados bajo condiciones de producción comercial de poinsettia (Flor de Pascua). Comparamos el patrón actualmente usado de número de liberación semanal fija (3 hembras por planta por semana) a un patrón experimental en el cual más parásitos fueron liberados temprano en el cultivo (semanas 1-8), seguido por un número menor (semanas 9-17), con la liberación estacional promedio aun siendo 3 parásitos hembras por planta por semana. Adicionalmente comparamos el resultado de estos dos tratamientos (fijo y variable) a una baja incidencia de liberación (1 parásito por planta por semana) de *Encarsia formosa* Gahan, un parásito usado extensamente para control de la mosca blanca en cultivos de invernadero. En jaulas de control sin liberación de parásito, densidades de ninfas mosca blanca alcanzaron 15-32 ninfas vivas por hoja, que fue entre 7 y 16 veces mayor que el nivel aceptable al momento de cosecha del cultivo. En jaulas en las cuales liberación de parásito ocurrió, las densidades de ninfas mosca blanca fueron suprimidas 99.8%, 96.8, y 50.9% por *E. eremicus* en tratamientos fijo, variable, y tratamientos de baja incidencia de *E. formosa*, respectivamente. En poblaciones de invernadero, las densidades finales de ninfas vivas de mosca blanca por hoja fueron significativamente mayores en el tratamiento de *E. formosa* que en los dos tratamientos de *E. eremicus*. Liberaciones de bajas cantidades de *E. formosa* proporcionaron control aceptable en solo uno de dos invernaderos. No hubo diferencia entre los tratamientos fijo y variable de *E. eremicus*, indicando que supresión de mosca blanca no incremento como causa de concentrar la liberación de este parásito temprano en el cultivo.

Silverleaf whitefly (*Bemisia argentifolii* Bellows and Perring) (Homoptera: Aleyrodidae) is an important foliar pest of poinsettia (*Euphorbia pulcherrima* Willd. ex Koltz.) (Byrne et al. 1990; Bellows et al. 1994; Hoddle & Van Driesche 1996).

The principal parasitoid species used for its control have been *Encarsia formosa* Gahan and *Eretmocerus eremicus* Rose and Zolnerowich (Hymenoptera: Aphelinidae) (Drost et al. 1996, Hoddle & Van Driesche 1996; Rose & Zolnerowich 1997).

Trials to measure the efficacy of releases of commercial *E. formosa* (*Ef*), the Beltsville strain of *E. formosa* (*Ef*Belt), or *E. eremicus* (*Ee*) on poinsettia have examined constant weekly releases of either one or three females per plant per week (Hoddle & Van Driesche 1996; Hoddle et al. 1996; Hoddle et al. 1997abc; Hoddle & Van Driesche 1999ab). In small greenhouses holding 90 plants, whitefly mortality (1- survivorship from egg to adult) was 99% (*Ee* and *Bf*Belt, both high rate), 96% (*Ef*Belt, low rate), 95% (*Ef*, low rate), 92% (*Ef* high rate) and 88% (*Ee* low rate) (Hoddle et al. 1997abc).

While these mortality rates may seem uniformly high, the differences among them have practical importance because whiteflies in poinsettia have four generations in the crop cycle, each with high rates of population increase. With no mortality from pesticides or parasitoids, silverleaf whitefly egg-to-adult survival is about 75% (Hoddle et al. 1997abc). Combined with the per female fertility rates that *B. argentifolii* can achieve on poinsettia, this level of survival results in rates of increase of up to 25-fold per generation. With this rate of increase and an initial density on cuttings in the range of 0.1 nymphs per leaf (a typical value), an uncontrolled population has the potential to exceed 12,000 per leaf by the end of the crop (but actually would be constrained below that level by competition, other sources of mortality, and space on the leaf). By comparison, if parasitoid-caused mortality decreased egg-to-adult survival to 1% (99% mortality), whitefly density would decrease over the course of the crop to less than 0.0004 per leaf. Lower levels of parasitoid caused mortality are progressively less effective; 88% mortality, for example, would allow the whitefly population to reach 8.1 nymphs per leaf, an unacceptably high level. Thus small differences in parasitoid-caused mortality in the range observed (88-99%) are critically important in the success or failure of whitefly biological control in commercial crops.

Because differences in mortality rates interact with variation in realized fecundity and sex ratio in ways that cannot be easily predicted, to actually know how well a given parasitoid release rate or pattern works in limiting final whitefly densities, tests must be rigorously conducted under commercial conditions where this technology will be ultimately utilized. In both summer stock plants and fall Christmas crop plants, *E. eremicus* at 3 females per plant per week effectively suppressed silverleaf whitefly (Hoddle & Van Driesche 1999ab). Poinsettias at the end of the crop were acceptable to growers as a source of cuttings or, for the Christmas crop, for sale to retailers (with fewer than 2 live nymphs and pupae per leaf). This commercially acceptable level of control was not achieved with the same release rates of *E. formosa* Beltsville. This release rate, however, was too expensive for grower adoption. Con-

sequently, we chose to investigate whether or not the pattern of parasitoid releases, independent of number released, might be manipulated in ways that increased the level of parasitoid impact on whitefly populations.

We hypothesized that release patterns that concentrated parasitoid releases either in the early or late part of the crop cycle might be more effective for controlling *B. argentifolii*. In small greenhouses, we tested two variable-rate release patterns against a fixed weekly release rate of 3 females per plant. In pattern 1 ("low-high"), parasitoids were released at a low rate in the first half of the crop and then the number was increased once plants were larger (first 1, then 5 parasitoids per plant per week) and in pattern 2 ("high-low") this pattern was reversed (first 5 parasitoids per plant, then 1 in the second half of the crop cycle), concentrating highest parasitoid numbers on the smallest plants, early in the crop.

The argument for potential greater efficacy of release pattern 1 (low-high) was that, since parasitoid foraging efficiency declines as plant size increases (Hoddle et al. 1998), increasing parasitoid release rate in the later part of the crop when plants are largest might compensate for this decline in per parasitoid efficacy. Also, at lower release rates progeny production by *E. eremicus* within the greenhouse increases because fewer parasitized hosts die from multiple ovipositions and host feeding (see Hoddle 2000 for a review of this argument). The argument for greater efficacy of release pattern 2 (high-low) was that using higher release rates when plants were small might virtually exterminate the whitefly population, leaving too little time before crop sale for whiteflies to recover to damaging levels.

In fall 1995, we ran a trial with *E. eremicus* in small greenhouses (holding 90 plants) (Hoddle et al. 1999) to test the pest control value of these variable release patterns. We found that pattern 2 (high-low) resulted in 75% fewer live whitefly nymphs and pupae per plant at the end of the crop (week 14) than did pattern 1 (low-high) (Hoddle et al. 1999).

The main goal of the study presented here was to directly compare, under fully realistic production conditions, the better of these two variable *E. eremicus* release patterns (high-low) directly against the fixed released pattern. By design these two treatments have the same release rate of *E. eremicus*, allowing us to isolate any effects due to the single factor of parasitoid release pattern. We also took advantage of available greenhouses in this same trial to pursue a second goal, to assess the efficacy of a low release rate (one parasitoid per plant per week) of *E. formosa*. We did this because (1) the low rate of this species did perform reasonably well (causing 95% mortality) in earlier small greenhouse trials; (2) published work indicated that this parasitoid when used at



low rates might actually be more, not less, effective than high rates due to the effects of mutual interference among parasitoids (Hoddle et al. 1997a; Hoddle 2000); and (3) producers recommend use of low release rates of the species in European flower crops, but few trials in North America exist to support this recommendation.

## MATERIALS AND METHODS

### Study Site and Experimental Design

The study was conducted in commercial greenhouses in western Massachusetts (Fairview Farms, Whately, MA), for seventeen weeks between 15 August and 6 December, 1996. Six plastic hoop houses (each identical in size and construction, with dimensions of  $4.8 \times 29.3$  m) were used for the principal treatments.

Three treatments were examined, each randomly assigned to a whole greenhouse with two replications: (1) a fixed release rate of three females of *E. eremicus* per plant per week; (2) a variable release of *E. eremicus*, with five females released per plant per week for the first eight weeks of the trial (August 16–October 4) and one female per plant per week for the last nine weeks (October 11 to December 6); and (3) the commercial strain of *E. formosa* released at one female per plant per week. A seventh greenhouse, in which the grower used pesticides for whitefly management, was also examined 12 times between 26 August to 18 November to provide a further comparison to whitefly suppression levels seen in the biological control treatments.

In each test greenhouse (except for the chemical control greenhouse), two cages (95  $\mu$ m mesh over PVC frames with dimensions  $153 \times 92 \times 117$  cm) were used to isolate five pots, each with three poinsettia plants, as controls. One cage in each greenhouse (designated “control for treatment”) received no treatments of any kind to suppress whiteflies. The other cage (designated “control for caging effect”) received the same parasitoid treatment as the greenhouse in which it was placed. Initial densities of whitefly nymphs and pupae in cages were manipulated to match those in test greenhouses (see below).

### Crop Composition and Management

In each greenhouse, a poinsettia crop was established on August 15, 1996 using 1500 plants, all ‘Freedom’ varieties (1140 red, 200 white and 160 pink per house) from Paul Ecke Ranch (Encinitas, CA). These plants were planted in soilless media, three stems per 20-cm dia pot. Numbers of plants in each house remained constant until November 27, at which time removal of colored plants for Christmas sale began. Numbers of parasitoids released during the last three weeks

of the trial were reduced as needed to keep the parasitoid release rate per plant constant. Plants placed in cages were selected from plants in each test greenhouse at the start of the experiment, choosing plants so that the average density of whitefly nymphs on plants put in cages was the same as that of the whole greenhouse.

All whiteflies found on plants were *B. argentifolii* and were assumed to have entered the greenhouses on leaves of rooted cuttings purchased from suppliers.

All plants in all greenhouses were treated with the fungicide thiophanate methyl+etridiazole (Banrot 40WP®, Scotts Sierra Crop Protection Co., 1411 Scottslawn Rd., Marysville, OH 43041) to control root rot on 18 August and 21 September. Plants in the chemical control greenhouse were treated with imidacloprid (Marathon®) on 12 September. No insecticides were used in any of the six test greenhouses, except in one of the two greenhouses in which *E. formosa* was released. In this single greenhouse, insecticide smokes (Fulex Dithio®, sulfotep, Fuller Systems, Inc., Woburn, MA 01801) were applied on 14 and 27 November to reduce numbers of adult whiteflies before sale.

### Population Sampling

Whitefly densities in each of the six test greenhouses at the start of the trial were determined by examining all the leaves of 100 freshly potted cuttings with a 3.5 $\times$  head-mounted magnifying device. All live whitefly nymphs, pupae or adults were counted. Because initial whitefly densities were extremely low (0.0083 live stages per cutting), cohorts of *B. argentifolii* nymphs were created using a laboratory colony and infested plants were placed in each house to augment the population by an estimated 1.0 nymph per plant (=1500 *B. argentifolii* nymphs added per greenhouse). To achieve this, six infested plants were introduced into each greenhouse. Each infested plant had three leaves on which cohorts of *B. argentifolii* nymphs had been created by using leaf cages to confine groups of 5–6 pairs of adult whiteflies. Each cohort initially consisted of 105 eggs. Previous lifetable estimates (Hoddle et al. 1997abc) suggest that 80% of these eggs would hatch and this method was thus equivalent to adding 1512 nymphs per house. Plants were placed in greenhouses on 19 August and leaf cages were immediately removed from one infested leaf per plant. Leaf cages were removed from the second and third leaves on each infested plant on August 23 and 29. Staggered removal of leaf cages was intended to promote whitefly survival and enhance the establishment of a whitefly population despite ongoing parasitoid releases. In each of the control cages in each test greenhouse, one plant with one infested leaf bearing 19 eggs was added (being the equivalent of 15 nymphs, allowing for

the estimated 80% hatch). Cages thus received the same increase of one nymph per plant as did the whole greenhouses.

Growth of whitefly populations in test greenhouses and cages was measured by counting whiteflies on each of 3 leaves (one lower, one middle and one upper) on each of 90 randomly selected plants each week from each greenhouse. In cages, 8 plants were chosen at random for sampling, resulting in 24 leaves being examined per cage per week. Sampling was nondestructive, with whitefly numbers being counted with the aid a head mounted optical magnifier, as with the counts on cuttings. For each leaf, numbers were recorded of live nymphs, live pupae, dead nymphs, dead pupae, parasitized nymphs, whitefly pupal exuviae, whitefly exuviae bearing parasitoid emergence holes, and adult whiteflies.

#### Parasitoid Sources and Sampling

*Eretmocerus eremicus*. This parasitoid was initially supplied by Beneficial Insectary (Oak Run, CA) and after week 7 by Koppert Biological Systems, Inc. (Berkel en Rodenrijs, The Netherlands, through the North American office in Romulus, MI). *Eretmocerus eremicus* was received as loose parasitized *Trialeurodes vaporariorum* (Westwood) nymphs from the supplier and the number needed for release was calculated by assuming a 60% emergence rate and a 50/50 sex ratio (as determined in other trials, Hoddle & Van Driesche 1999b; Van Driesche et al. 1999). We weighed 10 aliquots of parasitized nymphs to estimate the number of parasitoid pupae in 0.02 g. We then weighed the quantity of parasitized *T. vaporariorum* nymphs needed to treat greenhouses or cages in view of plant number present and desired parasitoid release rate. Actual emergence rates and sex ratios were measured in the course of the trial (see below) and used to calculate the actual release rate achieved.

*Eretmocerus eremicus* was deployed by placing parasitized *T. vaporariorum* nymphs (not glued to cards) in styrofoam release cups (6 cm tall, 5.5 cm wide at bottom, 8.5 cm wide at top), which had the bottoms cut out and replaced with organdy (mesh 0.9  $\mu$ m) to allow for drainage. Cups were attached 10 cm above the canopy to wooden stakes (50 cm long) placed in the potting media. In each biological control greenhouse receiving this parasitoid, there were 15 release cups distributed evenly throughout the crop. Since watering was done via overhead hoses, workers were asked to avoid wetting release cups.

*Encarsia formosa*. This thelytokous parasitoid species was supplied by Applied Bionomics (Sidney, BC, Canada) as parasitized fourth instar nymphs of *T. vaporariorum* glued to release cards, with 200 parasitoids per card. Based on earlier trials and producer's advice, we assumed that 50% of pupae would emerge as adults. Numbers

of cards needed per greenhouse were then calculated using these estimates of numbers per card, the emergence rate, and the number of plants per greenhouse or cage.

*Encarsia formosa* were deployed in greenhouses by hanging the necessary number of manufacturer's release cards (15 per greenhouse) (bearing parasitized *T. vaporariorum* nymphs) on plants throughout the crop.

*Verification of Release Rates*. To verify our assumptions concerning the adult emergence rates for each parasitoid species, percentage emergence of pupae of each parasitoid species was determined weekly throughout the trial. No parasitoids emerged from our shipments before being placed in the test greenhouses. In each greenhouse, emergence cards (for *E. formosa*) or release cups (for *E. eremicus*) were collected and taken back to the laboratory after a one week exposure in the test greenhouses. For each parasitoid species, one hundred and fifty whitefly nymphal cadavers were selected randomly from the material on the release card or in the release cups and the number of successfully emerged parasitoids was determined based on observation of parasitoid emergence holes.

The proportion of emerging adults of *E. eremicus* that were female was determined on eight dates throughout the test. On each date, 100-200 pupae were removed from orders received from suppliers and held in the laboratory in glass vials at 22°C for emergence. Seven to ten days later, one hundred adults were randomly selected and sexed.

Information on emergence rate, sex ratio (*E. eremicus* only), and number of pupae placed in greenhouses was used to calculate the release rate achieved in each greenhouse in each week.

#### Statistical Analysis

The densities of live whitefly nymphs per leaf at harvest were compared among treatments with a nested ANOVA and treatment means separated by use of Tukey's studentized mean separation test (at  $P = 0.05$ ).

## RESULTS

#### Release Rates of Parasitoids and Quality Control

*Eretmocerus eremicus*. Parasitoid emergence, summed over all dates within each greenhouse, varied among greenhouses from 46.1-58.2% and averaged  $53.5\% \pm 1.9$  (SE) overall. The percentage of adult *E. eremicus* emerging from pupae held after receipt in the laboratory that were female ranged among dates from 39 to 58%. The seasonal average, based on 800 parasitoids, was  $48.1\% \pm 2.2$  (SE).

Because the proportion of *E. eremicus* that emerged successfully (53.5%) was less than what we assumed (60%), insufficient pupae were placed in some greenhouses on some dates to achieve the

intended release rate. Actual release rates (females parasitoids per plant per week) averaged 3.8 (replicate one,  $3.1 \pm 0.6$ ; replicate two,  $4.4 \pm 0.6$ ) and 0.8 (replicate one,  $0.8 \pm 0.5$ ; replicate two,  $0.8 \pm 0.7$ ) for the variable release rate treatment and 2.8 (replicate one,  $2.7 \pm 0.2$ ; replicate two,  $2.8 \pm 0.2$ ) for the fixed rate, rather than 5 and 1, and 3 as intended (Fig. 1).

*Encarsia formosa*. The commercial strain of *E. formosa* had seasonal average emergence rates of 44.9 and 44.5% for the two test greenhouses, for a grand seasonal average of  $44.7 \pm 2.5$  (SE). Numbers of pupae per card averaged  $199.6 \pm 7.2$  (SE). Numbers of female parasitoids per plant actually emerging in the greenhouse were  $1.2 \pm 0.1$  (SE) in each test greenhouse.

Whitefly Population Trends in Caged Controls

Whitefly nymphal populations in cages in which no treatments were made increased rapidly in density after mid November in the four greenhouses receiving *E. eremicus* releases (Fig. 2A, B). Whitefly nymphal populations in control cages in the two greenhouses receiving *E. formosa* releases increased in late November, but then declined in December (Fig. 2C). In cages in which *E. eremicus* releases were made (constant and

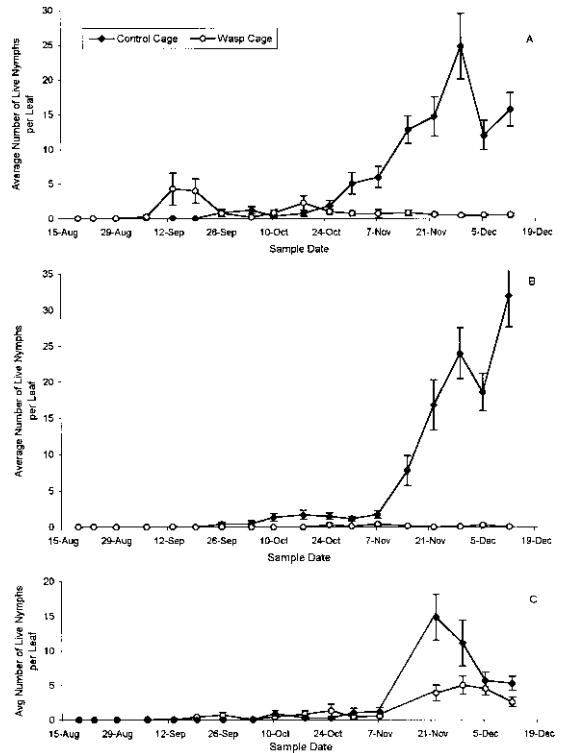


Fig. 2. Trends in average density of whitefly nymphs for control cages (control cage and parasitoid cage) for two greenhouses receiving the variable release rate of *Eretmocerus eremicus* (first 5 females per plant per week, then one female) (A); two greenhouses receiving the fixed rate of *E. eremicus* (3 females per plant per week) (B); two receiving the low rate of *Encarsia formosa* (1 female per plant per week) (C).

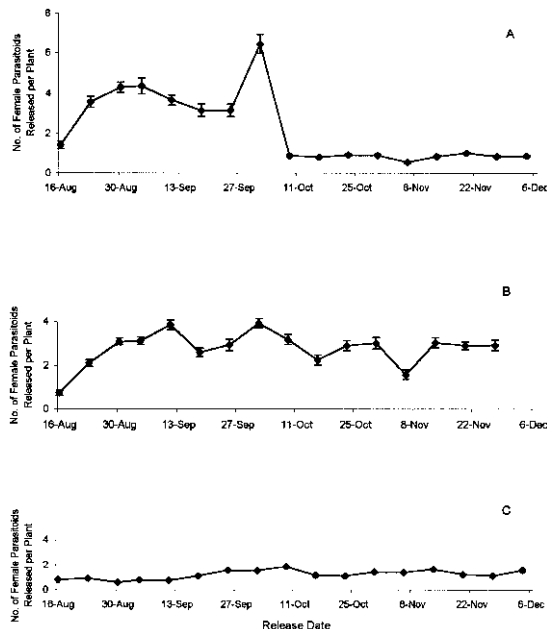


Fig. 1. Average release rates achieved (for two greenhouses each): greenhouses receiving the high-low release rate of *Eretmocerus eremicus* (first 5 females per plant per week, then one female) (A); greenhouses receiving the fixed release rate of *E. eremicus* (3 females per plant per week) (B); greenhouses receiving a low release rate of *E. formosa* (1 female per plant per week) (C).

variable), whitefly nymphal densities remained below 1 nymph per leaf during this same period, reflecting a high level of whitefly suppression (Fig. 2A, B). In cages in which *E. formosa* was introduced, nymphal densities in late November exceeded 4 nymphs per leaf (Fig. 2C). Comparison of whitefly densities at harvest in cages with and without parasitoid releases showed that nymphal densities per leaf in control cages were reduced 99.9% by the fixed release rate treatment of *E. eremicus*, compared to 96.8% for the variable (high-low) release rate treatment of the same species and 50.9% for the low release rate of *E. formosa*.

Whitefly Population and Parasitism Trends in Trial Greenhouses

Whitefly populations outside of cages in test greenhouses were low in all treatments and remained so throughout the trial (Fig. 3A-E). Densities of live nymphs remained below 1 nymph per leaf in all test greenhouses except in one of the two receiving releases of *E. formosa* (Fig. 3D), in

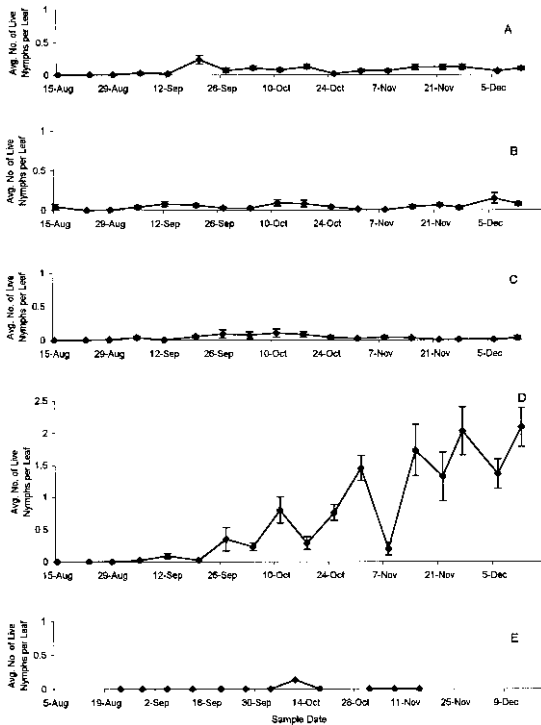


Fig. 3. Trends in density of whitefly nymphs per leaf on plants in greenhouses receiving the variable release rate of *Eretmocerus eremicus* (first 5 females per plant per week, then one female) (A, average of two replicates); the fixed release rate of *E. eremicus* (3 females per plant per week) (B, average of two replicates); the low rate of *Encarsia formosa* (1 female per plant per week) (C, D, replicates different and presented separately); or the chemically-treated greenhouse under grower management (E, one replicate).

which increasing densities exceeded 2 nymphs per leaf at harvest. Pupal and adult whitefly densities in all greenhouses were lower than nymphal densities, but followed similar trends, which are not presented. The trend of quick increase in whitefly nymphal numbers in one *E. formosa* greenhouse (Fig. 3D) prompted the grower to apply insecticidal smokes (Fulex Dithio® [sufotep]) to suppress whitefly adults on plants before sale.

Final densities of live nymphs per leaf were significantly different at harvest among treatments ( $F = 27.89$ ,  $df = 2$ ,  $p = 0.0001$ ) and whitefly nymphal densities in the low rate of *E. formosa* treatment were significantly higher than the two *E. eremicus* treatments, which were statistically similar. Whitefly densities in the chemical control greenhouse were highly suppressed throughout the trial.

Few parasitized whitefly nymphs were encountered in this trial in any of the test greenhouses and we were not able to draw any inferences about the effect of the treatments on

parasitism rates. However, low parasitism rates do not imply that total host mortality caused by parasitoids was low. For these parasitoid species, host feeding is often the major cause of host mortality, especially when parasitoid-to-host ratios are high, as would be the case in an augmentative biological control program when hosts are scarce. The effect of mortality from parasitoid host feeding is reflected in lowered host density, which has been analyzed above.

## DISCUSSION

We had two objectives with this experiment: (1) to determine if variable release rates of *E. eremicus* would be more effective than a fixed release rate of the same total number of parasitoids released over a complete poinsettia cropping period under commercial growing conditions and (2) to ascertain if a low fixed weekly release rate of *E. formosa* would suppress *B. argentifolii* densities on poinsettia to acceptable levels at time of harvest.

In this trial, there were no statistical differences in whitefly nymphal densities at harvest between greenhouses receiving fixed versus variable rate releases of *E. eremicus*. While starting whitefly densities in these greenhouses were very low, whitefly populations in control cages (in which *E. eremicus* was not applied) increased to high levels (25-35 live nymphs per leaf) by harvest. The absence of such increase in the four *E. eremicus*-release greenhouses (and their associated parasitoid-release cages) was thus due to natural enemy activity. Based on (1) the lack of evidence in this trial supporting the idea that a variable rate increases control and (2) lack of any such effect in a previous trial run in small greenhouses (Hoddle et al. 1999), we conclude that the variable release pattern has no discernible advantage over a fixed release rate pattern and do not recommend its use.

*Encarsia formosa* released at one parasitoid per plant per week provided inconsistent whitefly control. One of the two greenhouses receiving this treatment developed an increasing whitefly population that at harvest was still relatively low but was increasing enough that the grower intervened with chemical control measures. Because low rates of *E. formosa* have failed to consistently control *B. argentifolii* on poinsettia, we do not recommend use of this parasitoid on this crop in North America.

This trial demonstrates that tactics other than varying the release rates of *E. eremicus* will be needed to make use of this parasitoid both economical and effective. Insect growth regulators found in the laboratory to be compatible with *E. eremicus* (Hoddle et al. 2000) have enhanced the economic feasibility of using this parasitoid in greenhouses, allowing release rates to be reduced by two thirds (Van Driesche et al. 2000).

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***NEOTERMES PLATYFRONS*, A NEW DAMPWOOD TERMITE  
(ISOPTERA, KALOTERMITIDAE) FROM THE DOMINICAN REPUBLIC**

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ABSTRACT

*Neotermes platyfrons* n. sp. is described from the winged imago and the small and large soldier castes. This species is endemic to Hispaniola and surveys of the Dominican Republic indicate the greatest populations occur in the far-eastern coastal woodlands.

Key Words: new species, taxonomy, West Indies, Greater Antilles, Caribbean

RESUMEN

*Neotermes platyfrons* sp. n., es descrita basándose en los adultos alados y las castas de soldados grande y pequeña. La nueva especie descrita es endémica de la Española y nuestros hallazgos sugieren que las mayores poblaciones se hallan en los bosques costeros del extremo oriental de la Isla.

Twenty-two species of *Neotermes* occur in the Neotropical Region (Constantino 1998). Several *Neotermes* spp. are common elements of the termite fauna of coastal woodland and mangrove habitats in southern Florida and the West Indies (Krecek et al. 2000; Scheffrahn et al. 1994; Scheffrahn et al. 2000). While conducting termite surveys in the Dominican Republic between 1992 and 1996, we collected numerous samples of a *Neotermes* sp. that were preliminarily identified as *N. (=Kalotermites) jouteli* (Banks) (Harris 1955, 1971; Scheffrahn et al. 1994). We now recognize those specimens as a new species of *Neotermes* that is described herein for the first time.

MATERIALS AND METHODS

The description of *N. platyfrons* n. sp. is based on the examination of 27 colony samples, all in authors' collection, taken from 13 localities in the Dominican Republic (Fig. 1). Morphometric data from specimens preserved in 85% ethanol were obtained using a stereomicroscope fitted with an ocular micrometer. Measurements were adopted mainly from Roonwal (1970), while the color scheme of Sands (1965) was used. The terms "small" and "large" soldiers (Krishna 1961) are equal to the terms "short-headed" and "long-headed" soldiers (Banks & Snyder 1920), respectively. These terms are used to separate the two most common size morphs of soldiers occurring within some species of the Kalotermitidae (Nickle & Collins 1989).

Scanning electron micrographs (SEMs) were digitized at 600 dpi, the digital image outline traced using photograph-enhancing software

(Photo Magic, Micrografx, Inc., Richardson, TX), the background converted to black, and the scale bar digitally drawn. Collection localities were mapped (Fig. 1) using ArcView GIS version 3.0a software and relevant map data from Digital Map of the World version 1.0 (Environmental Systems Research Institute, Inc. Redlands, CA).

The holotype alate and a paratype soldier will be deposited in the American Museum of Natural History, New York. Additional paratype alates and soldiers will be deposited in the National Museum of Natural History (Smithsonian Institution), Washington, DC, and in the Florida State Collection of Arthropods, Florida Department of Agriculture and Consumer Services, Division of Plant Industry, Gainesville. Remaining paratypes are maintained in the authors' collection at the University of Florida Research and Education Center, Fort Lauderdale, FL.

*Neotermes platyfrons*, **New Species**

*Kalotermites* near *jouteli* Banks; Harris 1955 (Dominican Republic)

*Neotermes jouteli* (Banks); Harris 1971 (Dominican Republic)

*Neotermes jouteli* (Banks); Scheffrahn et al. 1994 (Dominican Republic records only)

Imago (Fig. 2 A-B, Table 1).

In dorsal view, body almost uniformly ferruginous with exception of darker chestnut brown V-shaped mark on frons and M-shaped mark on vertex. Mandibles chestnut brown at bases, almost

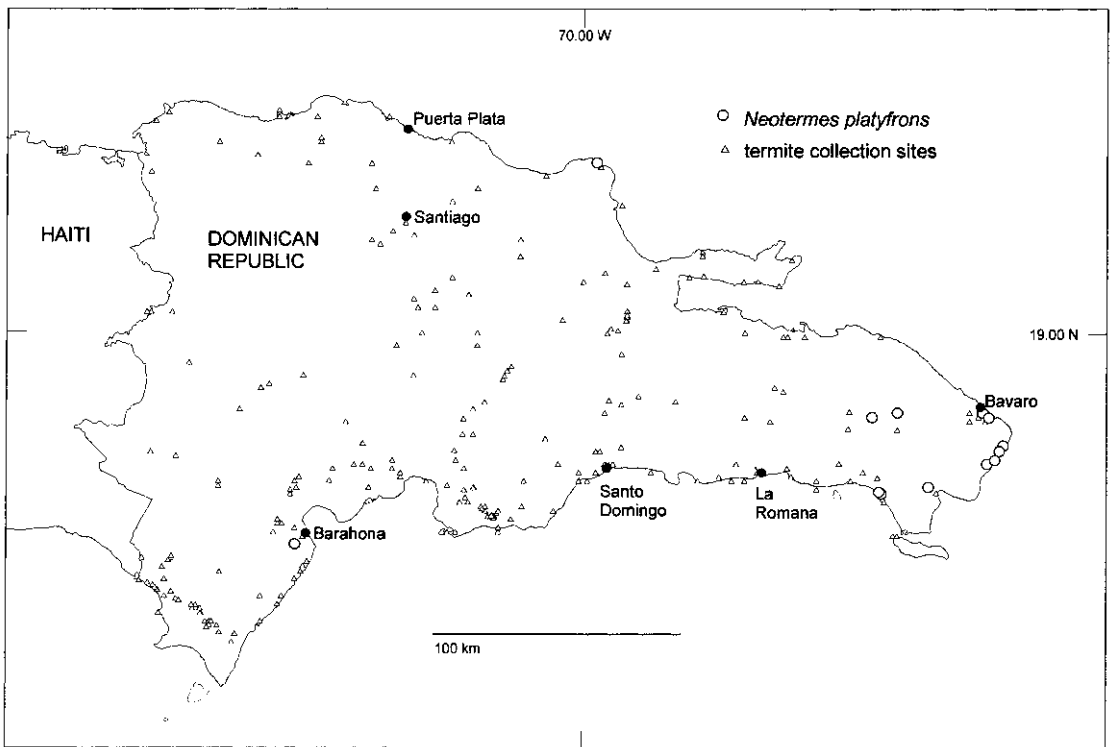


Fig. 1. *Neotermes platyfrons* localities and termite collection sites in the Dominican Republic from 1992-1996.

black distally. Anteclypeus pale yellow-orange. Antennae ferruginous with exception of darker chestnut-brown articles 2-4. Ocelli yellow-white, coloration contrasts conspicuously with head capsule. Pronotum chestnut-brown, periphery slightly darker and concolorous with distinct chevron pattern on wing scales of pterothorax. Femora pale yellow; tibiae and tarsi ferruginous. Arolia whitish.

In dorsal view, head capsule suboval; in ventral view, margins below eyes straight and convergent anteriorly. Frons moderately rugose, flat, and with shallow depression; surrounded on each side by ridges extending from ocelli to dorsal mandibular condyles. Frons rugosity terminates posteriorly as shallow median channel between ocelli. In lateral view, angle of slope between planes of frons and vertex  $\approx 10^\circ$ . Compound eyes large and protruding, subcircular or very slightly subreniform; margins along antennal sockets rectate or very faintly concave. Ocelli moderately large and slightly protruding, obliquely oval, and clearly separated from eyes. Basal striations on mandibles weak or absent. Epicranial depression weak or absent. Head with scattered short and fewer longer ( $\approx 0.2$ -mm-long) erect setae. Pronotum with similar pilosity, but with proportionally more longer setae than short setae along margins. Fore wing scale and proximal portion of fore wing with few short setae. Antennae with 18-20 articles,

usually 19; relative length formula  $2 = 3 > 4 = 5$  or  $2 > 3 > 4 = 5$ . Pronotum robust, with anterior margin deeply concave and peripheral rim elevated; median posterior margin faintly concave, and posterior corners subtruncate. Subcosta of fore wing closely parallels costal margin and joining costal margin at  $\frac{1}{4}$  wing length from suture. Radius of fore wing intersects costal margin near midwing, and radial sector with 4-6 branches; branching commences proximal to midwing. Sclerotized media of fore wing with several very faint diagonal anterior branches intersecting radial sector, and with posterior branches that fade into membrane save one or two that reach wing margin. Wing membrane texture with faint nodulations.

#### Comparisons.

Alates of *Neotermes platyfrons* are intermediate in size between those of the smaller *N. jouteli* and the larger *N. mona* (Banks) with some overlapping measurements among the three species. The most distinguishing range of measurements for *N. jouteli*, *N. platyfrons*, and *N. mona*, respectively, are as follows: head length to postclypeus 1.34-1.54, 1.55-1.68, and 1.63-1.97 mm; head height without postmentum 0.88-1.05, 1.05-1.11, and 1.11-1.21 mm; pronotum maximum length 1.06-1.32, 1.29-1.47, and 1.49-1.68 mm; pronotum

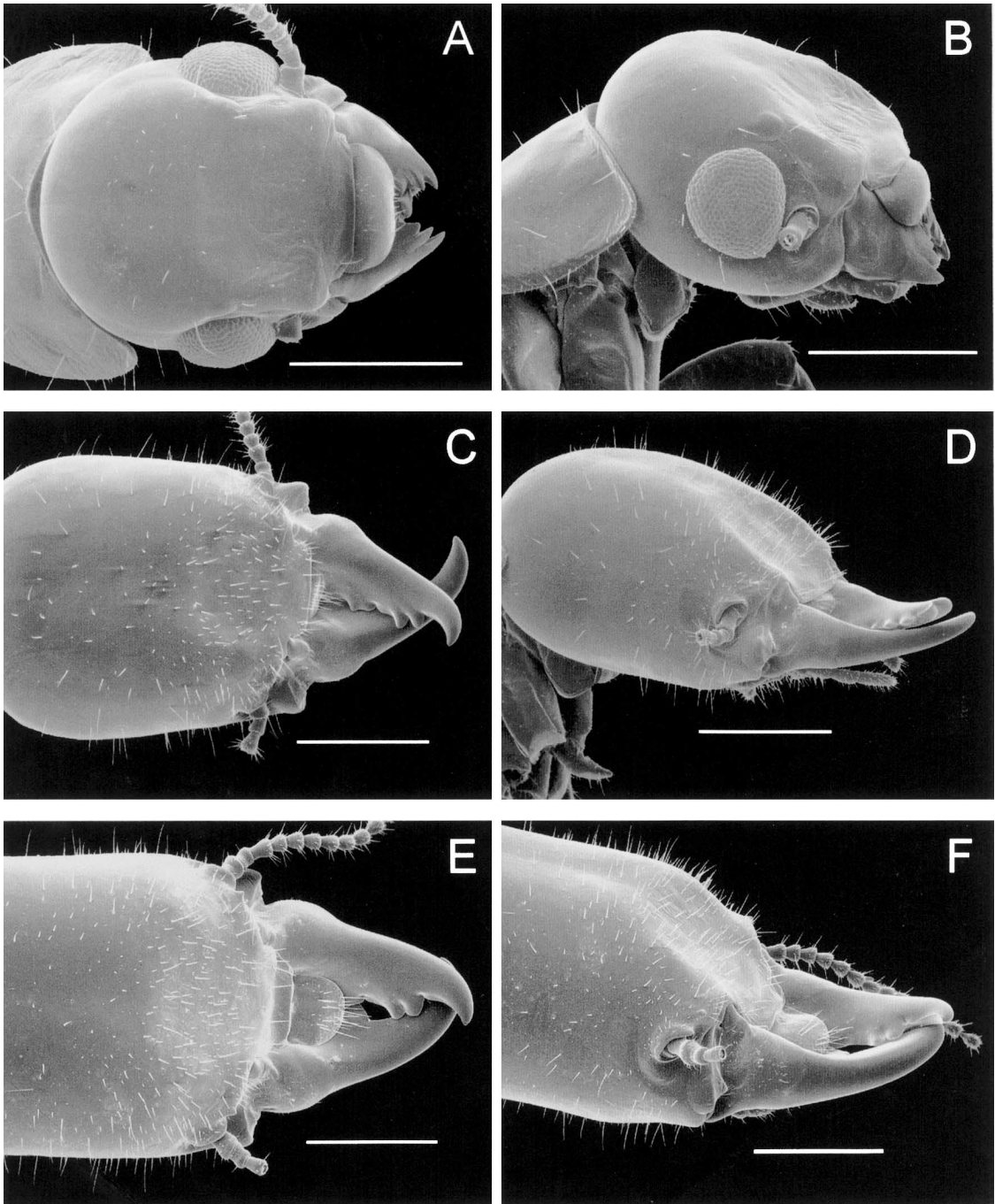


Fig. 2. Scanning electron micrographs of *Neoterмес platyfrons*: dorsal (A) and oblique (B) views of the imago head, dorsal (C) and lateral (D) views of the small soldier head, and dorsal (E) and lateral (F) views of the large soldier head. All specimens are from Punta Cana, La Altagracia Province, Dominican Republic. Antennae and palpi partially removed for clarity. Scale bar equals 1 mm.

maximum width 1.75-2.05, 2.00-2.22, and 2.22-2.47 mm, and head maximum width at eyes 1.59-1.81, 1.83-1.93, and 2.00-2.17 mm.

The distinct and darkened V- and M-shaped markings on the *N. platyfrons* imago head are absent on *N. jouteli* head. The long setae on the head



TABLE 1. MEASUREMENTS OF *NEOTERMES PLATYFRONS* IMAGO.

| Measurement in mm<br>(n = 6 males, 6 females from 4 colonies) | Range       | Mean $\pm$ S.D.  | Holotype |
|---|-------------|------------------|----------|
| Head length with labrum                                       | 1.91-2.20   | 2.03 $\pm$ 0.11  | 1.95     |
| Head length to postclypeus                                    | 1.55-1.68   | 1.60 $\pm$ 0.037 | 1.55     |
| Head width, maximum at eyes                                   | 1.83-1.93   | 1.89 $\pm$ 0.029 | 1.83     |
| Head height without postmentum                                | 1.05-1.11   | 1.07 $\pm$ 0.018 | 1.08     |
| Labrum width, maximum   | 0.67-0.77   | 0.72 $\pm$ 0.031 | 0.74     |
| Eye diameter with sclerite, maximum                           | 0.59-0.64   | 0.62 $\pm$ 0.015 | 0.63     |
| Eye to head base, minimum from sclerite                       | 0.25-0.32   | 0.32 $\pm$ 0.020 | 0.29     |
| Ocellus diameter, maximum                                     | 0.19-0.25   | 0.22 $\pm$ 0.020 | 0.23     |
| Ocellus diameter, minimum                                     | 0.15-0.18   | 0.16 $\pm$ 0.010 | 0.16     |
| Eye sclerite to ocellus, minimum                              | 0.01-0.06   | 0.03 $\pm$ 0.014 | 0.02     |
| Pronotum, maximum length                                      | 1.29-1.47   | 1.40 $\pm$ 0.052 | 1.39     |
| Pronotum, maximum width                                       | 2.00-2.22   | 2.08 $\pm$ 0.061 | 2.07     |
| Total length with wings                                       | 16.47-18.74 | 17.76 $\pm$ 0.82 | 17.61    |
| Total length without wings                                    | 7.95-10.22  | 9.06 $\pm$ 0.65  | 8.52     |
| Fore wing length from suture                                  | 12.78-14.34 | 13.64 $\pm$ 0.51 | 13.49    |
| Fore wing, maximum width                                      | 3.40-3.79   | 3.55 $\pm$ 0.13  | 3.43     |

and pronotum of *N. platyfrons* are about 0.2 mm long while those of *N. jouteli* are  $>5\times$  shorter. The shallow depression centered at the intersection of the epicranial suture in *N. jouteli* and *N. mona* is faint or absent in *N. platyfrons*. The anterior margin of the pronotum is more concave in *N. platyfrons* than in *N. jouteli*. The posterior corners of the *N. platyfrons* pronotum are subtruncate, while in *N. mona* they are more rounded and more pilose.

The imago of the sympatric *N. castaneus* (Burmeister) is unlike that of *N. platyfrons*. The general color of the former is castaneous, while that of *N. platyfrons* is orange. The eyes of *N. platyfrons* are  $\sim 1.5\times$  larger in diameter than those of *N. castaneus*, and the frons of *N. platyfrons* is flat-

tened and rugose while that of *N. castaneus* is convex and smooth.

Soldier. (Figs. 2 C-F, Tables 2-3).

Beyond size differences given in Tables 2 and 3, soldier caste dimorphism of this species is less distinctive when compared with some congeners. Consequently, the large and small soldier morphs are described together.

In dorsal view, head capsule, distal antennal articles, and labrum ferruginous except for chestnut brown anterior frons, postclypeus, frontal and antennal carinae, and 4 proximal antennal articles. Anteclypeus yellowish-white. Thorax and abdominal dorsum ferruginous orange. Mandibles

TABLE 2. MEASUREMENTS OF *NEOTERMES PLATYFRONS* SMALL SOLDIER.

| Measurement in mm (n = 13 from 11 colonies)                                | Range      | Mean $\pm$ S.D.  |
|--|------------|------------------|
| Head length to tip of mandibles  | 3.86-4.31  | 4.13 $\pm$ 0.16  |
| Head length to postclypeus   | 2.57-3.02  | 2.85 $\pm$ 0.15  |
| Head width, maximum  | 2.00-2.30  | 2.14 $\pm$ 0.077 |
| Antennal carinae, outside span   | 1.78-2.03  | 1.92 $\pm$ 0.072 |
| Head height, excluding postmentum  | 1.52-1.73  | 1.62 $\pm$ 0.072 |
| Labrum, maximum width  | 0.51-0.60  | 0.55 $\pm$ 0.030 |
| Postclypeus width, maximum   | 0.72-0.87  | 0.80 $\pm$ 0.042 |
| Left mandible length, tip to most distant visible point of ventral condyle | 1.89-2.01  | 1.97 $\pm$ 0.031 |
| Postmentum, length in middle   | 1.68-2.14  | 1.91 $\pm$ 0.14  |
| Postmentum, maximum width  | 0.72-0.83  | 0.77 $\pm$ 0.036 |
| Postmentum, minimum width  | 0.38-0.47  | 0.43 $\pm$ 0.035 |
| Pronotum, maximum width  | 2.30-2.52  | 2.39 $\pm$ 0.083 |
| Pronotum, maximum length   | 1.22-1.50  | 1.36 $\pm$ 0.095 |
| Hind tibia length  | 1.29-1.64  | 1.44 $\pm$ 0.096 |
| Total length   | 8.66-10.93 | 9.98 $\pm$ 0.75  |

TABLE 3. MEASUREMENTS OF *NEOTERMES PLATYFRONS* LARGE SOLDIER.

| Measurement in mm (n = 10 from 8 colonies)                                 | Range      | Mean $\pm$ S.D.  |
|--|------------|------------------|
| Head length to tip of mandibles  | 4.63-5.43  | 4.97 $\pm$ 0.26  |
| Head length to postclypeus   | 3.22-3.96  | 3.58 $\pm$ 0.24  |
| Head width, maximum  | 2.17-2.59  | 2.34 $\pm$ 0.13  |
| Antennal carinae, outside span   | 2.00-2.39  | 2.18 $\pm$ 0.12  |
| Head height, excluding postmentum  | 1.75-1.95  | 1.85 $\pm$ 0.074 |
| Labrum, maximum width  | 0.52-0.64  | 0.58 $\pm$ 0.033 |
| Postclypeus width, maximum   | 0.83-0.95  | 0.89 $\pm$ 0.040 |
| Left mandible length, tip to most distant visible point of ventral condyle | 2.05-2.30  | 2.17 $\pm$ 0.078 |
| Postmentum, length in middle   | 2.41-2.77  | 2.55 $\pm$ 0.11  |
| Postmentum, maximum width  | 0.83-0.95  | 0.87 $\pm$ 0.034 |
| Postmentum, minimum width  | 0.44-0.52  | 0.48 $\pm$ 0.030 |
| Pronotum, maximum width  | 2.47-3.13  | 2.76 $\pm$ 0.17  |
| Pronotum, maximum length   | 1.38-1.71  | 1.57 $\pm$ 0.11  |
| Hind tibia length  | 1.52-1.70  | 1.63 $\pm$ 0.071 |
| Total length   | 8.66-13.35 | 10.91 $\pm$ 1.25 |

glossy, almost black except for very dark chestnut brown bases. Epicranial sutures faint. Eyes blackish or fainter dark pigmentation. Femora yellowish-white; tibiae pale ferruginous. Sternum pale ferruginous orange. Ferruginous postmentum contrasting with pale ferruginous orange genae. Large soldiers are slightly more pigmented than small soldiers.

In dorsal view, head capsule moderately elongate in small soldier; relatively more elongated in large soldier. Sides of head subparallel. In small soldier, sides faintly convex or converging to anterior; in large soldier, sides usually very faintly concave to middle. Head capsule with posterior corners evenly rounded; posterior margin narrowly rectate in small soldier, widely rectate in large soldier. Head capsule and pronotum relatively densely covered with  $\approx$  0.1-mm-long setae. Frons depressed or slightly concave. Frontal carinae each with small conical tubercle. Labrum, when exposed, linguiform. Mandibles robust; in large soldier slightly more stout than in small soldier; bases faintly humped and with few minute setae on large soldier; left apical tooth hooked  $\approx$  60°, dentition conspicuous. Antennae with 14-16 articles in small soldier, usually 14-15; 13-16 in large soldier, usually 15-16; relative length formula  $2 < 3 > 4 = 5$ . Third antennal article subclavate, proximal articles moniliform, distal articles slightly elongate. Pronotum large and relatively short, noticeably wider than head capsule. Anterior margin of pronotum broadly and shallowly concave; lateral margins convex; posterior margin widely subrectate, usually with very faint median emargination.

In lateral view, head capsule slightly dorsoventrally flattened. Plane of frons slopes  $\approx$  20° below plane of vertex in small soldiers;  $\approx$  30° in large soldiers. Eyes oval. Mandibles moderately curved upward in lateral view. Femora moderately broad in small soldier; noticeably swollen in large soldier.

Comparisons.

Morphometric differentiation of the small soldiers of *Neotermes platyfrons* and *N. jouteli* is very tenuous although hind tibia lengths are 1.29-1.64 mm and 1.54-1.72 mm, respectively. Less character overlap occurs in large soldiers but this morph is also difficult to differentiate by measurements. The maximum pronotum lengths of large soldiers range from 1.38-1.71 mm in *N. platyfrons* and 1.71-1.85 mm in *N. jouteli*, and the hind tibia lengths are 1.52-1.70 mm and 1.68-1.93 mm, respectively.

Both soldier morphs of *N. platyfrons* differ from those of *N. jouteli* with the former having proportionally more elongate head capsules and mandibles (e.g., head length/maximum head width ratio in minor soldiers is 1.35 in *N. platyfrons* and 1.2 in *N. jouteli*). The left apical tooth of *N. platyfrons* is noticeably hooked, while in *N. jouteli* it is only faintly hooked. The labrum of *N. platyfrons* is apically rounded while subtruncate in *N. jouteli*. Basal mandibular pilosity in *N. platyfrons* is almost absent while well developed in *N. jouteli*.

Compared to the sympatric *N. mona*, both small and large soldiers of *N. platyfrons* are smaller in the following respective measurements (mm): outer span of antennal carinae (small soldiers 2.18-2.80 vs. 1.78-2.03 and large soldiers 2.57-3.10 vs. 2.00-2.39), labrum maximum width (small 0.64-0.87 vs. 0.51-0.60 and large 0.72-0.83 vs. 0.52-0.64), left mandible length (small 2.24-2.84 vs. 1.89-2.01 and large 2.67-3.07 vs. 2.05-2.30), pronotum maximum length (small 1.63-2.02 vs. 1.22-1.50 and large 2.00-2.37 vs. 1.38-1.71), and pronotum maximum width (small 2.73-3.28 vs. 2.30-2.52 and large 3.37-3.96 vs. 2.47-3.13). The soldier head capsule, including the postmentum, pronotum, and legs of *N. mona*, is more pilose than those of *N. platyfrons*.

Both soldier morphs of *N. platyfrons* differ with those of the sympatric *N. castaneus* considerably. The *N. castaneus* soldier is significantly larger than that of *N. platyfrons*. The *N. castaneus* soldier has unpigmented eye spots, while the eyes of *N. platyfrons* are pigmented. The mandibles of *N. castaneus* are distinctly paler proximally than distally, while those of *N. platyfrons* are almost concolorous throughout.

#### Etymology.

The species name is derived from the Greek "*platys*" which refers to the depressed or rather planer character of the frons in imagos and soldiers.

#### Type Material.

**Holotype. Dominican Republic.** La Altagracia Province, 2.6 km S.W. of Punta Cana, 18.533°N 68.368°W, 6-XI-1996, J. Chase & J. Krecek, 1 male holotype alate, 5 paratype alates, 3 paratype small soldiers and 3 paratype large soldiers (DR 1480). Additional specimens from this colony used for SEMs (Fig. 2).

**Paratypes. Dominican Republic.** La Altagracia Province, 4.4 km N.W. of Boca de Yuma, 18.392°N 68.647°W, 5-XI-1996, J. Chase & J. Krecek, 2 alates, 1 small soldier, 1 large soldier (DR 1378); 1 km N.W. of Playa Bavaro, 18.665°N 68.409°W, 5-XI-1996, J. Chase & J. Krecek, 1 small soldier (DR 1389); 1 km W. of Playa Dominicus, 18.366°N 68.831°W, 6-XI-1996, J. Chase & J. Krecek, 1 large soldier (DR 1409); same data, 1 small soldier (DR 1410); 0.5 km W. of Club Med., 18.554°N 68.353°W, 6-XI-1996, J. Chase & J. Krecek, 1 small soldier, 1 large soldier (DR 1448); Bejucal, 18.667°N 68.867°W, 10-VI-1992, R. Scheffrahn, J. Chase, J. Mangold, and J. de la Rosa, 1 small soldier, 1 large soldier (DR 504); Bavaro, 18.683°N 68.433°W, 11-VI-1992, R. Scheffrahn, J. Chase, J. Mangold, and J. de la Rosa, 1 small soldier (DR 512); Juanillo, 18.483°N 68.417°W, 11-VI-1992, R. Scheffrahn, J. Chase, J. Mangold, and J. de la Rosa, 1 small soldier (DR 567); Playa Bavaro, 18.683°N 68.767°W, 1-IV-1993, J. Chase & J. de la Rosa, 1 large soldier (DR 827); Barahona Province, Camino a Santa Elena, 18.167°N 71.133°W, 30-III-1993, J. Chase & J. de la Rosa, 2 alates, 1 small soldier, 1 large soldier (DR 793); Maria Trinidad Sanchez Province, Cabo Frances, 19.667°N 69.950°W, 9-VIII-1992, J. Chase, 2 alates, 1 small soldier, 1 large soldier (DR 682).

**Additional Material Examined.** Same data as DR 512 above, 1 small soldier (DR 514), 1 small soldier each (DR 526 and DR 538); same data as DR 682, 1 small soldier (DR 680); same data as DR 1378, 1 small soldier, 1 large soldier (DR 1379), 1 large soldier (DR 1380); same data as DR 1448,

1 small soldier, 1 large soldier (DR 1445), 1 small soldier, 1 large soldier (DR 1446), 1 queen (DR 1449); same data as DR 1480, 1 small soldier (DR 1481), and 1 small soldier, 1 large soldier (DR 1482); La Altagracia Province, 0.5 km E. of Playa Bayahibe, 18.372°N 68.839°W, 6-VI-1992, J. Chase & J. de la Rosa, 1 king, 1 queen (DR 1423).

#### DISCUSSION

We postulate that *N. platyfrons* was first listed as *N. (=K.) jouteli* by Harris (1955) in an intercepted wood shipment from the Dominican Republic to England. Harris (1971), Araujo (1977), and Scheffrahn et al. (1994) also accepted this initial determination. Based on current records, *N. castaneus*, *N. mona*, and *N. platyfrons* are the only *Neotermes* recorded from the Dominican Republic. Collections of *N. platyfrons* have been most prominent in extreme eastern Dominican Republic although two remote locations, one southwestern and one north-central, have also yielded specimens (Fig. 1). *Neotermes mona* has a broader distribution in the Dominican Republic (Krecek et al. 2000) that encompasses the distribution of *N. platyfrons* in the extreme east. The distribution of *N. castaneus* in the Dominican Republic is also more widespread than *N. platyfrons*, but these two species have only been recorded to be sympatric near Barahona. Except for the first few instars, all castes of *N. platyfrons* exhibit some degree of wing bud formation.

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## COLLECTION OF FALL ARMYWORM (LEPIDOPTERA: NOCTUIDAE) ADULTS AND NONTARGET HYMENOPTERA IN DIFFERENT COLORED UNITRAPS

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### ABSTRACT

Field experiments were conducted to determine the effectiveness of different colored pheromone-baited traps in capture of Fall Armyworm, *Spodoptera frugiperda*, males and nontarget Hymenoptera. Plastic Universal Moth Traps (Unitraps) of different colors were baited with commercial sex pheromone and were placed in peanut and corn fields in northern Florida. In one study, standard-colored (green canopy, yellow funnel, white bucket) traps collected more moths than all-white or all-green traps. More Sphecoidea were found in white traps while more Vespoidea were collected in standard traps. In another study, trap capture was compared among standard, all-white, and standard traps with buckets painted two different yellow colors. Results showed that there were few differences in capture among traps with different colors, however, contrasts between traps with yellow buckets or traps with white buckets suggested more moths could be captured in yellow-bucket traps. Very few Hymenoptera were collected, although more Apoidea were found in white traps.

Key Words: insect behavior, *Spodoptera*, monitoring, pheromone traps, Hymenoptera

### RESUMEN

Experimentos de campo fueron conducidos para determinar la efectividad de trampas de distintos colores con señuelo de feromona para capturar al machos del gusano *Spodoptera frugiperda*, y miembros de Himenóptera. Trampas Plásticas Universales de Mariposa (Unitraps) de colores diferentes fueron preparadas con señuelo de feromona comercial de sexo y colocadas en campos de maíz y maní en el norte de Florida. En un estudio, trampas de color estándar (toldo verde, embudo amarillo, cubeta blanca) colectaron mas mariposas que trampas de color blanco o verde solamente. Mas Sphecoidea fueron encontrados en trampas blancas mientras que más Vespoidea fueron colectados en trampas estándar. En otro estudio, captura por trampa fue comparada entre estándar, blanca, y trampas estándar con cubetas pintadas con dos colores amarillos diferentes. Los resultados demuestran que hubo poca diferencia en capturas entre trampas con diferentes colores, sin embargo, contrastes entre trampas con cubetas amarillas o trampas con cubetas blancas sugirieron que más mariposas pueden ser capturadas con trampas de cubeta amarilla. Muy pocos Himenóptera fueron colectados, aunque mas Apoidea fueron encontrados en trampas blancas.

Traps for insect pests can be categorized into those that catch insects randomly (interception or passive traps such as Malaise, sticky pane, or pit-fall traps) and those that attract and elicit orientating behavior (active traps such as light or baited traps) (Southwood 1978). Traps that attract insects use visual cues (yellow sticky cards, yellow pans, red spheres, etc.), chemical cues (pheromones, kairomones), edible baits, or any combination of the three. Workers involved in pest management research have developed traps that combine visual and chemical cues to attract insects, such as the work conducted with tephritids (Robacker et al. 1990, Stark & Vargas 1992, Epsky et al. 1995).

Few studies have documented the influence of visual cues in monitoring of noctuid moths. Decoys (dead female moths) were shown to improve capture of *Helicoverpa zea* (Boddie) males in electrocutor grid and sticky traps (Gross et al. 1983).

Trap color has been shown to be important in the collection of several noctuids such as *Heliothis virescens* (F.) (Hendricks et al. 1972), *Anticarsia gemmatalis* Hübner (Mitchell et al. 1989), *Agrotis ipsilon* (Hufnagel), and *Pseudaletia unipuncta* (Haworth) (Hendrix & Showers 1990). Plastic bucket traps with green canopies, yellow funnels, and white buckets collected more *Spodoptera* spp. males than all-green traps in several studies (Mitchell et al. 1989; Pair et al. 1989; Lopez 1998). All-white traps are also commercially available, but have not been bioassayed for Fall Armyworm, *S. frugiperda* (J. E. Smith) capture. My studies were designed to compare capture of Fall Armyworm males using the same chemical cue (commercially available sex pheromone) but different visual cues (trap colors).

Few studies have documented the species and number of nontarget Hymenoptera that are captured in traps intended for agricultural lepi-

dopteran pests. Meagher & Mitchell (1999) found species from Apoidea, Pompiloidea, Scolioidea, Sphecoidea, and Vespoidea in traps using sex pheromone and synthetic floral volatiles as lures, however, all traps used white buckets. Because of the deleterious effect on foraging pollinators and natural enemies (Meagher & Mitchell 1999), and the increased time required to service traps containing these insects (Gross & Carpenter 1991), an additional objective of this study was to collect and identify nontarget aculeate Hymenoptera attracted to different trap colors.

#### MATERIALS AND METHODS

1998

Peanuts (*Arachis hypogaea* L., 'Georgia Green') were planted during summer in northwestern Alachua County, Florida. This experiment was designed to compare moth and aculeate Hymenoptera capture in three Universal Moth Trap (Nitrap) (Great Lakes IPM, Vestaburg, MI) colors (all white, all green, or standard = green canopy, yellow funnel, white bucket). Traps were placed along roads and edges in two fields, and the experiment was designed as a randomized complete block with four replications of the three trap colors. Trap location within a replication was randomized weekly, and trapping began 21 July and ended 30 September. Both Trécé® (Trécé, Inc., Salinas, CA) red septa and Scentry® (Ecogen, Inc., Langhorne, PA) gray septa *S. frugiperda* pheromone lures were used. These lures were alternated and replaced every two weeks. All traps contained insecticide strips (Hercon® Vaportape II containing 10% dichlorovos, Hercon Environmental Co., Emigsville, PA) to kill captured insects. Trap contents were removed three or more times per week. Moth numbers per night and Hymenoptera numbers per day were compared across trap colors using a split-block analysis of variance (ANOVA) (Steel & Torrie 1980). To satisfy ANOVA assumptions, counts were  $\log(x + 1)$  transformed before analysis. Means for each trap color were separated using a LSD mean separation test (SAS Institute 1996). Untransformed means ( $\pm$ SE) are given in text and figures, whereas statistical results refer to transformed data.

1999

Four different trapping treatments were placed at the University of Florida, Hastings Research and Education Center's Yelvington Farm, St. John's County during July and August. The four trap colors were standard, white, and standard traps with buckets spray painted Fluorescent Yellow (Painter's Touch #1942, Rust-Oleum Corp., Vernon Hills, IL) or Sun Yellow Gloss (Painter's Touch #1945). Traps were placed along

roads among large plots of silage corn (*Zea mays* L.). The experiment was designed as a randomized complete block with four replications of the four trap colors. Trap location within a replication was randomized weekly, and trapping began 13 July and ended 19 August. Scenturion® (Scenturion Inc., Clinton, WA) gray septa Fall Armyworm pheromone lures were used, and were replaced every two weeks. All traps contained Vaportape II insecticide strips to kill moths that were captured. Trap contents were removed three or more times per week. Moth numbers per night and Hymenoptera numbers per day were compared across trap colors using a split-block ANOVA with  $\log(x + 1)$ -transformed data. Means for each trap color or each trap color combination were separated using a LSD mean separation test or orthogonal contrasts (PROC GLM, CONTRAST statement, SAS Institute 1996).

#### RESULTS

1998

Adult male Fall Armyworm numbers in the trap samples were high during July and August, and gradually decreased during September (Fig. 1). Comparison of moth captures among three trap colors showed that the standard traps captured significantly larger numbers of moths in 20 of 37 sampling dates. Most of the sampling dates where standard traps captured larger numbers of moths were during July and August, when traps were collecting over 50 moths per night. Only three sampling dates in September produced significant differences among traps. The overall average showed that standard traps captured more moths than white traps; green traps caught the fewest number of moths ( $54.0 \pm 4.0$ ,  $24.1 \pm 1.8$ ,  $11.9 \pm 1.2$  moths per night, respectively) ( $F = 51.2$ ;  $df = 2, 6$ ;  $P = 0.0002$ ).

Collection of nontarget bees and wasps, both in number of individuals and number of species, was lower than previously documented (Fig. 2) (Meagher and Mitchell 1999). Species collected included *Melissodes* sp., *Apis mellifera* (L.), *Bombus pennsylvanicus* (De Geer), *Campomeris plumipes fossulana* (F.), *Cerceris bicornuta* Guérin, *Ammophila* spp., and *Polistes* spp. More Sphecoidea were collected in white traps than standard or green traps ( $F = 7.3$ ;  $df = 2, 6$ ;  $P = 0.0249$ ); more Vespoidea were collected in standard traps ( $F = 4.1$ ;  $df = 2, 6$ ;  $P = 0.0743$ ) (Fig. 3). There was a trend toward fewer Apoidea in green traps.

1999

Consistently large numbers of Fall Armyworm males were collected in July and August from the surrounding corn fields (Fig. 3). There were no differences in number of males per night collected

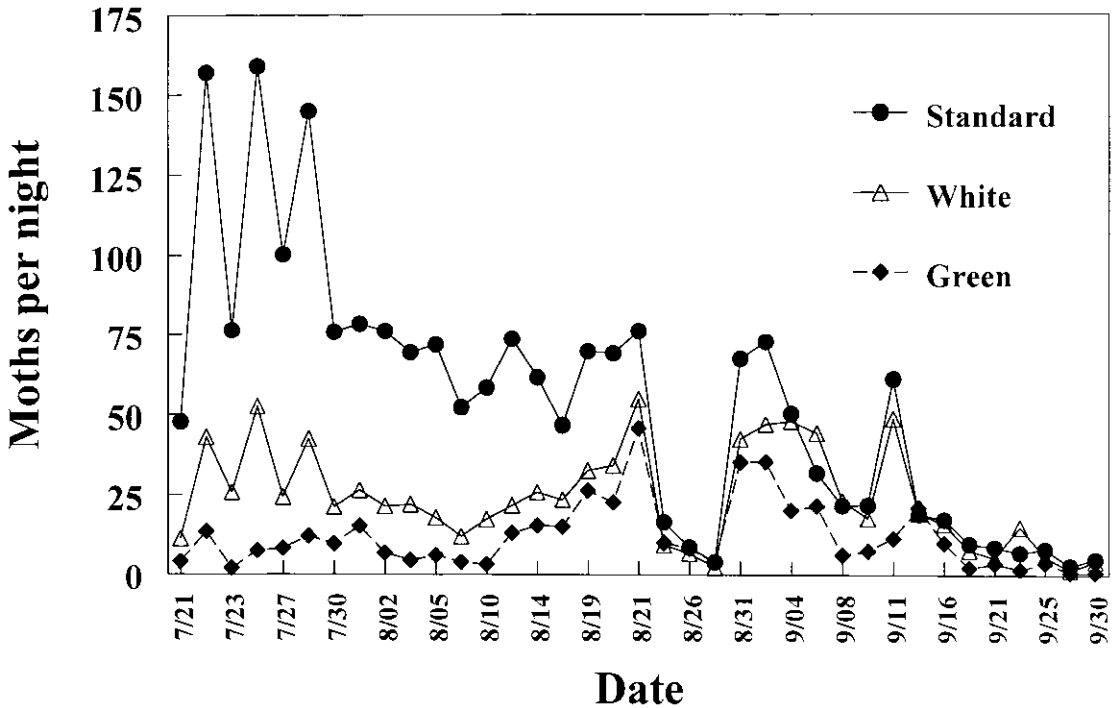


Fig. 1. Capture of male Fall Armyworm in standard (green canopy, yellow funnel, white bucket), all-white, or all-green Unitraps baited with sex pheromone lures, Alachua, FL, 1998.

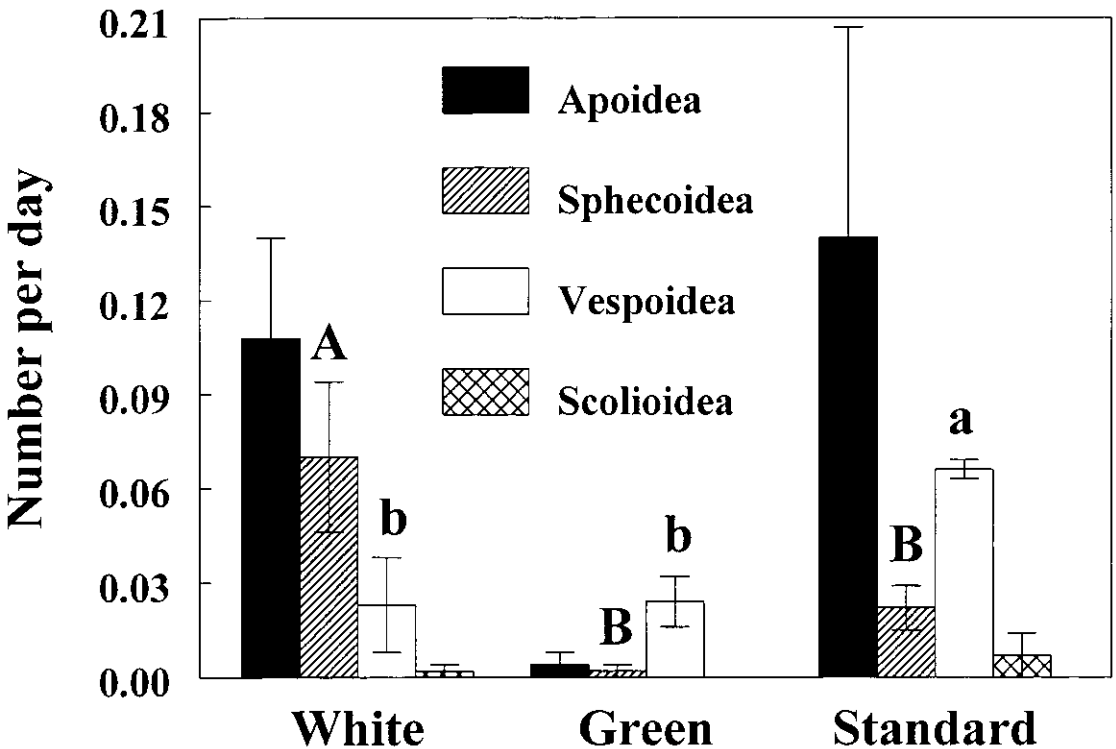


Fig. 2. Number of aculeate Hymenoptera captured per day in Fall Armyworm pheromone lure bucket traps in peanut, Alachua, FL, 1998. Means within Sphecoidea followed by the same uppercase letter and Vespoidea followed by the same lowercase letter, are not significantly different.

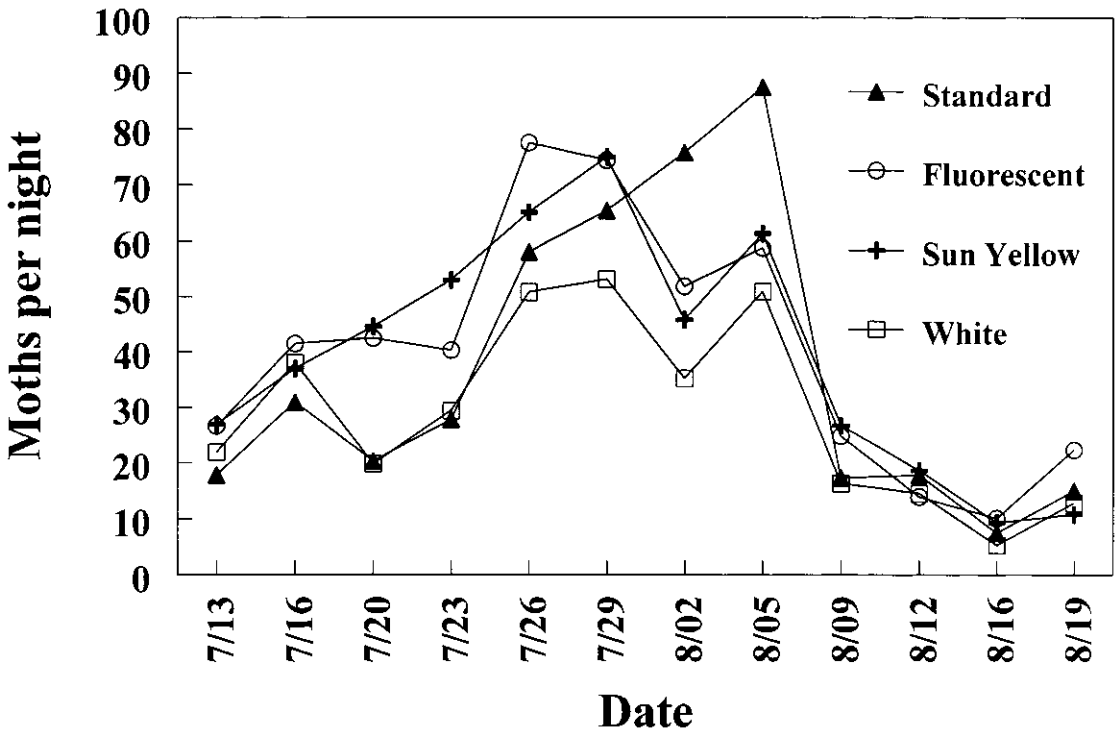


Fig. 3. Capture of male Fall Armyworm in standard (green canopy, yellow funnel, white bucket), fluorescent (green canopy, yellow funnel, fluorescent yellow bucket), sun yellow (green canopy, yellow funnel, sun yellow bucket), or all-white Unitraps baited with sex pheromone lures, Hastings, FL, 1999.

in traps with different colors when analyzed across dates (fluorescent yellow  $40.4 \pm 4.2$ , sun yellow  $39.5 \pm 4.6$ , standard  $36.7 \pm 4.7$ , white  $29.0 \pm 3.3$ ;  $F = 2.7$ ;  $df = 3, 9$ ;  $P = 0.1089$ ). When each of the 12 dates was analyzed separately, only 2 dates (20 July, 2 August) had differences in males captured among all trap colors ( $P < 0.05$ ). However, when different trap color combinations were subjected to contrasts, differences in capture were noted. Generally, traps with yellow buckets (fluorescent yellow and sun yellow traps) captured

more moths than traps with white buckets (standard and white traps), and all-white traps collected fewer moths (Table 1).

Very few Vespoidea, Sphecoidea, Tiphioidea, and Scolioidea were collected. Since more *Bombus* spp. were collected, their numbers were analyzed separately from other species of Apoidea. More Apoidea were collected from white traps than the other traps, whereas there were no differences among trap colors in collection of *Bombus* spp. (Fig. 4).

TABLE 1. CONTRAST OF NUMBERS OF FALL ARMYWORM MALES CAPTURED BETWEEN TRAPS WITH DIFFERENT COLORED BUCKETS, HASTINGS, FL, 1999

| Contrast                                  | F-value | Pr > F | N         | Means ± SE                       |
|---|---------|--------|-----------|----------------------------------|
| fluorescent + sun<br>vs. white            | 7.4     | 0.0238 | 96<br>48  | $39.9 \pm 3.1$<br>$29.0 \pm 3.3$ |
| fluorescent + sun<br>vs. standard         | 2.7     | 0.1350 | 96<br>48  | $39.9 \pm 3.1$<br>$36.7 \pm 4.7$ |
| fluorescent + sun<br>vs. standard + white | 7.1     | 0.0257 | 96<br>96  | $39.9 \pm 3.1$<br>$32.9 \pm 2.9$ |
| fluorescent + sun + standard<br>vs. white | 5.3     | 0.0470 | 144<br>48 | $38.9 \pm 2.6$<br>$29.0 \pm 3.3$ |



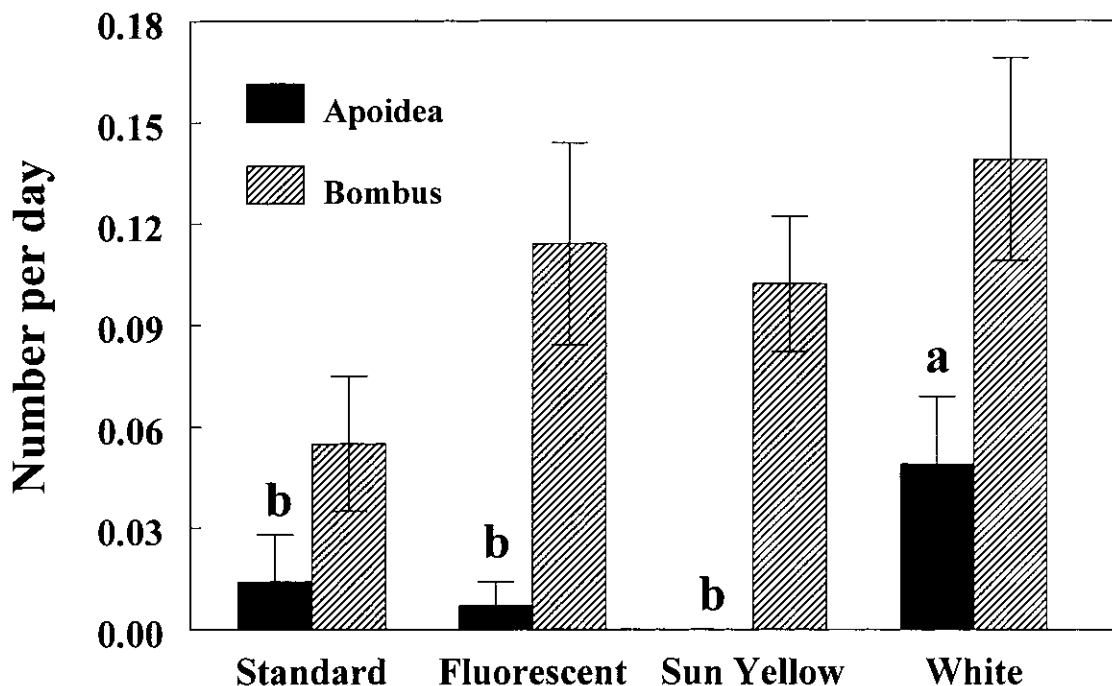


Fig. 4. Number of Apoidea and *Bombus* spp. captured per day in Fall Armyworm pheromone lure Unitraps in corn, Hastings, FL, 1999. Means within Apoidea followed by the same letter are not significantly different.

#### DISCUSSION

Trap color was shown to influence Fall Armyworm capture in previous trials (Mitchell et al. 1989; Pair et al. 1989), however in those studies all-white traps and traps with yellow funnel and buckets were not compared. My results showed that more moths were collected in standard traps than all-white or all-green traps, a result similar to what has been documented with *S. exigua* (Hübner) (Lopez 1998). Spectral analysis of the white, yellow, and green components of bucket traps indicates a possible factor responsible for decreased capture of moths in green traps was low light reflectance at wavelengths where moth eyes are most sensitive (Mitchell et al. 1989). However, too much reflectance may decrease moth capture since all-white traps captured fewer moths than standard traps.

Traps composed of yellow funnels and buckets tended to collect more males than traps with white buckets, but differences were not significant on all dates. Spectral analysis showed higher reflectance for fluorescent yellow than sun yellow in the 500-560 nm range (unpublished data), although this difference appears not to have influenced trap capture. Therefore, if the objective of monitoring for Fall Armyworm is to collect the largest number of moths, than it can be concluded

from this research and from previous research (Meagher & Mitchell 2001), that the standard Unitrap with green canopy, yellow funnel, and white bucket is the best collector of male Fall Armyworm when used with available commercial sex pheromones.

The attraction of *Bombus* spp. to traps using noctuid sex pheromone lures has been previously documented (Adams et al. 1989; Hendrix & Showers 1990; Gross & Carpenter 1991; Meagher & Mitchell 1999). Trap color seems to be important in the capture of bumblebees (Hamilton et al. 1971), although chemical cues either from the pheromone, the insecticidal strips, or both, may contribute to capture of these insects (Gross & Carpenter 1991). As far as capture of other aculeate Hymenoptera (Sphecoidea, Vespoidea, and Scolioida) in different colored traps, very little has been documented.

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## AERIAL FEEDING APHIDS OF CORN IN THE UNITED STATES WITH REFERENCE TO THE ROOT-FEEDING *APHIS MAIDIRADICIS* (HOMOPTERA: APHIDIDAE)

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### ABSTRACT

A brief summary of taxonomic characters, usual hosts, and distribution within the United States are given for each species. *Aphis craccivora* Koch, *Aphis fabae* Scopoli, *Aphis gossypii* Glover, *Aphis maidiradicis* Forbes, *Hysteroneura setariae* (Thomas), *Macrosiphum euphorbiae* (Thomas), *Metopolophium dirhodum* (Walker), *Myzus persicae* (Sulzer), *Rhopalosiphum maidis* (Fitch), *Rhopalosiphum padi* (L.), *Sipha flava* (Forbes), *Schizaphis graminum* (Rondani), and *Sitobion avenae* (Fabricius) are included in the present paper. Pictorial and dichotomous keys are included to aid personnel charged with detection, identification, and control of aphids associated with corn in the United States.

Key Words: Taxonomic keys, identification, control, distribution, *Zea mays*

### RESUMEN

Un breve resumen de características taxonómicas, hospederos usuales, y distribución dentro de los Estados Unidos son dados para cada especie. *Aphis craccivora* Koch, *Aphis fabae* Scopoli, *Aphis gossypii* Glover, *Aphis maidiradicis* Forbes, *Hysteroneura setariae* (Thomas), *Macrosiphum euphorbiae* (Thomas), *Metopolophium dirhodum* (Walker), *Myzus persicae* (Sulzer), *Rhopalosiphum maidis* (Fitch), *Rhopalosiphum padi* (L.), *Sipha flava* (Forbes), *Schizaphis graminum* (Rondani), y *Sitobion avenae* (Fabricius) son incluidos en este trabajo. Claves pictóricas y dicotómicas son incluidas para apoyar al personal encargado de detectar, identificar, y controlar los áfidos asociados con el maíz algodón en los Estados Unidos.

Corn or maize (*Zea mays* L.) ranks first among the agricultural crops in both area devoted to its cultivation and the value of the annual crop in the United States. For the 1997 production year, over 80 million acres in the United States were planted in corn for grain with Iowa the leading state with over 12 million acres (Anonymous 1999a; Anonymous 1999b). The total value of production of corn for grain was over \$24 billion in 1997 (Anonymous 1999a). Sweet corn and popcorn varieties serve for human consumption while the field corn varieties are live-stock food, and the herbage is used for forage. In addition, corn-based products are used for a wide variety of items.

In 1996, insecticides were used on 32% of the total acreage planted for corn (Anonymous 1999c). Over 200 species of insects, several of which are aphids, have been recorded as injurious to corn during some part of its life cycle or as a stored product (Bailey 1935). When aphid colonies are large, they can greatly diminish a plant's vigor or even kill the plant through mechanical injury by removal of sap during feeding. Besides mechanical injury, some aphids are able to transmit diseases that affect corn (Chan et al. 1991). Aphids also have the capability of transmitting

nonpersistent viruses between plants that would not otherwise be considered hosts. Aphids also produce a sticky substance called honeydew during feeding. This substance may be problematic when it fouls the corn tassel and interferes with pollination and encourages fungal growth.

The aerial aphid fauna includes at least 12 species that commonly colonize corn in the United States. Several other taxa also feed on the roots of corn (e.g., *Anoecia* spp., *Geoica* spp., *Pemphigus* spp.) however, only the most commonly found, the corn root aphid, *Aphis maidiradicis* Forbes, will be addressed in this paper. A brief summary of taxonomic characters, hosts, worldwide distribution, and U.S. distribution is given for each of the 13 included species: *Aphis craccivora* Koch, *Aphis fabae* Scopoli, *Aphis gossypii* Glover, *Aphis maidiradicis* Forbes, *Hysteroneura setariae* (Thomas), *Macrosiphum euphorbiae* (Thomas), *Metopolophium dirhodum* (Walker), *Myzus persicae* (Sulzer), *Rhopalosiphum maidis* (Fitch), *Rhopalosiphum padi* (L.), *Sipha flava* (Forbes), *Schizaphis graminum* (Rondani), and *Sitobion avenae* (Fabricius). Descriptions as well as written and illustrated keys are included as an aid for detection, identification, and control of aphids associated with corn in the United States.

## MATERIALS AND METHODS

In the synonymy section, one asterisk (\*) represents the name used by Palmer (1952) and two asterisks (\*\*) represent the name appearing in Blackman & Eastop (1984). Common names are those approved by the Entomological Society of America (ESA) (Bosik 1997).

Information on distribution and hosts is taken from labels on slides in the National Collection of Insects, Beltsville, Maryland, and from records in Palmer (1952), Smith & Parron (1978), and Blackman & Eastop (1984).

In the illustrated keys, the species are grouped by morphological differences of the antennae and antennal tubercles, body and antennal setae, pigmentation of the abdomen, coloration of cornicle, size of cauda, number of caudal setae, and wing venation. Characters used in the keys are apparent with a dissecting microscope with a minimum power of 16 $\times$  and are best seen at 50 $\times$ . Relative body size of the aphid species follows the division proposed by Blackman & Eastop (1984): body length <2.00 mm are "small," 2.00-3.00 mm are "medium," and >3.00 mm are "large." Figure 1 includes a figure illustrating general characters of a wingless and a winged adult. Body length is measured dorsally from the center of the frons to the end of the abdomen, excluding the cauda. Length of the antennal "terminal process" is measured as the distance from the large primary sensorium to the tip of the last antennal segment. Length of the "base" of the antenna is measured from the basal portion of the last antennal segment to the apex of the primary sensorium. Caudal length is measured along the midline from the beginning of the sclerotized portion to the apex. The keys are not intended for identification of single, errant aphids but should be used for individuals fully colonizing corn. Ideally winged aphids should develop from nymphs collected from a colony on the plant.

*Aphis craccivora* Koch 1854  
Figs. 1, 4, 6-7

## Synonymy:

\**Aphis medicaginis* Koch 1854  
(misidentification)

\*\**Aphis craccivora* Koch

ESA approved common name: cowpea aphid.

Other common names: black legume aphid, groundnut aphid.

Taxonomic characters: Wingless adult female.—In life, body shiny black with large black patch on dorsum of abdomen; legs strikingly white with black area near apex of femur and tibia; immatures often covered with grayish wax. Small sized, body length 1.2-1.9 mm, rounded. Antenna 6 segmented; tubercles not well developed; terminal process approximately 1 $\frac{2}{3}$ -2 $\frac{1}{3}$  times length of base of antennal segment VI; antennal segments III-V without secondary sensoria; longest setae

on antennal segment III shorter than diameter of segment. Cornicle black, cylindrical; approximately 3 $\frac{1}{3}$ -4 $\frac{3}{4}$  times as long as wide, longer than length of cauda. Cauda black, with 2-4 (usually 3) pairs of lateral setae and 0-1 dorsal preapical seta.

Winged adult female.—In life body shiny black with black lateral areas and variable bands on dorsum of abdomen, legs similar to wingless adult female; forewing with media twice branched, hind wing with 2 oblique veins; small to medium sized, body length 1.4-2.0 mm. Antenna 6 segmented; tubercles not developed; terminal process approximately 2 times length of base of antennal segment VI; antennal segment III with 5-7 secondary sensoria, 1 or 2 noticeably larger than the others, longest setae on antennal segment III shorter than diameter of segment; antennal segments IV-V without secondary sensoria. Cornicle black, cylindrical; 4-5 times as long as wide, longer than length of cauda. Cauda black, with 2-3 pairs of lateral setae and 0-1 dorsal preapical seta.

Hosts: Polyphagous with a preference for the Leguminosae.

Distribution in the United States: Throughout the United States.

Distribution in the world: Virtually worldwide.

Comments: *Aphis craccivora* transmits 51 plant viruses but is not listed as a vector of a corn virus (Chan et al. 1991).

*Aphis fabae* Scopoli 1763  
Figs. 1, 4-7

## Synonymy:

\* & \*\**Aphis fabae* Scopoli

ESA approved common name: bean aphid.

Other common name: black bean aphid.

Taxonomic characters: Wingless adult female.—In life body black, but may appear dull black due to waxy covering; immatures often covered with wax. Small to medium sized, body length 1.1-2.5 mm, rounded. Antenna 6 segmented; tubercles not well developed; terminal process approximately 2 $\frac{2}{3}$ -3 $\frac{2}{3}$  times length of base of antennal segment VI; antennal segments III-IV without secondary sensoria; longest setae on antennal segment III longer than diameter of segment. Cornicle dark, cylindrical; 2-4 times as long as wide, longer than length of cauda. Cauda dark, elongate with 4-7 pairs of lateral setae and 0-1 dorsolateral setae.

Winged adult female.—In life body dull black, usually with dark lateral areas and bands on dorsum of abdomen; forewing with media twice branched, hind wing with 2 oblique veins; small to medium sized, body length 1.7-2.2 mm. Antenna 6 segmented; tubercles not well developed; terminal process approximately 2 $\frac{1}{3}$ -4 times length of base of antennal segment VI; 9-20 secondary sensoria on antennal segment III; 0-6 secondary sensoria on antennal segment IV; longest setae on antennal segment III longer than diameter of segment.

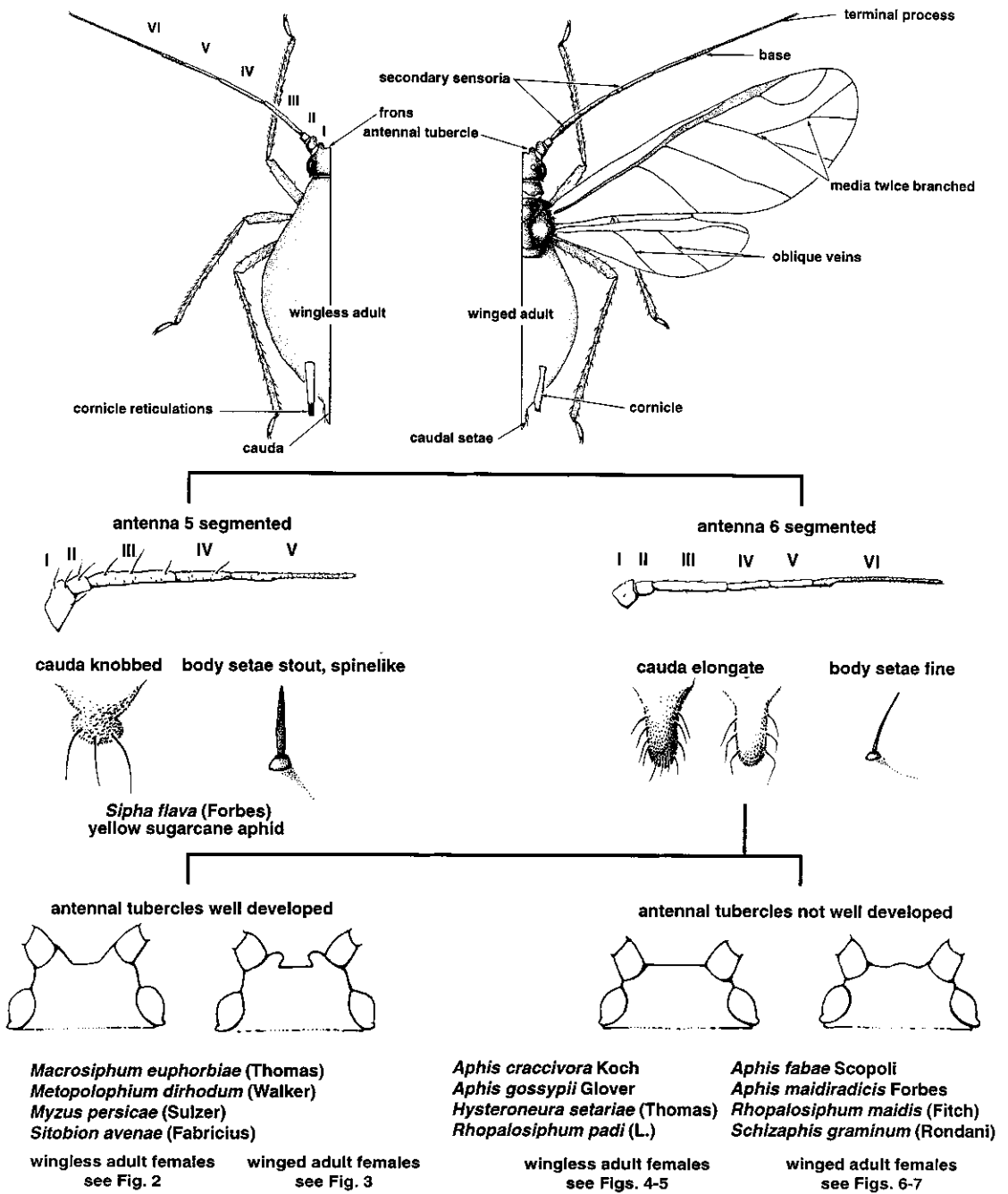


Fig. 1. Pictorial key to 13 aphid species that commonly colonize corn in the United States.

Cornicle dark, cylindrical; approximately 2½-4 times as long as wide, longer than length of cauda. Cauda dark, elongate with 5-8 pairs of lateral setae and 0-2 dorsolateral setae.

Hosts: Polyphagous and damaging to many plants of economic importance.

U.S. distribution: Widespread.

Distribution in the world: Virtually worldwide.

Comments: *Aphis fabae* transmits 42 plant viruses but is not listed as a vector of a virus of corn (Chan et al. 1991). Several subspecies have been described in the *A. fabae* complex.

*Aphis gossypii* Glover 1877

Figs. 1, 4-7

## Synonymy:

\* & \*\**Aphis gossypii* Glover

ESA approved common name: cotton or melon aphid.

Other common names: none.

Taxonomic characters: Wingless adult female.—In life, body color varying from dark green to pale yellow or nearly white. Small sized, body length 1.0-1.4 mm, body rounded. Antenna 5-6 segmented; tubercles not well developed, terminal process approximately 2¼-3 times length of base of antennal segment VI; antennal segment III without secondary sensoria; longest setae on antennal segment III shorter than diameter of segment. Cornicle dark, cylindrical with slight tapering; approximately 2½-5 times as long as wide, longer than length of cauda. Cauda pale to dusky, with 2-3 pairs of lateral setae.

Winged adult female.—In life, body shape and coloration similar to wingless adult female; forewing with media twice branched, hind wing with 2 oblique veins; small sized, body length 1.2-1.8 mm, rounded. Antenna 6 segmented; tubercles not well developed; terminal process approximately 2¾-3½ times length of base of antennal segment VI; antennal segment III with 3-8 secondary sensoria in a row, longest setae on antennal segment III shorter than diameter of segment; antennal segments IV-V without secondary sensoria. Cornicle dark, cylindrical with slight tapering; approximately 2½-3¾ times as long as wide, longer than length of cauda. Cauda pale to dusky, elongate with 2-3 pairs of lateral setae.

Hosts: Polyphagous and very damaging to many plants of economic importance.

U.S. distribution: Widespread.

Distribution in the world: Virtually worldwide.

Comments: *Aphis gossypii* transmits 76 plant viruses including sugarcane mosaic virus which is listed as a virus of corn (Chan et al. 1991).*Aphis maidiradicis* Forbes 1891

Figs. 1, 4, 6

## Synonymy:

\**Aphis maidi-radicis* Forbes\*\**Aphis maidiradicis* Forbes

ESA approved common name: corn root aphid.

Other common names: none.

Taxonomic characters: Wingless adult female.—In life body bluish green with dark head and dusky transverse thoracic and abdominal bands. Small to medium sized, body length 1.5-2.0 mm long, body rounded. Antenna 6 segmented; tubercles not well developed, terminal process approximately 1½-2 times length of base of antennal segment VI; antennal segment III without secondary sensoria; longest setae on antennal segment III shorter than diameter of segment.

Cornicle dusky, cylindrical; approximately 1½-2½ times as long as wide, subequal to length of cauda. Cauda dusky, with 7-8 pairs of lateral setae and 1-2 preapical setae.

Winged adult female.—In life head and thorax black, abdomen light green with dusky markings; forewing with media twice branched, hind wing with 2 oblique veins; body length 1.4-1.9 mm body, rounded. Antenna 6 segmented; tubercles not well developed; terminal process approximately 1½-2¼ times length of base of antennal segment VI, antennal segment III with 4-11 secondary sensoria, longest setae on antennal segment III shorter than diameter of segment; antennal segment IV with 0-2 secondary sensoria, antennal segment IV without secondary sensoria. Cornicle dusky, cylindrical; 2-3 times as long as wide, subequal to length of cauda. Cauda dusky, with 5-8 pairs of lateral setae and 1-2 preapical setae.

Hosts: *Aphis maidiradicis* is principally known as a pest of corn in the U.S. but has been collected on the roots of a wide range of hosts (Blackman & Eastop 1984).

Distribution in the United States: Widespread.

Distribution in the world: Brazil, Jamaica, U.S.A.

Comments: *Aphis maidiradicis* is not recorded as transmitting plant viruses (Chan et al. 1991). This species is found on roots and is often tended by ants. The taxonomic status of *A. maidiradicis*, *A. middletonii*, *A. armoraciae*, and others are unclear and require detailed taxonomic and biological research.

*Hysteroneura setariae* (Thomas 1878)

Figs. 1, 4-6

## Synonymy:

*Siphonophora setariae* Thomas\**Aphis setariae* (Thomas)\*\**Hysteroneura setariae* (Thomas)

ESA approved common name: rusty plum aphid.

Other common names: none.

Taxonomic characters: Wingless adult female.—In life body dark reddish brown, apical area of tibiae dark, cornicles dark to almost black, cauda pale to nearly white. Small to medium sized, 1.2-2.2 mm, body rounded. Antenna 6 segmented; tubercles not well developed, terminal process approximately 4½-5¾ times length of base of antennal segment VI; antennal segments III-V without secondary sensoria; longest setae on antennal segment III shorter than diameter of segment. Cornicle dark to nearly black, tapered apically; approximately 3½-4½ times as long as wide, longer than length of cauda. Cauda pale to nearly white, elongate, with 2-3 (usually 2) pairs of lateral setae.

Winged adult female.—In life coloration similar to wingless adult female; forewing with media twice branched, hind wing with one oblique vein;

small sized, body length 1.2-1.8 mm. Antenna 6 segmented; tubercles not well developed, terminal process approximately 6-7½ times length of base of antennal segment VI; antennal segment III with 12-17 secondary sensoria, longest setae on antennal segment III shorter than diameter of segment; antennal segment IV with 0-6 secondary sensoria; antennal segment V without secondary sensoria. Cornicle dark to nearly black, tapered apically, approximately 4½-6½ times as long as wide, longer than length of cauda. Cauda pale to nearly white, with 2 pairs of lateral setae.

Hosts: Primary hosts include *Prunus* spp. (Blackman & Eastop 1984) however, secondary hosts include numerous species of Gramineae including corn.

U.S. distribution: Widespread.

Distribution in the world: Virtually worldwide.

Comments: *Hysteroneura setariae* transmits six plant viruses including guinea grass mosaic virus and sugarcane mosaic virus which are listed as viruses affecting corn (Chan et al. 1991).

*Macrosiphum euphorbiae* (Thomas 1878)

Figs. 1-3

Synonymy:

*Siphonophora euphorbiae* Thomas

\**Macrosiphum solanifolii* (Ashmead 1882)

\*\**Macrosiphum euphorbiae* (Thomas)

ESA approved common name: potato aphid.

Other common names: none.

Taxonomic characters: Wingless adult female.—

In life, body color varies between shades of green or pink. Medium to large sized, body length 2.2-3.3 mm, pear shaped. Antenna 6 segmented; tubercles well developed with inner faces divergent; terminal process approximately 4½-6 times length of base of antennal segment VI; antennal segment III with 1-6 secondary sensoria, longest setae on antennal segment III shorter than diameter of segment; antennal segments IV-V without secondary sensoria. Cornicle pale or becoming increasingly dusky towards apex, approximately 6½-9 times as long as wide, longer than length of cauda, with slight apical constriction and several rows of polygonal reticulations in constricted area. Cauda pale, with 3-4 pairs of lateral setae and 1-3 dorsal preapical setae.

Winged adult female.—In life, body usually of varying shades of green or pink; forewing with media twice branched, hind wing with 2 oblique veins; medium to large sized, body length 2.2-3.0 mm, pear shaped. Antenna 6 segmented; frontal tubercles well developed with inner faces divergent; terminal process approximately 5½-7 times length of base of antennal segment VI; antennal segment III with 11-20 secondary sensoria, longest setae on antennal segment III shorter than diameter of segment; antennal segments IV-V without secondary sensoria. Cornicle sometimes pale but

usually progressively darker towards apex, with slight apical constriction and several rows of polygonal reticulations in constricted area, approximately 7½-10½ times as long as wide, longer than length of cauda. Cauda pale, with 4-5 pairs of lateral setae and 1-2 dorsal preapical setae.

Hosts: *Macrosiphum euphorbiae* is polyphagous and damaging to many plants of economic importance.

U.S. distribution: Widespread.

Distribution in the world: Virtually worldwide.

Comments: *Macrosiphum euphorbiae* transmits 67 plant viruses but is not listed as vector of a corn virus (Chan et al. 1991).

*Metopolophium dirhodum* (Walker 1849)

Figs. 1-3

Synonymy:

*Aphis dirhodum* Walker

\**Macrosiphum dirhodum* (Walker)

\*\**Metopolophium dirhodum* (Walker)

ESA approved common name: none.

Other common names: rose-grass aphid, rose-grain aphid.

Taxonomic characters: Wingless adult female.—In life, body green to yellow green with dark green longitudinal stripe along the middle of the dorsum. Small to medium sized, body length 1.7-2.7 mm, elongate. Antenna 6 segmented with dark bands on apices of antennal segments III-V, the base of VI dark near the primary sensoria, and the terminal process dark; tubercles well developed with inner faces divergent; terminal process approximately 3¼-4 times length of base of antennal segment VI; antennal segment III with 1-2 secondary sensoria, longest setae on antennal segment III shorter than diameter of segment; antennal segments IV-V without secondary sensoria. Cornicle pale with darker apices, cylindrical with slight tapering to apical flange; approximately 3¾-5½ times as long as wide, longer than length of cauda. Cauda pale, with 2-4 pair of lateral setae and 2-3 dorsal preapical setae.

Winged adult female.—In life, abdomen green without markings; forewing with media twice branched, hind wing with 2 oblique veins; small to medium sized, body length 1.9-2.6 mm, elongate. Antenna 6 segmented; frontal tubercles well developed with inner faces divergent; terminal process approximately 3¼-4 times length of base of antennal segment VI; antennal segment III with 14-21 secondary sensoria over most of the length, longest setae on antennal segment III shorter than diameter of segment; antennal segments IV-V without secondary sensoria. Cornicle pale with darker apices, cylindrical with slight tapering to apical flange, approximately 4-6½ times as long as wide, longer than length of cauda. Cauda pale, with 3-4 pairs of lateral setae and 2-4 dorsal preapical setae.

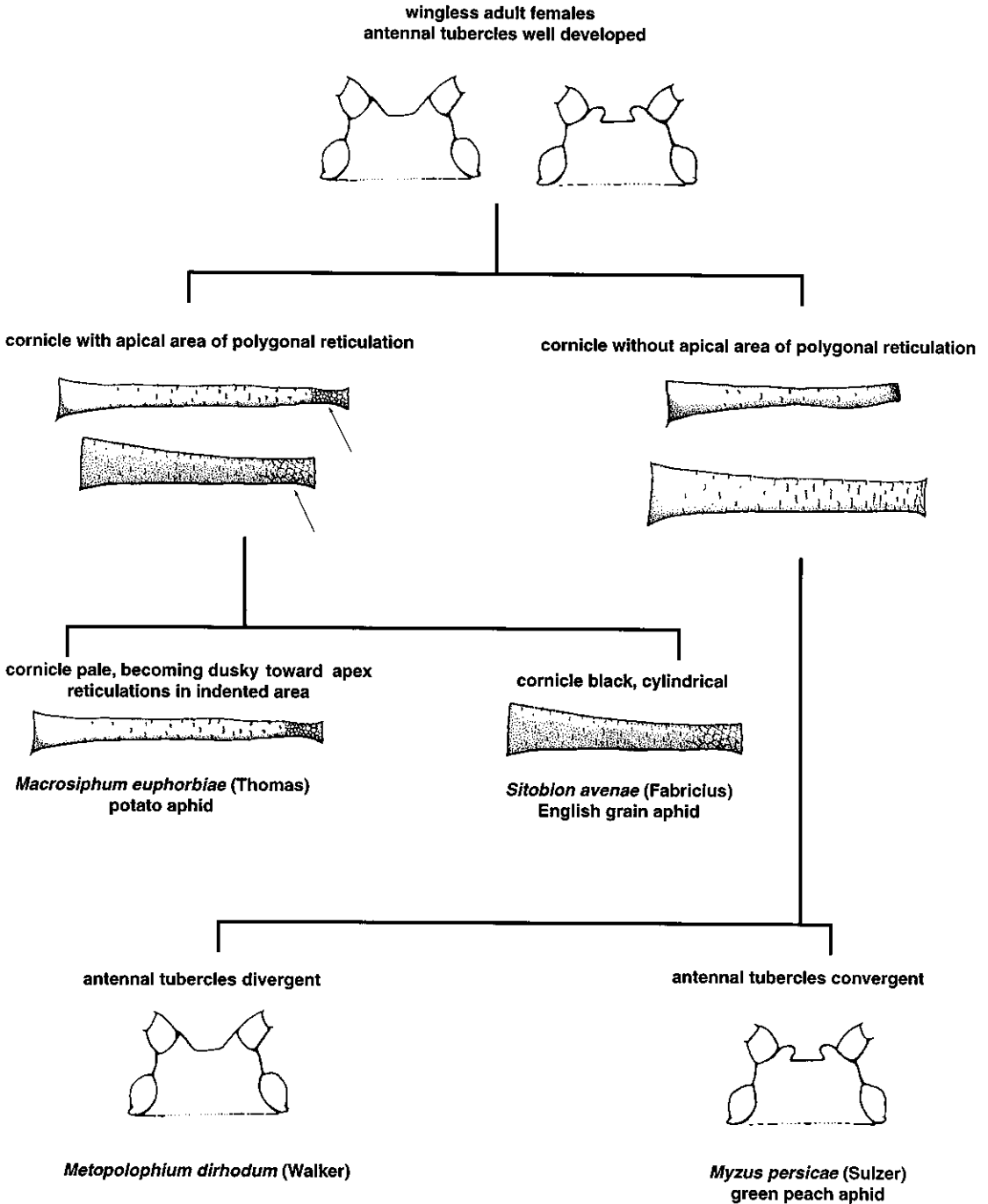


Fig. 2. Pictorial key to wingless adult females of four aphid species that commonly colonize corn in the United States and have well developed antennal tubercles.

Hosts: Primary hosts of *M. dirhodum* include wild and cultivated species of *Rosa*, however, secondary hosts include several species of Gramineae including corn.

U.S. distribution: Widespread except in tropics.  
Distribution in the world: Africa, Central Asia, Europe, the Middle East, New Zealand, North America, and South America.



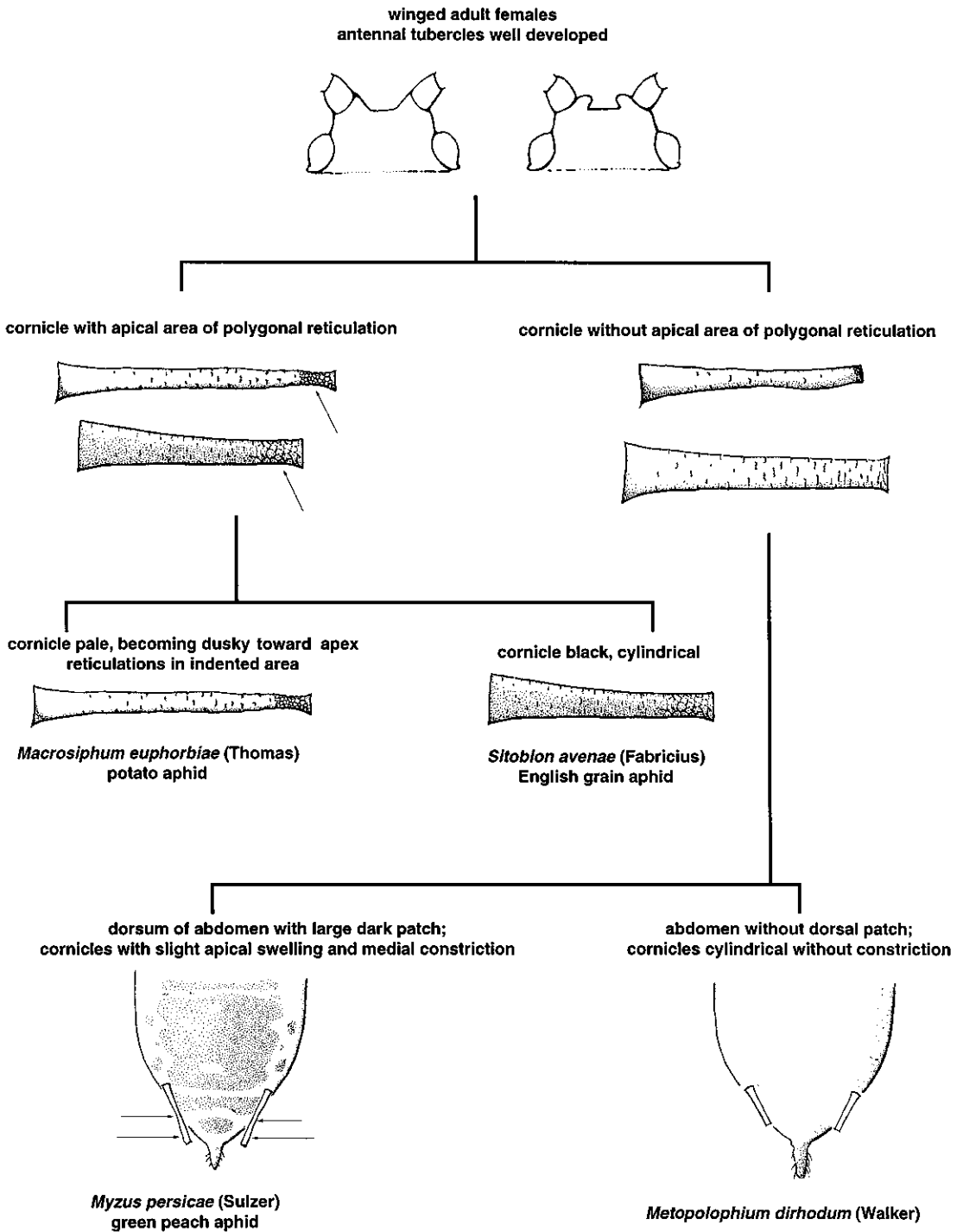


Fig. 3. Pictorial key to winged adult females of four aphid species that commonly colonize corn in the United States and have well developed antennal tubercles.

Comments: *Metopolophium dirhodum* transmits three plant viruses including barley yellow

dwarf virus which is listed as a virus affecting corn (Chan et al. 1991).

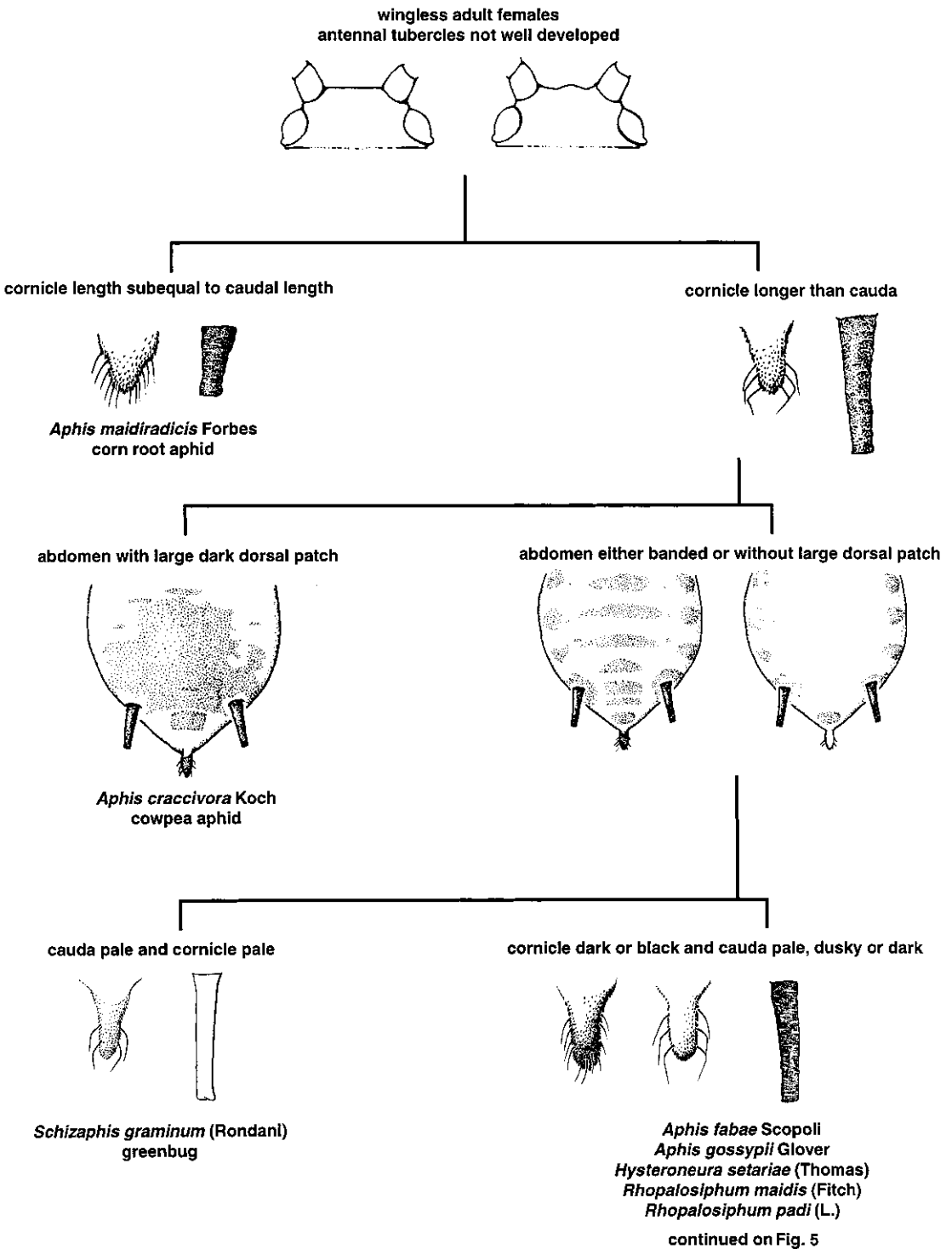


Fig. 4. Pictorial key to wingless adult females of eight aphid species that commonly colonize corn in the United States and have antennal tubercles not developed.

*Myzus persicae* (Sulzer 1776)

Figs. 1-3

## Synonymy:

*Aphis persicae* Sulzer\* & \*\**Myzus persicae* (Sulzer)

ESA approved common name: green peach aphid.

Other common name: peach-potato aphid.

Taxonomic characters: Wingless adult female.—In life, body color varies from green to pale yellow. Small to medium sized, body length 1.5-2.1 mm, pear shaped. Antenna 6 segmented; tubercles well developed with inner faces convergent; terminal process approximately  $3\frac{1}{4}$ - $4\frac{1}{2}$  times length of base of antennal segment VI; antennal segments III-V without secondary sensoria, longest setae on antennal segment III shorter than diameter of III. Cornicle pale but apex may be dark, slight apical swelling and slight medial constriction; approximately  $4\frac{2}{3}$ - $7\frac{3}{4}$  times as long as wide, longer than length of cauda. Cauda pale to dusky, with 3 pairs of lateral setae.

Winged adult female.—In life, body varies from green to pale yellow with a large dark patch on dorsum of abdomen; forewing with media twice branched, hind wing with 2 oblique veins; small to medium sized, body length 1.4-2.2 mm, pear shaped. Antenna 6 segmented; tubercles well developed with inner faces convergent; terminal process approximately  $2\frac{3}{4}$ - $4\frac{2}{3}$  times length of base of antennal segment VI; antennal segment III with 6-16 secondary sensoria, longest setae on antennal segment III shorter than diameter of segment; antennal segments IV-V without secondary sensoria. Cornicle dusky to dark but apex sometimes darker, slight apical swelling and slight medial constriction; approximately  $4\frac{1}{3}$ -9 times as long as wide, longer than length of cauda. Cauda pale to dusky, with 3 pairs of lateral setae.

Hosts: Primary hosts include several species of *Prunus*, however *M. persicae* is polyphagous and damaging to many other plants of economic importance.

U.S. distribution: Widespread.

Distribution in the world: Virtually worldwide.

Comments: *Myzus persicae* transmits more than 182 plant viruses including barley yellow dwarf virus and sugarcane mosaic virus which are listed as viruses affecting corn (Chan et al. 1991).

*Rhopalosiphum maidis* (Fitch 1856)

Figs. 1, 4-7

## Synonymy:

*Aphis maidis* Fitch\* & \*\**Rhopalosiphum maidis* (Fitch)

ESA approved common name: corn leaf aphid.

Other common names: none.

Taxonomic characters: Wingless adult female.—In life, body color blue green to olive green with

reddish-purple areas around cornicle bases, occasionally wax covered. Small sized, body length 1.7-2.6 mm, pair shaped. Antenna 6 segmented; tubercles not well developed, terminal process approximately 2-2 $\frac{1}{2}$  times length of base antennal segment VI; antennal segments III-V without secondary sensoria, longest setae on antennal segment III longer than diameter of segment. Cornicle dark, slightly constricted apically,  $2\frac{1}{2}$ - $3\frac{1}{3}$  times as long as wide, longer than length of cauda. Cauda dark, with 2 pairs of lateral setae.

Winged adult female.—In life, abdominal color yellow green to dark green; forewing with media twice branched; hind wing with 2 oblique veins; small sized, body length 1.6-2.0 mm. Antenna 6 segmented, terminal process approximately  $1\frac{1}{2}$ - $2\frac{1}{3}$  times length of base antennal segment VI; antennal segment III with 11-20 secondary sensoria, longest setae on antennal segment III shorter than diameter of segment; antennal segment IV with 1-6 secondary sensoria; antennal segment V with 0-4 secondary sensoria. Cornicle dark, slightly constricted apically,  $2\frac{1}{2}$ - $3\frac{1}{2}$  times as long as wide, longer than length of cauda. Cauda dark, with 2 pairs of lateral setae.

Hosts: *Rhopalosiphum maidis* feeds on numerous species of Gramineae including many that are economically important.

U.S. distribution: Widespread.

Distribution in the world: Virtually worldwide.

Comments: *Rhopalosiphum maidis* transmits more than 15 plant viruses, including barley yellow dwarf virus, guinea grass mosaic virus, and sugarcane mosaic virus which are listed as affecting corn (Chan et al. 1991).

*Rhopalosiphum padi* (Linnaeus 1758)

Figs. 1, 4-7

## Synonymy:

*Aphis padi* Linnaeus\* & \*\**Rhopalosiphum padi* (Linnaeus)

ESA approved common name: bird cherry-oat aphid.

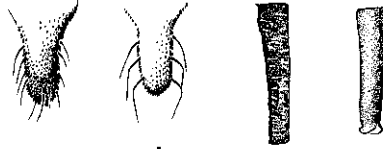
Other common names: oat bird-cherry aphid.

Taxonomic characters: Wingless adult female.—In life, body color varies from light yellow green mottling to dark green, often with orange patches around base of cornicles. Small to medium sized, body length 1.5-2.1 mm, pear shaped. Antenna 6 segmented; tubercles not well developed, terminal process approximately 4-5 $\frac{1}{2}$  times length of base antennal segment VI; antennal segments III-V without secondary sensoria, longest setae on antennal segment III shorter than diameter of segment. Cornicle dark, cylindrical, slightly constricted apically; approximately  $2\frac{1}{3}$ -4 times as long as wide, longer than length of cauda. Cauda dark, with 2-3 (usually 2) pairs of lateral setae.

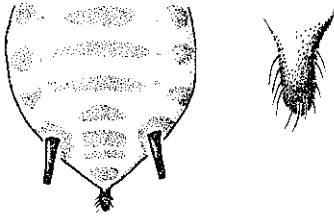
Winged adult female.—In life, abdominal color light green to dark green; forewing with media twice branched, hind wing with 2 oblique veins;

continued from Fig. 4

cornicle dark or black and cauda pale, dusky or dark

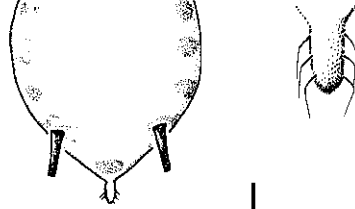


abdomen with dorsal dark bands;  
cauda with 4-7 pairs of lateral setae



*Aphis fabae* Scopoli  
bean aphid

abdomen without dorsal dark bands;  
cauda with 3 or fewer pairs of lateral setae



cauda pale or dusky

cauda dark or black

apex of femur dark or black

apex of femur without dark or black coloration



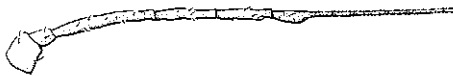
*Hysteroneura setariae* (Thomas)  
rusty plum aphid



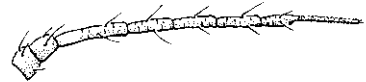
*Aphis gossypii* Glover  
cotton or melon aphid

terminal process >3 times length of base;  
antennal setae shorter than width of segment

terminal process < 3 times length of base;  
antennal setae longer than width of segment



*Rhopalosiphum padi* (L.)  
bird cherry-oat aphid



*Rhopalosiphum maidis* (Fitch)  
corn leaf aphid

Fig. 5. Continued pictorial key to wingless adult females of eight aphid species that commonly colonize corn in the United States and have antennal tubercles not developed.

small to medium sized, body length 1.6-2.0 mm. Antenna 6 segmented, terminal process approximately 4 1/3-5 1/3 times length of base antennal segment VI; antennal segment III with 11-21 secondary senso-

ria, longest setae on antennal segment III shorter than diameter of segment; antennal segment IV with 3-9 secondary sensoria; antennal segment V with 0-1 secondary sensoria. Cornicle dark, cylin-

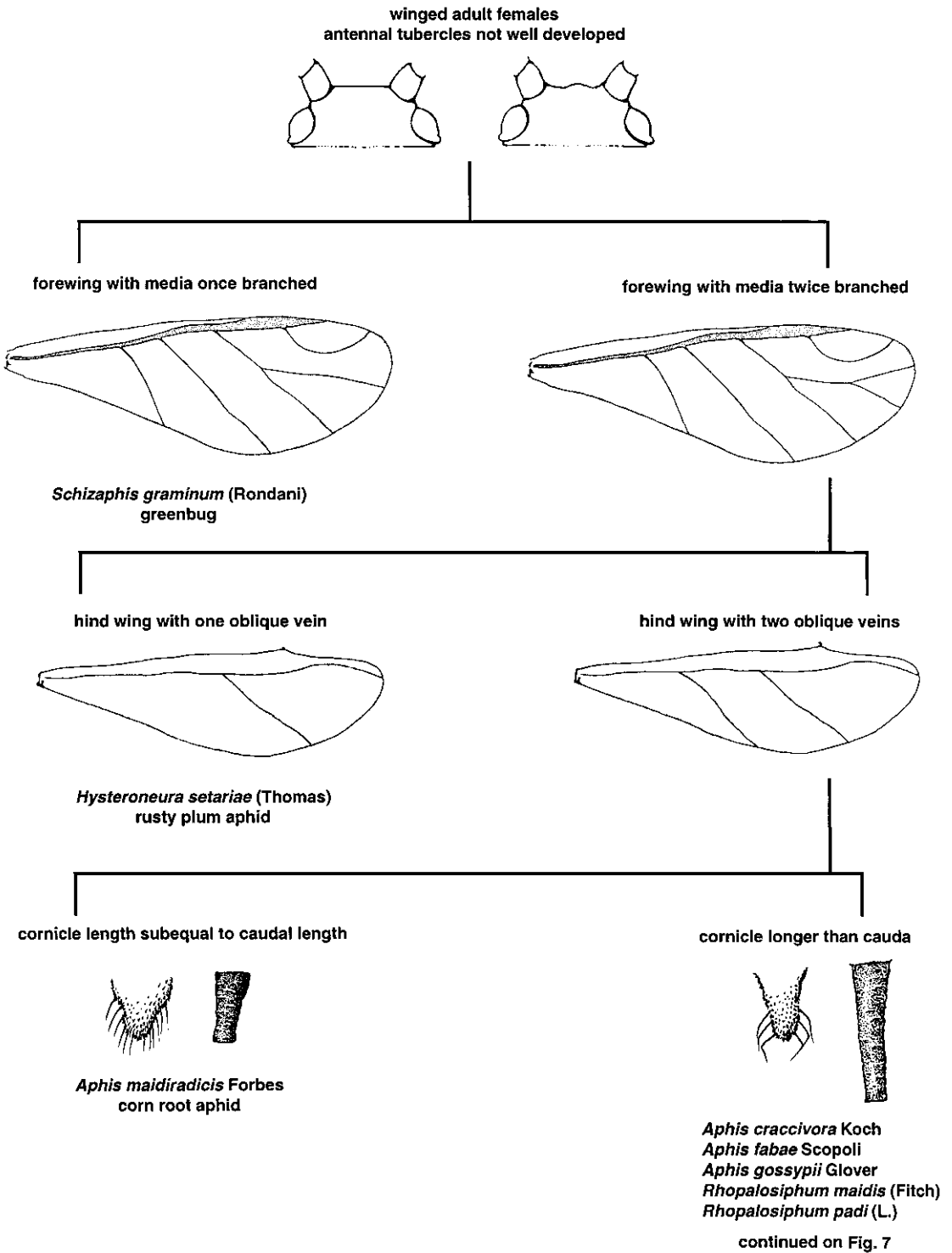


Fig. 6. Pictorial key to winged adult females of eight aphid species that commonly colonize corn in the United States and have antennal tubercles not developed.

continued from Fig. 6

cornicle longer than cauda

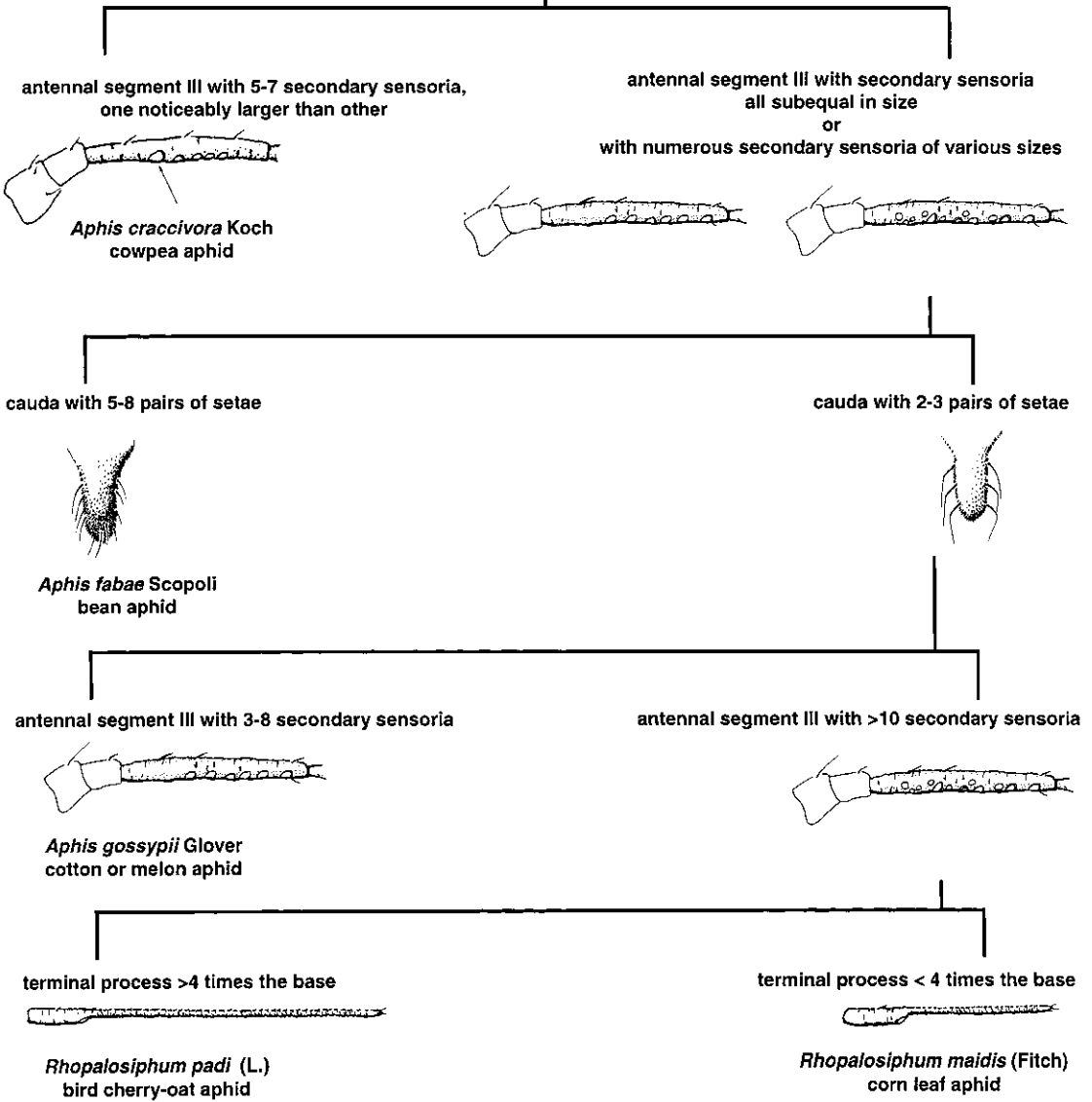


Fig. 7. Continued pictorial key to winged adult females of eight aphid species that commonly colonize corn in the United States and have antennal tubercles not developed.

drical, slightly constricted apically; approximately 4-6½ times as long as wide, longer than length of cauda. Cauda dark, with 2 pairs of lateral setae.

Hosts: Primary host of *R. padi* in North America is *Prunus virginiana*, however it also feeds on numerous species of Gramineae including corn.

U.S. distribution: Widespread.

Distribution in the world: Virtually worldwide.

Comments: *Rhopalosiphum padi* transmits more than 15 plant viruses, including barley yellow dwarf virus, maize leaf fleck virus, and sugarcane mosaic virus which are listed as affecting corn (Chan et al. 1991).

*Schizaphis graminum* (Rondani 1852)

Figs. 1, 4, 6

Synonymy:

*Aphis graminum* Rondani

\**Toxoptera graminum* (Rondani)

\*\**Schizaphis graminum* (Rondani)

ESA approved common name: greenbug.

Other common names: none.

Taxonomic characters: Wingless adult female.—

In life, body green to yellow green, dorsum often with median longitudinal stripe. Small to medium sized, body length 1.6-2.2 mm, elongate; body setae fine, inconspicuous. Antenna 6 segmented; tubercles not well developed; terminal process approximately  $3\frac{3}{4}$ - $4\frac{3}{4}$  times length of base of antennal segment VI; antennal segments III-V without secondary sensoria; longest setae on antennal segment III shorter than diameter of segment. Cornicle pale, sometimes apically dusky, approximately  $4\frac{1}{2}$ - $5\frac{3}{4}$  times as long as wide, longer than length of cauda. Cauda pale with 2-3 pairs of lateral setae.

Winged adult female.—In life, head and prothorax yellow brown, abdomen green to yellow green; forewing with media once branched, hind wing with 2 oblique veins; small to medium sized, body length 2.6-3.0 mm, body setae fine, inconspicuous. Antenna 6 segmented; tubercles not well developed; terminal process approximately  $3\frac{2}{3}$ - $4\frac{2}{3}$  times length of base of antennal segment VI; antennal segment III with 5-9 secondary sensoria, longest setae on antennal segment III shorter than diameter of segment; antennal segments IV-V without secondary sensoria. Cornicle pale, sometimes apically dusky, approximately  $3\frac{1}{2}$ -5 times as long as wide, longer than length of cauda. Cauda pale, with 2-3 pairs of lateral setae.

Hosts: Hosts are several species of Gramineae.

U.S. distribution: Widespread.

Distribution in the world: Africa, Central Asia, Central America, Japan, Korea, Middle East, Nepal, North America, Pakistan, South America, Taiwan, and Thailand.

Comments: *Schizaphis graminum* transmits 3 plant viruses, including barley yellow dwarf virus and sugarcane mosaic virus which are listed as affecting corn (Chan et al. 1991).

*Sipha flava* (Forbes 1884)

Fig. 1

Synonymy:

*Chaitophorus flava* Forbes

\**Sipha flava* (Forbes), in key

\*\**Sipha flava* (Forbes)

ESA approved common name: yellow sugarcane aphid.

Other common names: yellow sugar cane aphid.

Taxonomic characters: Wingless adult female.—In life, body yellow to green, often with paired intersegmental marking on dorsum. Small sized, body length 1.6-2.1mm, oval shaped, covered with stout, spinelike setae. Antennae 5 segmented; tubercles not well developed; terminal process approximately  $4\frac{1}{4}$ - $6\frac{1}{4}$  times length of base of antennal segment V; antennal segments III-IV without secondary sensoria; longest setae on antennal segment II longer than diameter of segment. Cornicle dusky, stout, approximately half as long as wide, shorter than length of cauda. Cauda pale, knobbed, with 3 pairs of lateral setae and 0-1 preapical setae.

Winged adult female.—In life, abdomen yellow with variable dorsal dark markings; forewing with media twice branched, hind wing with 2 oblique veins; small to medium sized, body length 1.5-2.0 mm, covered with stout, spine-like setae. Antennae 5 segmented; tubercles not well developed; terminal process approximately  $1\frac{3}{4}$ - $2\frac{3}{4}$  times length of base of antennal segment V; antennal segment III with 3-6 secondary sensoria, longest setae on antennal segment III longer than diameter of segment; antennal segment IV without secondary sensoria. Cornicle dusky, stout, approximately half as long as wide, shorter than length of cauda. Cauda pale, knobbed, with 3-4 pairs of lateral setae and 0-1 preapical setae.

Hosts: Hosts are several species of Gramineae.

U.S. distribution: Widespread.

Distribution in the world: Caribbean, Central America, North America, South America.

Comments: *Sipha flava* transmits sugarcane mosaic virus which is listed as affecting corn (Chan et al. 1991).

*Sitobion avenae* (Fabricius 1775)

Figs. 1-3

Synonymy:

*Aphis avenae* Fabricius

\**Macrosiphum granarium* (Kirby 1798)

\*\**Sitobion avenae* (Fabricius)

ESA approved common name: English grain aphid.

Other common names: grain aphid.

Taxonomic characters: Wingless adult female.—In life, body yellow green to red brown, dorsum often with faint intersegmental markings. Small to medium sized, body length 2.2-3.8 mm, elongate; body setae fine, inconspicuous. Antenna 6 segmented; tubercles well developed, inner faces divergent; terminal process approximately  $4\frac{1}{2}$ - $6\frac{1}{2}$  times length of base of antennal segment VI; antennal segments III-V without secondary sensoria; longest setae on antennal segment III shorter

than diameter of segment. Cornicle black, cylindrical and apically reticulated, approximately 4¼-5 times as long as wide, subequal to length of cauda. Cauda pale, with 2-5 pairs of lateral setae and 0-1 preapical setae.

Winged adult female.—In life, coloration similar to wingless adult female but intersegmental markings are more distinct; forewing with media twice branched, hind wing with 2 oblique veins; small to medium sized, body length 1.8-2.8 mm; body setae fine, inconspicuous. Antenna 6 segmented; tubercles well developed, inner faces divergent; terminal process approximately 5½-6½ times length of base of antennal segment VI; antennal segment III with 5-13 secondary sensoria, longest setae on antennal segment III shorter than

diameter of segment; antennal segments IV-V without secondary sensoria. Cornicle black, cylindrical and apically reticulated, approximately 4½-6½ times as long as wide, subequal to length of cauda. Cauda pale, with 3-5 pairs of lateral setae and 1-2 preapical setae.

Hosts: Several species of Gramineae, including all major cereals and pasture grasses.

U.S. distribution: Widespread.

Distribution in the world: Africa (in part), Central Asia, Central America, India, the Mediterranean, Middle East, Nepal, North America, Pakistan, and South America

Comments: *Sitobion avenae* transmits 4 plant viruses, including barley yellow dwarf virus which is listed as affecting corn (Chan et al. 1991).

KEY TO THE AERIAL FEEDING WINGLESS ADULT FEMALE APHIDS OF CORN IN THE UNITED STATES

With Reference to the Root-feeding, *Aphis maidiradicis* Forbes

- 1. Antenna 5-segmented; body setae stout, spine like; cornicle short, its length approximately half of its width . . . . . yellow sugarcane aphid, *Sipha flava* (Forbes)
- Antenna 6-segmented; body setae fine, inconspicuous; cornicle elongate, its length greater than half its width . . . . . 2
- 2(1) Antennal tubercles not well developed, not extending beyond frons or approximately even with frons (Fig. 1) . . . . . 3
- Antennal tubercles well developed, extending beyond frons (Fig. 1) . . . . . 10
- 3(2) Longest setae on antennal segment III longer than the diameter of the segment . . . . . 4
- Longest setae on antennal segment III shorter than the diameter of the segment . . . . . 5
- 4(3) Cornicle with apical constriction; cauda with 2 pairs of lateral setae . . . . . corn leaf aphid, *Rhopalosiphum maidis* (Fitch)
- Cornicle cylindrical, without apical constriction; cauda with 4-7 pairs of lateral setae . . . . . bean aphid, *Aphis fabae* Scopoli
- 5(3) Cornicle pale, sometimes apically dusky . . . . . greenbug, *Schizaphis graminum* (Rondani)
- Cornicle dark . . . . . 6
- 6(5) Cornicle length subequal to caudal length . . . . . corn root aphid, *Aphis maidiradicis* Forbes
- Cornicle longer than cauda . . . . . 7
- 7(6) Abdomen with large dark dorsal patch . . . . . cowpea aphid, *Aphis craccivora* Koch
- Abdomen without large dark dorsal patch, abdomen may have small dorsal marking or no markings . . . . . 8
- 8(7) Cornicle cylindrical with apical constriction; cauda dark . . . . . bird cherry-oat aphid, *Rhopalosiphum padi* (Linnaeus)
- Cornicle cylindrical or tapered without apical constriction; cauda pale or dusky . . . . . 9
- 9(8) Terminal process >4 times length of base; cauda pale to white . . . . . rusty plum aphid, *Hysteroneura setariae* (Thomas)
- Terminal process < 4 times length of base; cauda dusky to pale . . . . . cotton or melon aphid, *Aphis gossypii* Glover
- 10(2) Cornicle with polygonal reticulation . . . . . 11
- Cornicle without polygonal reticulation . . . . . 12
- 11(10) Cornicle black, 5 times as long as wide, subequal to length of cauda . . . . . English grain aphid, *Sitobion avenae* (Fabricius)
- Cornicle sometimes pale or becoming increasing dusky toward apex, 6½ times as long as wide, longer than the length of cauda . . . . . potato aphid, *Macrosiphum euphorbiae* (Thomas)



- 12(10) Antennal tubercles with inner faces convergent; cornicle with slight apical swelling and slight medial constriction . . . . . green peach aphid, *Myzus persicae* (Sulzer)  
 Antennal tubercle with inner face divergent; cornicle cylindrical with slight tapering to an apical flange . . . . . rose-grass aphid, *Metopolophium dirhodum* (Walker)

## KEY TO THE AERIAL FEEDING WINGED ADULT FEMALE APHIDS OF CORN IN THE UNITED STATES

With Reference to the Root-feeding, *Aphis maidiradicis* Forbes

1. Antenna 5-segmented; body setae stout, spine like; cornicle short, its length approximately half of its width . . . . . yellow sugarcane aphid, *Sipha flava* (Forbes)  
 Antenna 6-segmented; body setae fine, inconspicuous; cornicle elongate, its length greater than half its width . . . . . 2
- 2(1). Antennal tubercles not well developed, not extending beyond frons or approximately even with frons (Fig. 1) . . . . . 3  
 Antennal tubercles well developed, extending beyond frons (Fig. 1) . . . . . 10
- 3(2) Hind wing with one oblique vein; terminal process 6 times the length of the base of antennal segment VI. . . . . rusty plum aphid, *Hysterononeura setariae* (Thomas)  
 Hind wing with two oblique veins; terminal process < 6 times the length of the base of antennal segment VI. . . . . 4
- 4(3) Forewing with media once branched; cornicle pale sometimes dusky apically . . . . . greenbug, *Schizaphis graminum* (Rondani)  
 Forewing with media twice branched; cornicle black or dusky . . . . . 5
- 5(4) Cornicle length subequal to caudal length . . . . . corn root aphid, *Aphis maidiradicis* Forbes  
 Cornicle longer than cauda. . . . . 6
- 6(5) Longest setae on antennal segment III longer than diameter of segment; cauda with 5-8 pairs of lateral setae and 0-2 preapical setae . . . . . bean aphid, *Aphis fabae* Scopoli  
 Longest setae on antennal segment III shorter than diameter of segment; cauda with <5 pairs of lateral setae . . . . . 7
- 7(6) Antennal segment III with < 11 secondary sensoria and antennal segment IV without secondary sensoria . . . . . 8  
 Antennal segment III with 11 secondary sensoria and antennal segment IV with secondary sensoria . . . . . 9
- 8(7) Cornicle and cauda black . . . . . cowpea aphid, *Aphis craccivora* Koch  
 Cornicle dark and cauda pale to dusky . . . . . cotton or melon aphid, *Aphis gossypii* Glover
- 9(7) Terminal process < 3 times the length of the base . . . . . corn leaf aphid, *Rhopalosiphum maidis* (Fitch)  
 Terminal process > 4 times the length of the base . . . . . bird cherry-oat aphid, *Rhopalosiphum padi* (Linnaeus)
- 10(2) Cornicle with polygonal reticulation . . . . . 11  
 Cornicle without polygonal reticulation . . . . . 12
- 11(10) Cornicle black, 7 times as long as wide, subequal to length of cauda . . . . . English grain aphid, *Sitobion avenae* (Fabricius)  
 Cornicle sometimes pale or becoming increasing dusky toward apex, >7 times as long as wide, longer than length of cauda . . . . . potato aphid, *Macrosiphum euphorbiae* (Thomas)
- 12(10) Abdomen with large dark dorsal patch; cornicle with slight apical swelling and slight medial constriction . . . . . green peach aphid, *Myzus persicae* (Sulzer)  
 Abdomen without large dark dorsal patch; cornicle cylindrical with slight tapering to an apical flange . . . . . rose-grass aphid, *Metopolophium dirhodum* (Walker)

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SIX NEW SPECIES OF MOZENA FROM MEXICO  
(HETEROPTERA: COREIDAE: COREINAE: NEMATOPODINI)

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ABSTRACT

Six new species of *Mozena*, *M. atra*, *M. nogueirana*, *M. pardalota*, *M. perezae*, *M. preclara*, and *M. presigna*, collected in Mexico, are described; dorsal habitus, antennal segments, pronotum, abdomen in lateral view, paramere and male genital capsule of most of the species are illustrated.

Key Words: Insecta, Heteroptera, Coreidae, Nematopodini, *Mozena*, new species, Mexico

RESUMEN

Seis nuevas especies de *Mozena*, *M. atra*, *M. nogueirana*, *M. pardalota*, *M. perezae*, *M. preclara*, y *M. presigna*, colectadas en México son descritas; el cuerpo en vista dorsal, las antenas, el pronoto, el abdomen en vista lateral, los parámetros y la capsula genital del macho de la mayoría de las especies son ilustrados.

The genus *Mozena* Amyot and Serville (1843) is a large, complex group that ranges from the Southern United States, throughout Mexico, Cuba, and Central America to Northeastern South America including Colombia and Venezuela, with the greatest number of species being known from Mexico (Brailovsky 1999).

These are medium-sized to large coreids which are rather variable in color and size as well as in the general development of the humeral angles of the pronotum, development of the hind femur, and allometry of the scutellum.

Within the tribe Nematopodini, *Mozena* can be recognized by the triangular, flat, apically acute scutellum, the hind tibia only dilated ventrally, the humeral pronotal angles produced but not into sharp spines, the body usually longer than 20.00 mm, the tylus produced anterior to antennifers, the dorsal surface of the hind femur armed with tubercles or spines in three or more rows, the outer surface of the male hind coxa with large and robust tubercle (blunt in female without tubercle), and the mesosternum with a median sulcus between the bases of the fore coxae, with in which the rostrum usually lies. In the related genus *Piezogaster* Amyot and Serville the anterior third of the mesosternum lacks a median longitudinal sulcus (O'Shea 1980).

The genus is usually associated with Leguminosae, and has been collected on Mesquite (*Prosopis* spp.), sweet acacia (*Acacia* spp.), and *Schrankia uncinata* (Ward et al. 1977). The biology of most species is poorly known. Brailovsky et al. (1995) studied the immature stages, life history, and biological aspects of *Mozena lunata* (Burmeister).

Previous to this paper, 15 species and one subspecies of *Mozena* were known in Mexico (*M. affinis* (Dallas), *M. arizonensis* Ruckes, *M. brunnicornis*

(H. S.), *M. buenoi* Hussey, *M. gaumeri* Distant, *M. hector* Van Duzee, *M. lineolata* (H.S.), *M. lunata* (Burmeister), *M. lunata rufescens* Ruckes, *M. lurida* (Dallas), *M. lutea* (H.S.), *M. nestor* (Stål), *M. pallisteri* Ruckes, *M. rufula* Van Duzee, *M. scrupulosa* (Stål), and *M. ventralis* (Mayr). This contribution adds six new species.

The following abbreviations are used to identify institutions where types are deposited or which generously lent material for this paper: AMNH: American Museum of Natural History, New York; BMNH: The Natural History Museum, London, England; BYU: Brigham Young University, Monte L. Bean Life Science Museum, Provo, UT; CAS: California Academy of Sciences, San Francisco; CMN: Carnegie Museum of Natural History, Pittsburgh; CUIC: Cornell University, Insect Collection, Ithaca, NY; FNS: Forschungsinstitut und Naturmuseum Senckenberg, Germany; FMNH: Field Museum Natural History, Chicago, IL; FSCA: Florida State Collection of Arthropods, Gainesville, FL; LACM: Los Angeles County Museum, CA; NMW: Naturhistorisches Museum, Wien; NRE: Naturhistoriska Riksmuseet, Stockholm; TAMU: Texas A&M University, College Station; UCB: University of California, Berkeley; UCD: University of California, Davis; UGA: University of Georgia, Athens, GA; UNAM: Instituto de Biología, Universidad Nacional Autónoma de México; USNM: National Museum of Natural History, Smithsonian Institution, Washington, DC.

All measurements are given in millimeters.

*Mozena atra* Brailovsky and Barrera, **New Species**

Figs. 13, 26

**Description.** Male (holotype). Dorsal coloration: head including antennal segments I to IV or-

ange; pronotum bright chestnut orange with outer margin of humeral angles black, and small tubercles dark yellow; scutellum creamy yellow, with chestnut orange spot on basal and middle third; clavus and corium chestnut orange, with middle third of costal margin whitish; hemelytral membrane dark amber-like, with veins darker; connexival segments III to VII reddish brown, with anterior third yellow, and posterior border and spines black; dorsal abdominal segments light orange yellow. Ventral coloration. Bright orange with following areas yellow: acetabulae, tubercles nearest each acetabulae, posterior margin of propleura, and anterior third of pleural abdominal sterna III to VII; rostral segments orange with apex of IV black; propleura, mesopleura, and metapleura with clearly creamy yellow vittae obliquely directed; anterior and posterior lobe of metathoracic peritreme black; fore and middle legs bright orange, with coxae tinged with bright red; hind leg with coxa and trochanter bright orange red, femur dark orange with ventral spines black, tibia with inner surface bright black, and outer surface and distal third bright red to orange red, tarsus bright orange; creamy yellow vittae along each side of abdominal sterna broken into two sections on each sternite, the anterior one small and more or less discoidal, and posterior one bigger and enlarged posteriorly; rim of abdominal spiracle creamy yellow to yellow. Anterolateral margins of pronotum obliquely straight, uniformly dentate; humeral angles produced laterally, each margin dentate, and apically acute; posterolateral and posterior margin straight and smooth; calli behind with two short small tubercles laterad to midline (Fig. 13). Legs. External face of hind coxa with a large and robust tubercle; hind femur medially incrassate, with three rows of tubercles on dorsal surface, ventrally with two rows of lateral spines; hind tibiae conspicuously dilated. Scutellum longer than wide. Abdomen slightly dilated, wider than hemelytra, maximum width less than maximum width of pronotum across humeral angles.

**Genitalia.** Posteroventral edge of genital capsule simple, straight.

Coloration of females similar to holotype. Connexival segments VIII and IX black, with anterior third yellow to orange; dorsal abdominal segments VIII and IX orange yellow with posterior margin black; genital plates orange with outer border of paratergite VIII and IX black. Scutellum wider than long; external face of hind coxae blunt without tubercles; hind tibiae scarcely dilated. Plica located near the posterior border of abdominal sternite VI.

Body surface rather dull, seldom shiny, almost glabrous; bristle-like setae of antennal segments, legs, and body surface scattered, short and appressed; antennal segment I with erect setae; fore and middle leg with erect to suberect setae inter-

mixed with appressed setae; posterior third of pronotal disc, scutellum, clavus, corium, acetabulae, and posterior margin of prothorax, mesothorax, and metathorax sparsely to strongly punctate; head, anterior third of pronotal disc, calli, connexival segments, prosternum, mesosternum and metasternum, upper anterior third of propleura and metapleura, and abdominal sterna impunctate; posterior third of pronotal disc with scattered with minute tubercles; thorax with minute tubercles near upper margin of each acetabulum; genital segments of both sexes minutely punctate and tuberculate.

**Variation.** 1: Posterior region of scutellum including apex black, with anterolateral margins creamy yellow, and basal and middle third chestnut orange. 2: Creamy yellow vitta along each side of abdominal sterna III to VII complete and characteristically enlarged posteriorly.

**Measurements.** ♂ holotype first, then ♀: Head length 1.90, 1.80, width across eyes 2.40, 2.35, interocular space 1.55, 1.45, interocellar space 0.88, 0.79, preocular distance 1.35, 1.20; length of antennal segments: I, 3.85, 3.40, II, 3.80, 3.35, III, 2.90, 2.70, IV, 3.25, 3.00. Pronotum: Total length 5.68, 5.36, width across frontal angles 2.70, 2.75, width across humeral angles 10.25, 9.85. Scutellar length 3.25, 3.15, width 3.20, 3.25. Maximum width of abdomen 9.50, 9.70. Total body length 23.83, 23.20.

**Holotype.** ♂ Mexico: Sinaloa, 7mi S Culiacan, 23-VIII-1960 (R. L. Westcott). (CUIC). Paratypes. 1 ♂, 2 ♀: Same data as holotype. (CUIC, UNAM).

**Etymology.** From the Latin, *atra*, black, in reference to the color of the anterior and posterior lobes of the metathoracic peritreme.

**Discussion.** The laterally produced pronotal shape is somewhat similar to that of *M. buenoi* Hussey (Figs. 13, 14). However *M. atra* can be easily separated by having the anterior and posterior lobe of metathoracic peritreme entirely black, the antennal segments orange, the bristle-like setae of antennal segment I appressed, and intermixed with erect setae, and the rims of the abdominal spiracles creamy yellow to yellow. In *M. buenoi* the anterior lobe of the metathoracic peritreme is yellow with its inner margin narrowly black, the antennal segments are not entirely orange, antennal segment I is shorter (♂ 3.20, ♀ 2.75, against ♂ 3.85, ♀ 3.40) with bristle like setae appressed, and the rims of abdominal spiracles black to brown.

*Mozena nogueirana* Brailovsky and Barrera,

**New Species**

Figs. 6, 8, 17, 23

**Description.** Male (holotype). Dorsal coloration: head dark yellow hazel, with tylus, midline stripe running from tylus to apex, and upper margin of antenniferous tubercles reddish brown; an-

tennal segment I and II with dorsal face reddish brown and ventral face orange yellow to bright orange; segments III and IV bright orange; pronotum, scutellum, clavus and corium chestnut, with following areas creamy yellow to light yellow: few scattered dots on pronotal disc, clavus and corium, apex of scutellum and claval and corial veins; humeral angles black; hemelytral membrane amber-like with basal angle and veins brown; connexival segments II and III yellow, IV yellow with upper margin of posterior third light brown, and V to VII with anterior half yellow and posterior half including the spines brown; dorsal abdominal segments II to V yellow, VI yellow with two light brown discoidal spots near to posterior margin, and VII black with lateral margins light orange brown. Ventral coloration. Included buccula, rostral segments (apex of IV black), and coxae (outer margin chestnut brown) chestnut yellow; humeral angles, tubercles of thoracic mesopleura, and spines of pleural abdominal segments IV to VI black; anterior lobe of metathoracic peritreme black, with central area dark orange, and posterior lobe black; trochanters reddish brown with upper third chestnut yellow; fore and middle femora chestnut orange yellow, and hind femur chestnut orange yellow with dorsal tubercles light yellow, ventral spines reddish brown, and a wide stripe reddish brown running throughout dorsal surface; fore and middle tibiae with dorsal face reddish brown, and ventrally chestnut orange yellow, and hind tibiae dorsally reddish brown, and ventrally chestnut orange yellow with inner spines reddish brown; basal tarsi chestnut orange yellow, and middle and hind tarsi chestnut orange yellow to reddish brown; propleura, mesopleura and metapleura with the creamy yellow vitta poorly developed, discontinuous, those on metapleura sometimes lacking; creamy yellow to light orange vittae along each side of abdominal sterna III to VII characteristically enlarged posteriorly; rim of abdominal spiracles creamy yellow to whitish. Anterolateral margins of pronotum obliquely straight, with few tubercles on anterior half; humeral angles exposed, slightly elevated, small to medium sized, hemispheric, directed upward, with the tips slightly or conspicuously curved inwards, and apically acute; posterolateral margins straight, with upper half sparsely dentate, and inner half smooth; posterior margin entire, straight, sometimes rounded; calli lacking tubercles behind (Fig. 17). Legs. External face of hind coxa with large and robust tubercle; hind femur with three rows of tubercles on dorsal surface, and ventrally with two rows of lateral spines; hind tibiae medially dilated on male, and cylindrical and slender on female. Scutellum wider than long. Abdomen slightly dilated, slightly wider than hemelytra, with maximum width less than maximum width of pronotum across humeral angles.

Genitalia. Posteroventral edge of genital capsule simple, with a weakly concavity (Fig. 6). Paramere as in Fig. 8.

Coloration of females similar to male. Connexival segment VIII black with anterior third yellow, IX entirely black; dorsal abdominal segments II to VII black with lateral margins usually yellow; segments VIII and IX entirely black; abdominal sterna bright yellow, sometimes tinged with bright reddish orange; genital plate chestnut orange with external face of paratergite IX brown. Plica close to posterior border of abdominal sternite VI.

Body surface rather dull, seldom shiny. Head, pronotum, scutellum, clavus, corium, and thorax clothed with long erect to suberect bristle-like setae; bristle-like setae of antennal segments I to III, legs, and abdominal sterna usually short, appressed, intermixed with sparse suberect setae; posterior third of pronotal disc, clavus, and corium strongly punctate; head, anterior third of pronotal disc, calli, thorax, and abdominal sterna almost impunctate; scutellum with few scattered punctures; posterior third of pronotal disc, clavus and corium with few irregular scattered, and small tubercles or callosities; propleura, mesopleura and metapleura without tubercles; genital segments of both sexes minutely tuberculate and punctate.

**Measurements.** ♂ holotype first, then ♀: Head length 1.85, 1.75, width across eyes 2.35, 2.25, interocular space 1.44, 1.35, interocellar space 0.68, 0.62; length of antennal segments: I, 4.25, 3.60, II, 4.25, 3.50, III, 3.15, 2.60, IV, 3.30, 2.75. Pronotum: Total length 4.84, 4.35, width across frontal angles 2.25, 2.05, width across humeral angles 6.90, 6.80. Scutellar length 2.70, 2.40, width 2.85, 2.60. Maximum width of abdomen 5.32, 5.38. Total body length 20.32, 18.83.

Holotype. ♂ Mexico: Jalisco, Alista, 1100m, 16-VI-1996 (G. Nogueira). (UNAM). Paratypes. 1 ♀, same data as holotype. (UNAM). 1 ♀ Mexico: Jalisco, Copala, 1170m, 17-IX-1995 (G. Nogueira). (UNAM). 1 ♀ Mexico: Michoacan, 48 km S Nueva Italia, 8-VIII-1978 (Plitt and Schaffner). (TAMU). 1 ♀ Mexico: Michoacán, 4.8 km N. Capirio, 12-VII-1981 (E. M. Fisher). (UNAM). 2 ♂, 2 ♀ Mexico: Michoacán, Apatzingan, 1200', 12-VIII-1941 (H. Hoogstraal). (FSCA). 1 ♀ Mexico: Guerrero, Tecpan de Galeana, 19-VIII-1971 (H. Brailovsky). (UNAM). 1 ♂ Mexico: Puebla, Acatlan, 26-VII-1942 (F. Islas). (UNAM). 2 ♀ Mexico: Puebla, Matamoros, 28-VII-1942 (F. Islas). (UNAM). 1 ♀ Mexico: Oaxaca, 17km, SE from Huajuapán de León, 29-VI-1996 (H. Brailovsky and E. Barrera). (UNAM).

**Etymology.** Named for Guillermo Nogueira, our friend and excellent collector.

**Discussion.** *Mozena nogueirana* resembles *M. buenoi* Hussey by having the maximum width of abdomen less than maximum width of pronotum across humeral angles, the antennal segment I without erect setae (Fig. 2), and the

creamy yellow to orange vittae along each side of abdominal sterna III to VI enlarged posteriorly (Fig. 4). In *M. buenoi* the humeral pronotal angles are produced laterally and the antennal segment I is robust and shorter than 3.30. In the new species the humeral angles are moderately raised, hemispheric, and the antennal segment I is slender and longer than 3.50.

*Mozena pardalota* Brailovsky and Barrera,

**New Species**

Figs. 9, 18, 24

**Description.** Male (holotype). Dorsal coloration: orange yellow with a olivaceous tinge; antennal segments I to III orange yellow, with apical joint reddish brown, and IV yellowish with a brownish tinge; pronotal disc at middle third with one dark orange longitudinal stripe running from anterior margin to near posterior margin (best seen with naked eye); apex and lateral margins of scutellum light yellow; hemelytral membrane amber-like; connexival segments light orange with inner third of anterior margin yellow, upper margin yellow with one to four black spots; dorsal abdominal segments black, with lateral margins of V and VI bright orange yellow, and VII bright orange yellow with two anterior spots black and lateral to midline. Ventral coloration. Including rostral segments (apex of IV black), legs, and genital capsule bright orange yellow; spines and tubercles of femora light yellow; anterior lobe of metathoracic peritreme yellow with outer margin narrowly black, and posterior lobe black; propleura, mesopleura, and metapleura with the creamy yellow vitta well developed, and obliquely straight; acetabulae, thoracic tubercles, and posterior margin of propleura, mesopleura, and metapleura creamy yellow; upper margin of pleural abdominal sterna light yellow; creamy yellow vittae along each side of abdominal sterna III to VII uniformly straight, not enlarged posteriorly; rim of abdominal spiracles yellow. Anterolateral margins of pronotum obliquely straight, conspicuously dentate; humeral angles slightly exposed, subacute, directed laterally, almost at right angles to longitudinal axis of body, each border dentate; posterolateral and posterior margin straight, and entire; calli behind with two tubercles lateral to midline (Fig. 18). Legs. External face of hind coxa with a large and robust tubercle; fore and middle femora with three rows and hind femur with four rows of tubercles on dorsal surface, ventrally with two rows of lateral spines; hind tibia slightly incrassate near the middle third. Scutellum wider than long. Abdomen dilated, wider than hemelytra, with maximum width wider than maximum width of pronotum across humeral angles.

Genitalia. Posteroventral edge of genital capsule simple, transversely straight. Paramere as in Fig. 9.

Coloration of females similar to holotype. Connexival segments dark orange with tubercles yellow, and upper margin yellow with one to four black spots; dorsal abdominal segments bright orange; gonocoxae I bright orange, and paratergite VIII and IX orange yellow. External face of hind femur blunt, without tubercle; hind tibia cylindrical. Plica close to posterior margin of abdominal sternite VI.

Body surface rather dull, seldom shiny, and almost glabrous; antennal segment I clothed with erect setae; antennal segments II to IV clothed with appressed setae; femora and tibiae with erect to suberect setae; calli behind with six tubercles in transverse row; posterior third of pronotal disc, scutellum, clavus, corium, acetabulae, posterior margin of propleura, mesopleura and thoracic metapleura punctate; head, calli, connexival segments, and abdominal sterna impunctate; connexival segments scarcely tuberculate; genital segments of both sexes minutely tuberculate and punctate.

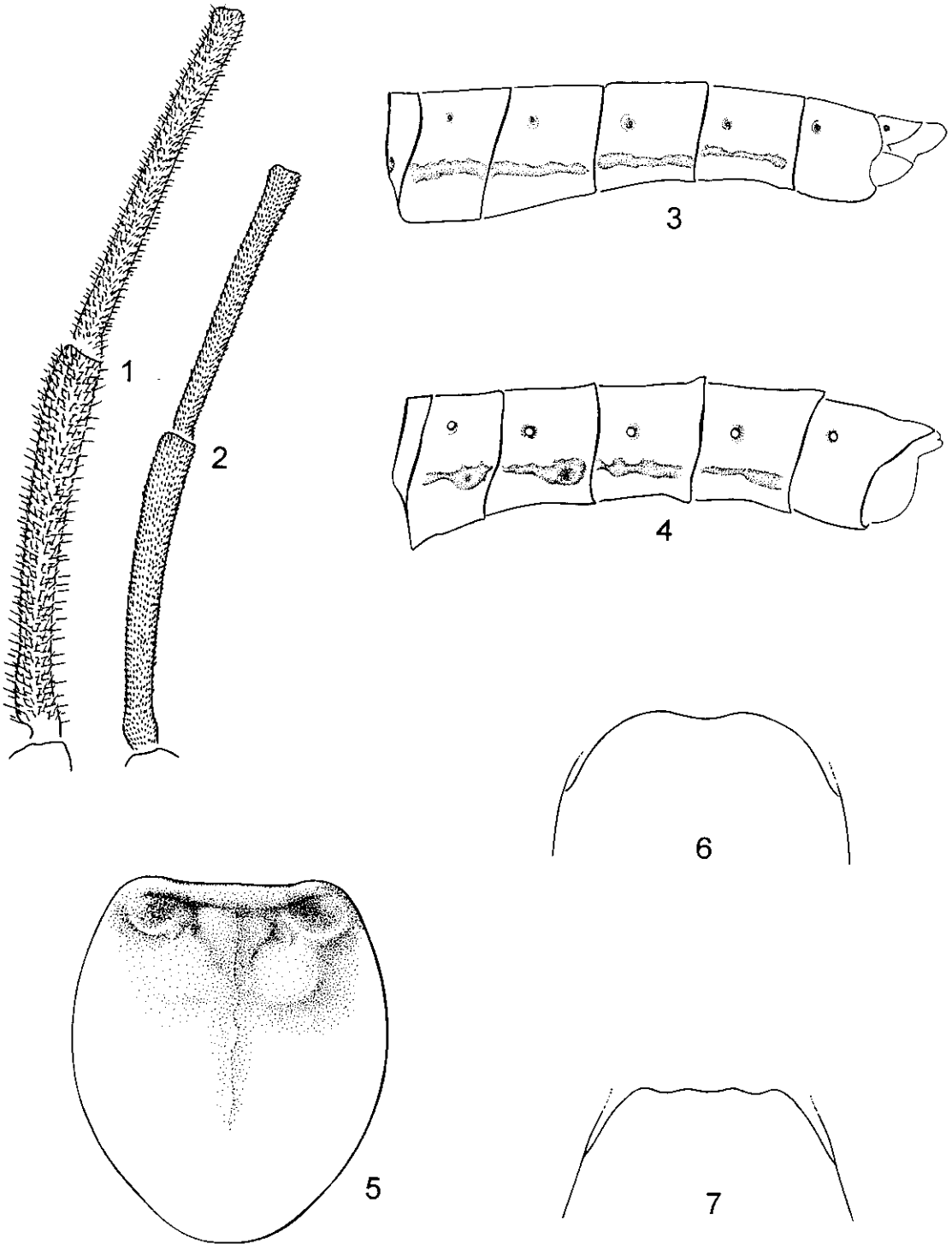
Variation. 1: Body light yellow, with antennal segments, clavus, and corium bright orange, and upper margin of connexival segments with one to four black spots.

**Measurements.** ♂ holotype first, then ♀: Head length 1.57, 1.50, width across eyes 1.95, 1.95, interocular space 1.20, 0.98, interocellar space 0.65, 0.65, preocular distance 1.10, 1.00; length of antennal segments: I, 2.75, 2.20, II, 2.65, 2.10, III, 2.20, 1.65, IV, 2.75, 2.20. Pronotum: Total length 4.35, 4.45; width across frontal angles 2.55, 2.35; width across humeral angles 7.00, 7.55. Scutellar length 2.55, 2.55, width 2.75, 2.85. Maximum width of abdomen 8.45, 8.85. Total body length 17.30, 18.50.

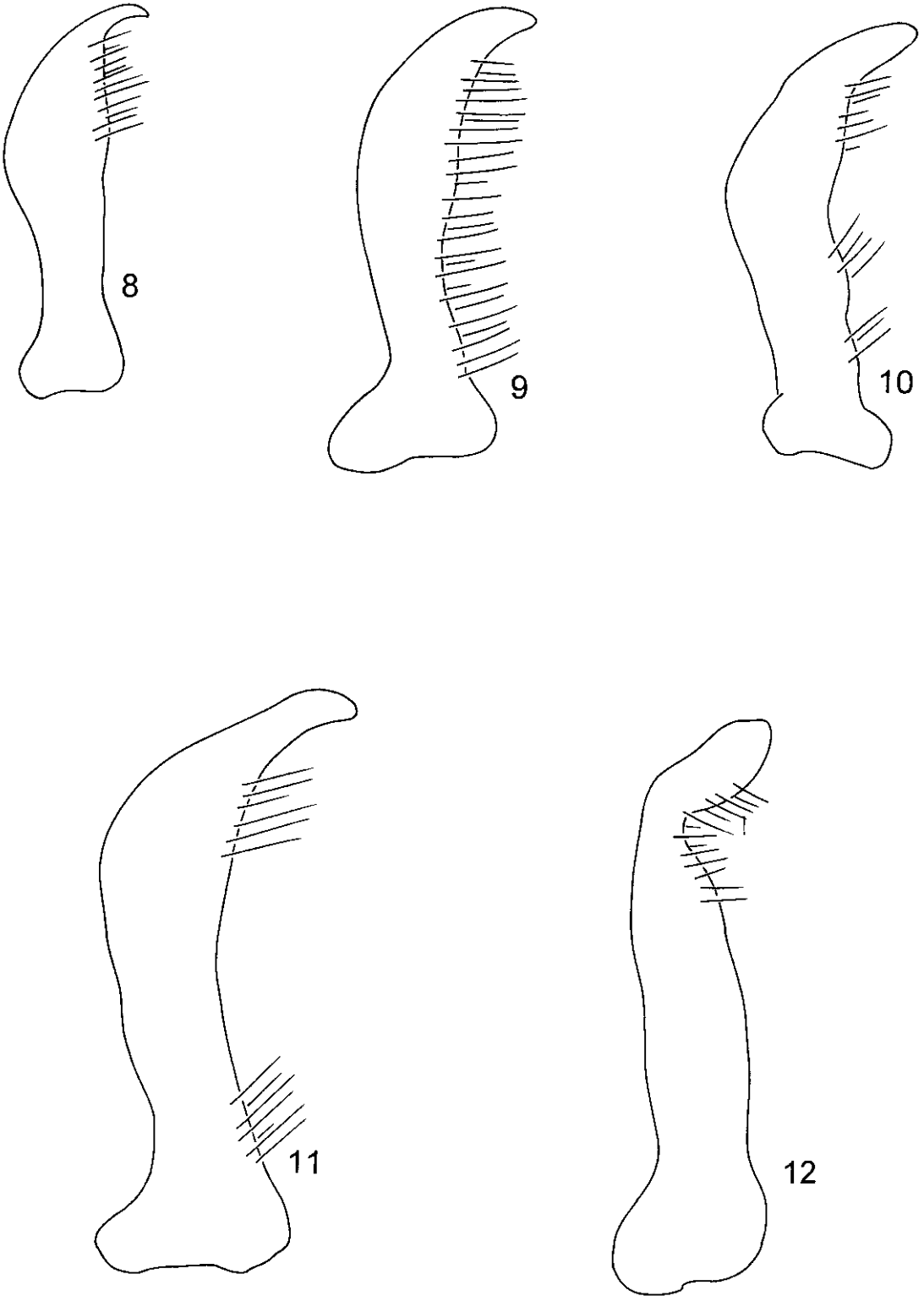
Holotype. ♂ Mexico: Jalisco, km 4 El Tuito-Tehuamixtle, 23-XI-1991 (J. Villa and E. Ramirez). (UNAM). Paratypes. 2 ♂, 1 ♀: Same data as holotype. 1 ♂ Mexico: Jalisco, Chamela, Tomatlán, 7-IX-1979 (H. Brailovsky). (UNAM). 2 ♀ Mexico: Nayarit, 44 mi NW Tepic, 30-VIII-1971 (J. L. Petty). (BYU). 1 ♂ Mexico: Nayarit, 68 mi SE Tepic, 31-VIII-1971 (J. L. Petty). (BYU).

**Etymology.** The specific name is from the Greek *pardalota* meaning "spotted like a leopard" in reference to its spotted color pattern on upper margin of connexival segments.

**Discussion.** The pronotal shape including the humeral angles is somewhat similar to those of *M. buenoi* Hussey (Figs. 14, 18). *M. pardalota* is recognized by having the maximum width of abdomen wider than maximum width of pronotal disc across humeral angles, antennal segment I orange yellow, slender, and clothed with erect setae (Fig. 1), calli behind with two lateral tubercles, apical margin of corium unicolorous in reference to corial disc, and upper margin of connexival segments yellow with one to four black spots. In *M. buenoi* the maximum width of abdomen is less

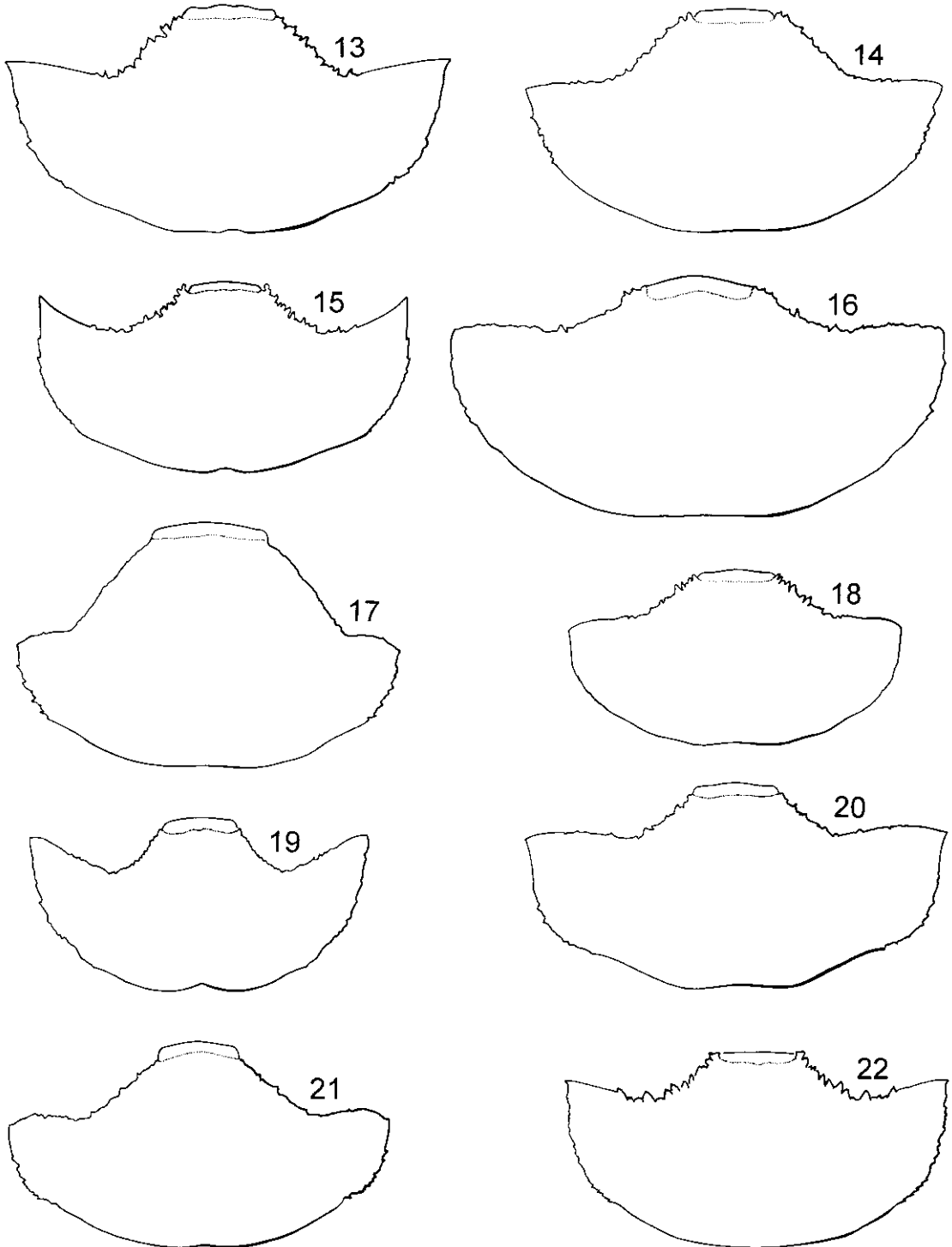


Figs. 1-7. *Mozena* spp. Figs. 1-2. Antennal segments I-II. 1, *M. nestor* (Stål). 2, *M. lunata* (Burmeister). Figs. 3-4. Abdomen in lateral view. 3, *M. perezae* Brailovsky and Barrera, **New Species**. 4, *M. presigna* Brailovsky and Barrera, **New Species**. Figs. 5-7. Caudal view of male genital capsule. 5, *M. perezae* Brailovsky and Barrera, **New Species**. 6, *M. nogueirana* Brailovsky and Barrera, **New Species**. 7, *M. presigna* Brailovsky and Barrera, **New Species**.



Figs. 8-12. Paramere of *Mozena* spp. 8, *M. nogueirana* Brailovsky and Barrera, **New Species**. 9, *M. pardalota* Brailovsky and Barrera, **New Species**. 10, *M. perezae* Brailovsky and Barrera, **New Species**. 11, *M. preclara* Brailovsky and Barrera, **New Species**. 12, *M. presigna* Brailovsky and Barrera, **New Species**.





Figs. 13-22. Pronotum of *Mozena* spp. 13, *M. atra* Brailovsky and Barrera, **New Species**. 14, *M. buenoi* Hussey. 15, *M. lunata* (Burmeister). 16, *M. nestor* (Stål). 17, *M. nogueirana* Brailovsky and Barrera, **New Species**. 18, *M. pardalota* Brailovsky and Barrera, **New Species**. 19, *M. perezae* Brailovsky and Barrera, **New Species**. 20, *M. preclara* Brailovsky and Barrera, **New Species**. 21, *M. presigna* Brailovsky and Barrera, **New Species**. 22, *M. lunata rufescens* Ruckes.

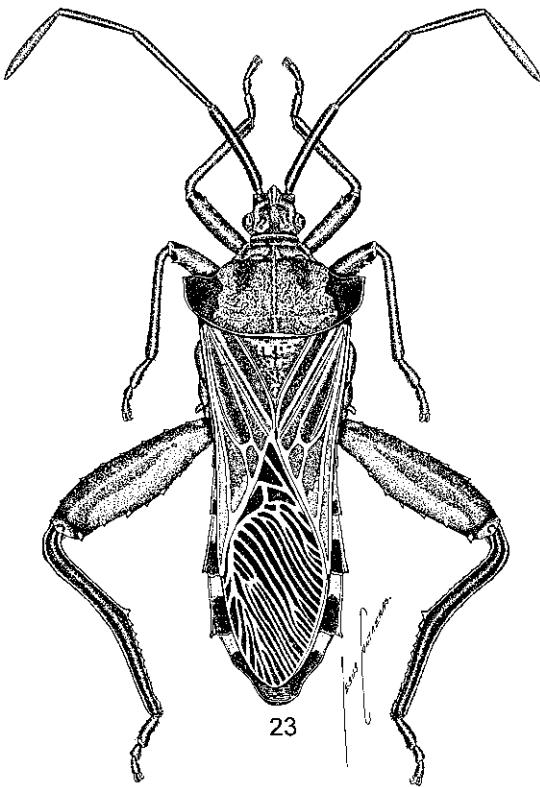


Fig. 23. *Mozena nogueirana* Brailovsky and Barrera, **New Species**, dorsal view (male).

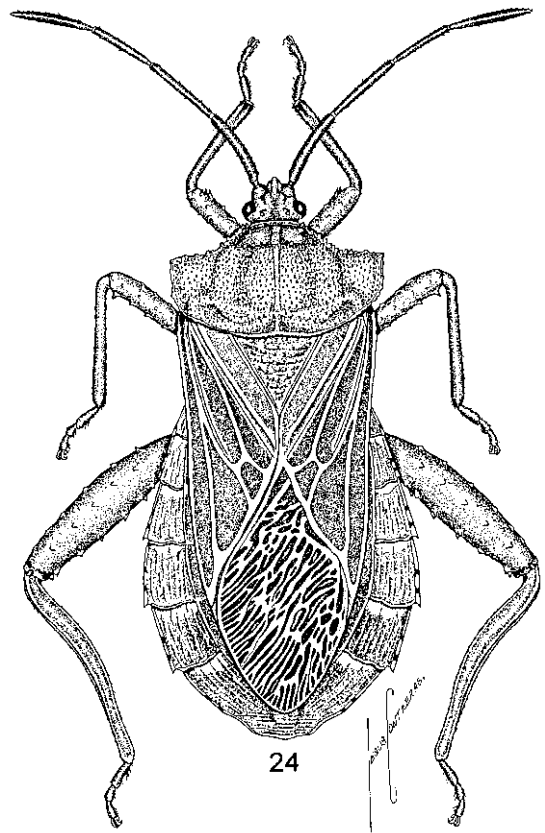


Fig. 24. *Mozena pardalota* Brailovsky and Barrera, **New Species**, dorsal view (male).

than maximum width of pronotal disc across humeral angles, antennal segment I dorsally black, ventrally orange, robust, without or with few erect setae, calli behind lacking tubercles or scarcely developed, apical margin of corium light yellow to ivory and contrasting with corial disc, and upper margin of connexival segments with anterior half yellow, and posterior half brown to orange, without black discoidal spots.

In *M. rufula* Van Duzee (Fig. 29) the body and antennal segments I to IV are orange, but the maximum width of abdomen is less than maximum width of pronotal disc across humeral angles.

*Mozena perezae* Brailovsky and Barrera, **New Species**  
Figs. 3, 5, 10, 19, 28

**Description.** Male (holotype). Dorsal coloration: head dark orange; tylus and space between eye and antenniferous tubercle black; antennal segment I dorsally black, and ventrally orange except basal third black; segments II to IV bright orange; calli dark orange; collar and posterior lobe dark yellow with callosities and two prominent tubercles behind calli light yellow; humeral angles black; scutellum light yellow with irregular black dots, and punctures chestnut orange; clavus

and corium yellow with olivaceous tinge, punctures chestnut orange, and anterior half of costal margin of corium brown; hemelytral membrane amber-like with basal angle and veins darker; connexival segments III to VII black, with anterior third light yellow; dorsal abdominal segments black with scent gland scars IV-V and V-VI yellow. Ventral coloration. Head including buccula black, with the area neighbouring eyes yellow; rostral segments chestnut red; thorax bright orange with anterior third of propleura, each margin of humeral expansion, and great portion of meta-acetabulae black to reddish brown; propleura, mesopleura and metapleura with conspicuous creamy yellow vittae; upper margin of metapleura posteriorly yellow; anterior lobe of metathoracic peritreme yellow and posterior lobe black; coxae bright orange with inner third black to reddish brown; trochanters bright orange; fore and middle femora bright orange with apical third black; hind femur bright orange with apical third, tubercles and spines black to reddish brown; fore and middle tibiae bright orange, with sulcate region black to reddish brown; hind tibia with dorsal face black, and ventrally orange; tarsi

chestnut orange with reddish brown dots; laevigate creamy yellow and polished line along each side of abdominal sterna III to VII uniformly straight (Fig. 3); rim of abdominal spiracles black; pleural abdominal segments III to VII black with anterior third yellow; abdominal sterna bright orange; genital capsule black with lateral margin almost orange. Anterolateral margins of pronotum obliquely straight, with few tubercles on anterior half; humeral angles exposed, elevated, strongly developed, elongate, produced laterally, out and slightly upwards directed, and apically subacute; posterolateral margins straight, and smooth; posterior margin sinuate, smooth, with shallow concavity at middle third; calli behind with two prominent tubercles lateral to midline (Fig. 19). Legs. External face of hind coxa with blunt, inconspicuous tubercle; hind femur with three rows of tubercles on dorsal surface, ventrally with two rows of lateral spines; hind tibia conspicuously dilated. Scutellum wider than long. Abdomen slightly dilated, wider than hemelytra, with maximum width less than maximum width of pronotum, across humeral angles.

Genitalia. Posteroventral edge of genital capsule simple, with shallow medial plate emargination (Fig. 5). Paramere as in Fig. 10.

Coloration of females similar to male. Head black with area around ocelli orange; collar and calli black; scutellum light yellow, with black squarish mark on basal third; clavus yellow with olivaceous tinge, and black punctures along anal suture; connexival segments VIII and IX, and abdominal segments VIII and IX black; thorax black, with mesosternum and metasternum orange yellow, with following areas yellow: one irregular spot on propleura, mesopleura and metapleura, anterior tubercle of mesopleura, and posterior third of upper margin of metapleura; anterior and posterior lobe of metathoracic peritreme yellow; legs black with dark orange trochanters; laevigate creamy yellow polished line running from III to VI abdominal sterna uniformly straight; genital plates black. External face of hind coxa blunt, with inconspicuous tubercle; hind tibia conspicuously dilated. Plica located far from posterior border of abdominal sternite VI.

Body surface rather dull, almost entirely glabrous; bristle-like setae of antennal segments I to III appressed; bristle-like setae of femora, and hind tibiae scattered, appressed, and on fore and middle tibiae densely appressed or suberect; head, anterior third of pronotal disc, calli, thorax (acetabulae usually punctate) connexival segments and abdominal sterna impunctate; posterior third of pronotal disc, clavus, and corium strongly punctate, and each puncture with short decumbent bristle-like setae; scutellum with few scattered punctures; posterior third of pronotal disc with irregular, small tubercles or callosities; propleura, mesopleura and metapleura without

tubercles; genital segments of both sexes minutely tuberculate and punctate.

Variation. 1: Head, pronotum, and scutellum mostly black. 2: Head, antennal segments I to IV, pronotum (humeral angles black), scutellum, clavus, corium, thorax, fore and middle legs, and abdomen bright orange. 3: Anterior and posterior lobe of metathoracic peritreme black. 4: Trochanters black. 5: Pleural abdominal sterna III to VII black with small yellow spot on anterior third of sterna V to VI.

**Measurements.** ♂ holotype first, then ♀: Head length 1.65, 1.65, width across eyes 2.07, 2.10, interocular space 1.28, 1.25, interocellar space 0.74, 0.74, preocular distance 1.12, 1.15; length antennal segments: I, 4.30, 3.85, II, 3.50, 3.30, III, 2.55, 2.35, IV, 3.15, 3.00. Pronotum: Total length 4.30, 4.30, width across frontal angles 2.20, 2.15, width across humeral angles 8.60, 8.65. Scutellar length 2.45, 2.50, width 2.55, 2.60. Maximum width of abdomen 5.16, 5.23. Total body length 22.20, 20.78.

Holotype ♂ Mexico: Chiapas, Aguacero, 16km W from Ocozocoautla, 15-VI-1987 (D. B. Thomas). (UNAM). Paratypes. 1 ♀ Mexico: Chiapas, La Trinidad, 1-X-1983 (E. Barrera). (UNAM). 1 ♀ Mexico: Chiapas, Tuxtla Gutierrez, 10-VIII-1983 (E. Barrera). (UNAM). 1 ♀ Mexico: Chiapas, Reserva El Ocote, 2-10-XII-1993 (G. Ortega León, E. Barrera, and A. Casasola). (UNAM). 1 ♂ Mexico: Chiapas, Municipio Ocozocoautla, El Aguacero, 2200', 6-VIII-1990 (J. C. Schaffner). (TAMU).

**Etymology.** Named for Olga Leticia Perez Ramirez, friend for many years.

**Discussion.** *Mozena perezae* is related to *M. nogueirana*, sharing with it the following characters: maximum width of abdomen less than maximum width of pronotum across humeral angles, and antennal segment I without erect setae (Fig. 2). In *M. perezae* the body is slender, and narrowed, the humeral angles strongly developed (Fig. 19), and the creamy yellow to orange vitta along each side of abdominal sterna III to VI not enlarged posteriorly (Fig. 3).

In *M. lunata* (Burmeister) and *M. lunata rufescens* Ruckes the humeral angles are not conspicuously developed (Figs. 15, 22), and the connexival segments II and III are yellow and not black which is characteristic of *M. perezae*.

*Mozena preclara* Brailovsky and Barrera, **New Species**  
Figs. 11, 20, 25

**Description.** Male (holotype). Dorsal coloration: head dark orange with the interocular space reddish brown; antennal segment I black dorsally, orange ventrally, segments II and III orange, and IV reddish brown with basal joint orange; pronotum dark chestnut orange with humeral angles black; scutellum light yellow, and basally with dark chestnut orange central area; clavus and

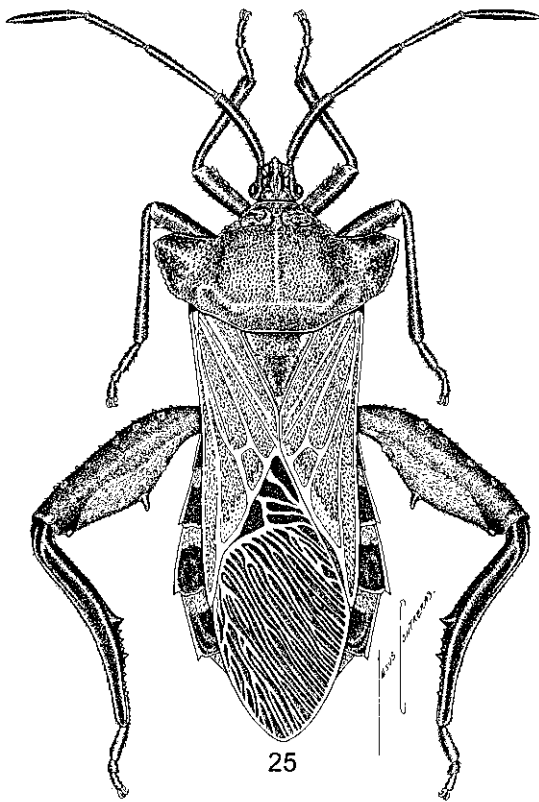


Fig. 25. *Mozena preclara* Brailovsky and Barrera, **New Species**, dorsal view (male).

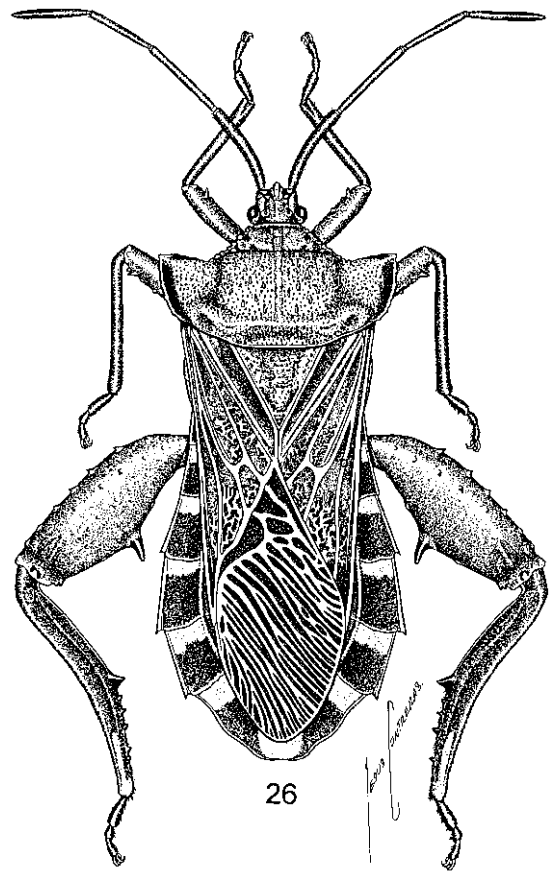


Fig. 26. *Mozena atra* Brailovsky and Barrera, **New Species**, dorsal view (male).

corium chestnut orange with veins, apical margin, anterior third of costal margin, and small tubercles light yellow to dark orange; hemelytral membrane dark amber-like, with basal angle and veins brown; connexival segments reddish brown with anterior third yellow; dorsal abdominal segments bright orange, with posterior margin of segment VII black. Ventral coloration. Bright chestnut orange; rostral segments orange with apical third of IV black; anterior lobe of metathoracic peritreme black, with central area orange, and posterior lobe black; propleura, mesopleura and metapleura with creamy yellow vitta slender, obliquely straight; acetabulae, few tubercles scattered on thoracic pleura, upper tubercle of anterior angle of mesopleura, and posterior angle of metapleura creamy yellow; legs orange with following areas black: fore and middle femora dorsally with slender longitudinal stripe, hind femur with tubercles, spines and dorsal surface with wide longitudinal stripe, and dorsal surface of hind tibia (apical third orange); creamy yellow vitta along each side of abdominal sterna III to VII uniformly broad; rim of abdominal spiracles yellow; genital capsule orange. Anterolateral margins of pronotum obliquely straight, uni-

formly dentate; humeral angles exposed, relatively quadrate, produced laterally, directed upwards, each margin dentate, and apically subacute with tip slightly turned backwards; posterolateral and posterior margin straight and smooth; calli lacking tubercles behind (Fig. 20). Legs. External face of hind coxa with large, and robust tubercle; hind femur medially incrassate, with three to four rows of tubercles on dorsal surface, and ventrally with two rows of lateral spines; hind tibia conspicuously dilated. Scutellum wider than long or as long as wide. Abdomen slightly dilated, wider than hemelytra, with maximum width less than maximum width of pronotum, across humeral angles.

Genitalia. Posteroventral edge of genital capsule simple, straight. Paramere as in Fig. 11.

Coloration of females similar to holotype. Antennal segment IV orange; scutellum light yellow, and basally light orange; connexival segments VIII and IX black, with anterior third yellow; dorsal abdominal segment VIII black, and IX dark orange with posterior margin black; anterior lobe of metathoracic peritreme yellow with outer bor-

der narrowly black, and posterior lobe black; fore and middle femora orange; hind femur orange and dorsally black; genital segments bright orange with outer border of gonocoxae I, paratergite VIII and IX yellow; abdominal sterna III to VII medially light yellow, laterally light orange; pleural sterna light orange, with anterior third yellow. External face of hind coxa blunt, without tubercle; hind tibia almost cylindrical. Plica located close to posterior border of abdominal sternite VI.

Body surface rather dull, seldom shiny ventrally, and clothed with short, decumbent, silvery pubescence, intermixed with few erect bristle-like setae; antennal segment I clothed with erect setae; antennal segments II to IV and legs with appressed and few suberect setae; posterior third of pronotal disc, scutellum, clavus, corium, and acetabulae punctate; head, anterior third of pronotal disc, calli, connexival segments, thorax, and abdominal sterna impunctate; posterior third of pronotal disc with few irregular or rather inconspicuous small tubercles or callosities; propleura, mesopleura, and metapleura with few tubercles, almost inconspicuous; genital segments of both sexes minutely tuberculate and punctate.

**Measurements.** ♂ holotype first, then ♀: Head length 1.95, 1.90, width across eyes 2.45, 2.40, interocular space 1.50, 1.47, interocellar space 0.83, 0.85, preocular distance 1.40, 1.40; length of antennal segments: I, 4.00, 3.70, II, 3.75, 3.50, III, 3.05, 2.85, IV, 3.25, 3.25. Pronotum: Total length 5.95, 5.93, width across frontal angles 2.60, 2.60, width across humeral angles 10.55, 11.15. Scutellar length 3.35, 3.65, width 3.35, 3.75. Maximum width of abdomen 9.80, 11.05. Total body length 28.00, 26.53.

**Holotype.** ♂ Mexico: Guerrero, Cañon del Zopilote, 23-VIII-1984 (H. Brailovsky). (UNAM). **Paratypes.** 3 ♀ Mexico: Guerrero, Valerio Trujano, 9-VII-1961, 8-VIII-1962, 21-VI-1964 (L. Vazquez). (UNAM). 1 ♂ Mexico: Guerrero, 8 km, W Iguala, 25-VIII-1983 (H. Brailovsky). (UNAM). 2 ♀ Mexico: Guerrero, Mezcala, 12-VII-1967 (H. Perez), 26-VI-1971 (E. Martin). (UNAM). 1 ♂ Mexico: Guerrero, 6mi W Iguala, 19-VIII-1981 (J. Chemsak and A. & M. Michelbacher). (UCB). 1 ♀ Mexico: Guerrero, 3km S Xalitla, 610m, 18-00 N-98 -24 W, 1-VII-1992 (C. L. Bellamy). (CMN). 1 ♀ Mexico: Puebla, km 78 carr. Cuautla-Oaxaca, 30-X-1987 (F. Arias, R. Barba, and E. Barrera). (UNAM).

**Etymology.** From the Latin *preclarus* meaning very beautiful, splendid, in reference to the general shape.

**Discussion.** *Mozena preclara* (Fig. 25) appears to be closely related to *M. nestor* (Stål) (Fig. 27) on the basis of the body surface clothed with short and decumbent silvery pubescence, intermixed with few erect bristle-like setae, antennal segment I clothed with erect setae (Fig. 1), with dorsal surface black, and ventrally orange, hu-

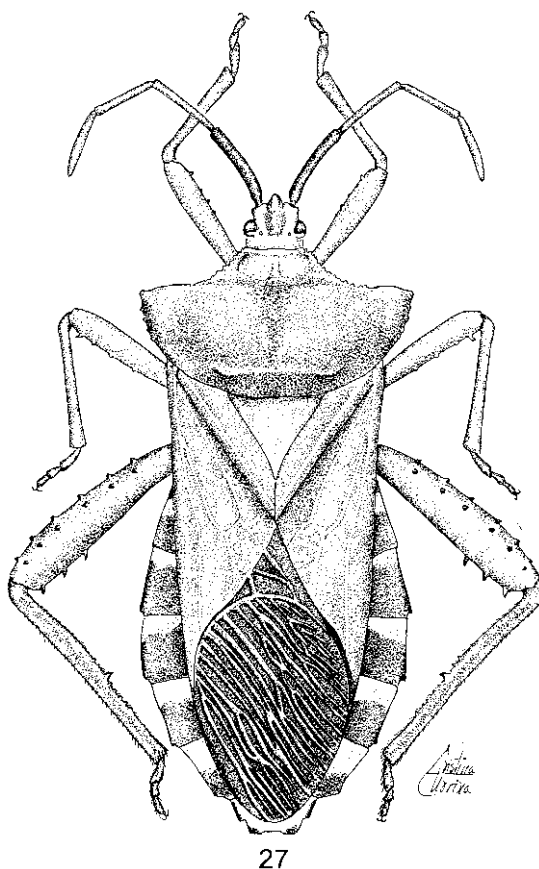


Fig. 27. *Mozena nestor* (Stål), dorsal view (female).

meral angles outwardly directed, slightly upwards, and not lunate, and calli lacking tubercles behind him. *M. nestor* is recognized by the widely dilated abdomen, with maximum width wider than maximum width of pronotum across humeral angles, male hind femur heavily incrassate, and humeral angles robust and medially produced (Fig. 16). In *M. preclara* the maximum width of abdomen is less than maximum width of pronotum across humeral angles, male hind femur medially incrassate, and humeral angles relatively quadrate, not widely expanded, and conspicuously produced (Fig. 20).

*Mozena presigna* Brailovsky and Barrera, **New Species**  
Figs. 4, 7, 12, 21

**Description.** Male (holotype). Dorsal coloration: head dark orange hazel, with tylus reddish brown, and space between ocelli dark orange; antennal segment I with dorsal face reddish brown, ventrally dark orange; segments II and III reddish brown, with basal and apical joint dirty yellow to dark orange; antennal segment IV bright orange; pronotum, scutellum, clavus and corium

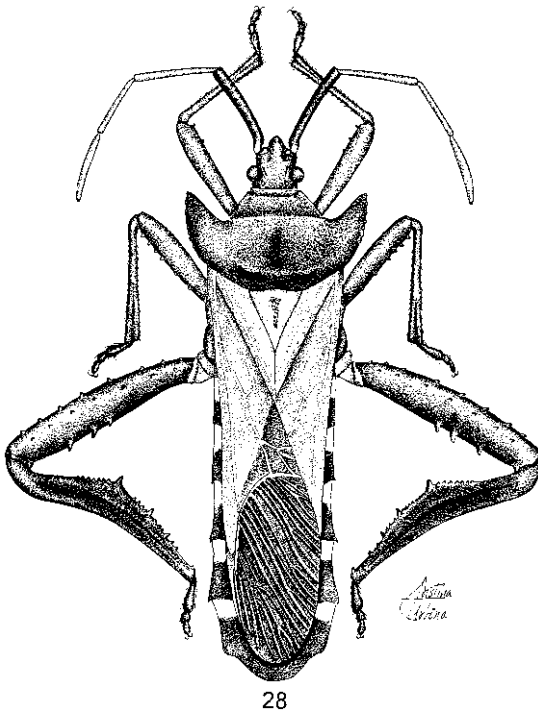


Fig. 28. *Mozena perezae* Brailovsky and Barrera, **New Species**, dorsal view (male).

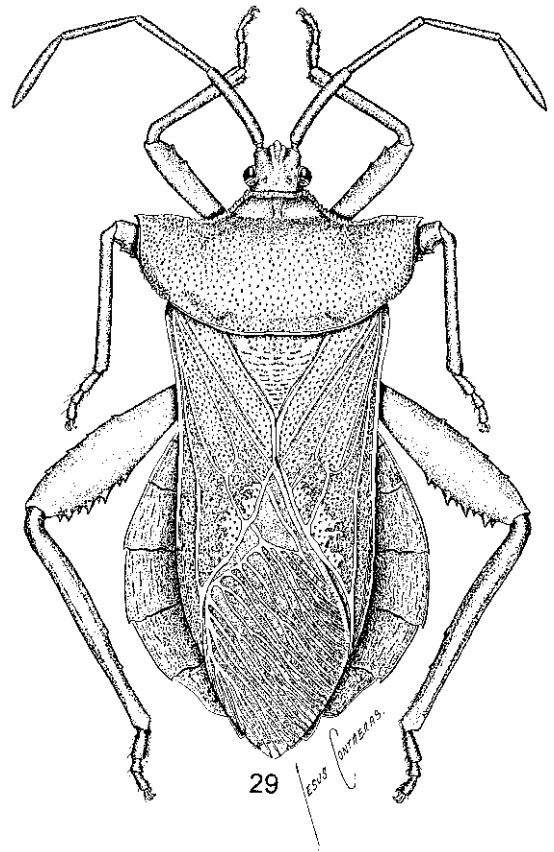


Fig. 29. *Mozena rufula* Van Duzee (female).

bright dark orange hazel with humeral angles black, and apex of scutellum yellow; hemelytral membrane amber-like with basal angle and veins brown; connexival segment III yellow with upper margin of posterior half reddish brown; connexival segments IV to VII reddish brown with anterior third yellow; dorsal abdominal segments bright orange, with black longitudinal stripe running from IV to VII segment. Ventral coloration. Included buccula, rostral segments (apex of IV black), and coxae (outer margin reddish brown) dark to light chestnut orange to chestnut yellow; outer margin of humeral angles, posterior third of pleural abdominal segments III to VII and spines black; anterior lobe of metathoracic peritreme black with central area dark orange, and posterior lobe black; trochanters dark chestnut orange, apical third reddish brown; fore and middle femora light chestnut yellow, distal joint reddish brown; hind femur chestnut orange yellow with dorsal tubercles, ventral spines and wide dorsal longitudinal stripe reddish brown; fore and middle tibiae reddish brown with ventral face lighter; hind tibia reddish brown; tarsi reddish brown to dark chestnut orange; propleura, mesopleura, and thoracic metapleura with creamy yellow vittae poorly developed, and discontinuous, those on metapleura sometimes lacking; creamy yellow to light orange vittae along each side of abdominal

sterna III to VI characteristically enlarged posteriorly (Fig. 4); rim of abdominal spiracle creamy yellow to whitish; genital capsule reddish brown with lateral margins and posteroventral edge dark chestnut orange. Anterolateral margins of pronotum obliquely straight, and sparsely tuberculate; humeral angles broad, exposed, raised, medium to large sized, hemispheric, directed upward, with tips slightly approaching inwards, and apically acute; posterolateral margins straight, with upper half sparsely dentate, and inner half smooth; posterior margin entire, straight; calli lacking tubercles behind (Fig. 21). Legs. External face of hind coxa with large, robust tubercle; hind femur with three or four rows of tubercles on dorsal surface, ventrally with two rows of lateral spines; hind tibia medially dilated on male, cylindrical and slender on female. Scutellum wider than long. Abdomen slightly dilated, wider than hemelytra, with maximum width less than maximum width of pronotum across humeral angles.

Genitalia. Posteroventral edge of male genital capsule simple, transversely sinuate (Fig. 7). Paramere as in Fig. 12.

Coloration of females similar to male. Connexival segments VIII and IX, and dorsal abdominal segments VIII and IX black; genital plates dark chestnut orange with external face of gonocoxae I, upper margin of paratergite IX, and great portion of paratergite VIII reddish brown. Plica close to posterior border of abdominal sternite VI.

Body surface rather dull, seldom shiny. Head, pronotum, scutellum, clavus, corium, thorax, and abdomen clothed with short suberect bristle-like setae; bristle-like setae of antennal segments I to III, and legs, short, appressed, intermixed with few suberect setae; posterior third of pronotal disc, clavus, and corium strongly punctate; head, anterior third of pronotal disc, calli, thorax, and abdominal sterna almost impunctate; scutellum sparsely punctate; posterior third of pronotal disc, clavus, and corium irregularly scattered with small tubercles or callosities; propleura and mesopleura without tubercles; metapleura with few and low cuticular structures like tubercles or callosities; genital segments of both sexes minutely tuberculate, and punctate.

Variation. 1: Antennal segments I to III chestnut orange. 2: Dorsal surface of head with reddish brown stripe running from tylus to vertex. 3: Scutellum black with apex dark chestnut orange. 4: Upper margin of connexival segments III to IX entirely black. 5: Dorsal abdominal segments II to IX black, or segments II to IV bright orange, and V to VII black with anterior margin bright orange. 6: Anterior lobe of metathoracic peritreme entirely black.

**Measurements.** ♂ holotype first, then ♀: Head length 1.80, 1.75, width across eyes 2.35, 2.30, interocular space 1.55, 1.47, interocellar space 0.72, 0.72, preocular distance 1.37, 1.27; length of antennal segments: I, 4.14, 3.50, II, 4.10, 3.40, III, 3.17, 2.50, IV, 3.25, 2.80. Pronotum: Total length 5.20, 5.20; width across frontal angles 2.70, 2.60; width across humeral angles 8.13, 8.28. Scutellar length 2.80, 2.77, width 2.87, 2.85. Maximum width of abdomen 6.08, 6.91. Total body length 21.17, 20.46.

Holotype. ♂ Mexico: Oaxaca, Totolapan, 16 39'52"N-96 16'75"W, 30-VI-1996 (H. Brailovsky and E. Barrera). (UNAM). Paratypes. 1 ♂, 1 ♀ Mexico: Oaxaca, Tehuantepec, 7-VII-1983 (H. Brailovsky). (UNAM). 1 ♂ Mexico: Oaxaca, 23 km Morro de Mazatlan-Salina Cruz, 24-III-1990 (E. Barrera and H. Brailovsky). (UNAM). 17 ♂, 20 ♀ Mexico: Oaxaca, 23 km S Matias Romero, 23-VII-1974 (Clark, Murray, Ashe, Schaffner). (TAMU, UNAM). 1 ♀ Mexico: Oaxaca, 11 km NW El Carmen, 17-VII-1981 (J. C. Schaffner). (TAMU).

**Etymology.** From the latin word *presignis*, distinguished or illustrious.

**Discussion.** *Mozena presigna* is similar to *M. nogueirana*, but may be separated by its uniformly dark coloration, the humeral angles broad, conspicuously raised and well developed (Fig. 21), and the posteroventral edge of the male genital capsule transversely sinuate (Fig. 7) without a weak concavity (Fig. 6) which is present in *M. nogueirana*, in which humeral angles are shorter (Fig. 17).

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SUITABILITY OF NINE MEALYBUG SPECIES  
(HOMOPTERA: PSEUDOCOCCIDAE) AS HOSTS FOR THE PARASITOID  
*ANAGYRUS KAMALI* (HYMENOPTERA: ENCYRTIDAE)

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ABSTRACT

The parasitoid *Anagyrus kamali* Moursi [Hymenoptera: Encyrtidae] has been recently introduced into the Caribbean as a biological control agent against the Hibiscus Mealybug, *Maconellicoccus hirsutus* Green [Homoptera: Pseudococcidae]. In order to understand host/parasitoid ecological interactions and optimize the mass-production system of this parasitoid, eight mealybug species (*Planococcus citri* (Risso), *Planococcus halli* Ezzat & McConnel, *Dysmicoccus brevipes* (Cockerell), *Pseudococcus elisae* Borchsenius, *Saccharococcus sacchari* (Cockerell), *Puto barberii* (Cockerell), *Nipaecoccus nipae* (Newstead), *Plotococcus neotropicus* (Williams & Granara de Willink)) common to Trinidad were tested to determine their potential as alternative hosts for the parasitoid. Susceptibility, preference and suitability tests were conducted. In addition to *M. hirsutus* ( $4.5 \pm 2.04$  hosts parasitized per female parasitoid in 30 min), *Planococcus citri* ( $1.1 \pm 1.23$  hosts parasitized) and *Planococcus halli* ( $0.8 \pm 1.41$  hosts parasitized) were the only species parasitized. However, the parasitoid did not complete its development in the latter two hosts. Out of nine mealybug species, *M. hirsutus* was the only suitable host for the complete development of *A. kamali* progeny. This level of host specificity by *A. kamali* may prevent adverse effect to other Caribbean mealybug species.

Key Words: *Maconellicoccus hirsutus*, hibiscus mealybug, parasitoid, Pseudococcidae, host species selection

RESUMEN

El parasitoido *Anagyrus kamali* Moursi [Hymenoptera: Encyrtidae] fue introducido en el Caribe para el control biológico de la cochinilla rosada, *Maconellicoccus hirsutus* Green [Homoptera: Pseudococcidae]. Para mejorar nuestro conocimiento del sistema plaga/parasitoido y optimizar la producción del parasitoido, ocho especies de cochinillas (*Planococcus citri* (Risso), *Planococcus halli* Ezzat & McConnel, *Dysmicoccus brevipes* (Cockerell), *Pseudococcus elisae* Borchsenius, *Saccharococcus sacchari* (Cockerell), *Puto barberii* (Cockerell), *Nipaecoccus nipae* (Newstead), *Plotococcus neotropicus* (Williams & Granara de Willink)) de Trinidad fueron probadas para determinar sus potencial como hospedantes alternativos del parasitoido. *M. hirsutus* ( $4.5 \pm 2.04$  hospedantes parasitados por parasitoido en 30 min), *Planococcus citri* ( $1.1 \pm 1.23$  hospedantes parasitados) y *Planococcus halli* ( $0.8 \pm 1.41$  hospedantes parasitados) fueron las únicas especies parasitadas, sin embargo, el parasitoido nunca completo su desarrollo en las dos últimas especies. *M. hirsutus* fue la única especie en la cual el parasitoido pudiera completar su desarrollo. Por eso, la introducción de *A. kamali* en el Caribe no debería afectar la biodiversidad local.

Since its accidental introduction into the island of Grenada in 1994, *Maconellicoccus hirsutus* Green (Homoptera: Pseudococcidae), commonly named the hibiscus or pink mealybug, has been

inexorably spreading through the Caribbean and is now present on 18 Islands (International Institute of Entomology 1997). *M. hirsutus* has high reproductive potential (384-540 eggs laid per fe-



male (Mani 1989)) and injects its saliva at the point of feeding, causing severe distortion of leaves, new shoots and fruit (Williams 1996). Because of its wide host range (ca. 125 host species (Malvaceae, etc.), Mani 1989) and its rapid geographic expansion on agricultural land, home gardens and forest areas, biological control appears to be the best strategy to reduce *M. hirsutus* populations.

The solitary endoparasitoid *Anagyrus kamali* Moursi (Hymenoptera: Encyrtidae) was imported from China by CABI Bioscience, for biological control of *M. hirsutus* in the Caribbean (Sagarra & Peterkin 2000). The preferred host of *A. kamali* is *M. hirsutus* (Moursi 1948), but it was also reported utilizing five other mealybug species in Jordan and India (Noyes & Hayat 1994), including *Nipaeococcus viridis* (Newstead) (Meyerdirk et al. 1988), and *Ferisia virgata* (Cockerell) (Cross & Noyes 1996). These mealybug species were found to be parasitized by other *Anagyrus* species that were subsequently considered as synonymous to *A. kamali*. However their suitability for the development of *A. kamali* has not been formally determined.

It is important to ensure that introduction of a newly introduced natural enemy will minimally disrupt local mealybug biodiversity. On the other hand, the impact of an insect parasitoid on target pest populations may be enhanced by the availability of alternative hosts in or around the crop especially in periods of host shortage (Powell 1986). It has been suggested that alternative hosts can help to improve synchrony between parasitoids and their pest hosts, improve parasitoid distribution and reduce intraspecific competition in the parasitoid population (van den Bosch & Telford 1964).

An understanding of host/parasitoid interactions is essential to determine if other co-existing mealybug species could be used by *A. kamali* as alternative hosts. Our objective was to determine the susceptibility, preference, and suitability of eight common mealybug species in Trinidad (i.e., *Planococcus citri* (Risso), *Planococcus halli* Ezzat & McConnel, *Dysmicoccus brevipes* (Cockerell), *Pseudococcus elisae* Borchsenius, *Saccharococcus sacchari* (Cockerell), *Puto barberi* (Cockerell), *Nipaeococcus nipae* (Newstead), *Plotococcus neotropicus* (Williams & Granara de Willink) and *M. hirsutus* to parasitization by *A. kamali*. The latter mealybug species are pantropical and have been present in Trinidad for more than a hundred year (Records from CABI insect collection).

## MATERIALS AND METHODS

### Rearing of *M. hirsutus*

Mealybugs were reared on sprouted potatoes in nylon mesh cages (32 potatoes per cage) supported on steel wire frames (48 × 48 × 68cm). The

cultures were maintained in the dark at 27 ± 2°C. Each week, 64 potatoes were individually infested with 20 adult female mealybugs having well-formed ovisacs. Weekly infestations ensured a continuous supply of different *M. hirsutus* nymphal instars. Three weeks after infestation, sprouted potatoes had *M. hirsutus* populations that consisted predominantly of second and third instar mealybugs. Adult females with ovisacs were available after 4-6 weeks. Size was used to distinguish *M. hirsutus* stages (Ghose 1971).

### Rearing of Other Mealybug Species

*Planococcus citri*, *P. halli*, *Dysmicoccus brevipes*, and *Pseudococcus elisae* were reared on sprouted potatoes in plastic boxes with a mesh cover (6 potatoes per box). The potato sprouts were individually infested with 20 adult female mealybugs of each species. *Saccharococcus sacchari* were reared on young sugar cane seedlings individually infested with 5 adults. *Puto barberi* were reared on young Bougainvillea seedlings individually infested with 30 adult mealybugs. *Nipaeococcus nipae* were reared on young coconut trees, and leaves were individually infested with 30 adults. *Plotococcus neotropicus* were reared on young Guava seedlings individually infested with 30 adult mealybugs. Four weeks after initial infestation, third instars and young adults of each species were collected and used for the experiments. Mealybug identification was confirmed by J. Etienne from INRA, Guadeloupe, and D. Matile-Ferrero, Musée d'Histoire Naturelle in Paris, France.

### Rearing of *A. kamali*

Each week, 100 *A. kamali* adult females were released into two cages each containing 32 infested sprouted potatoes supporting three-week old populations of *M. hirsutus* (after Sagarra and Vincent, 1999). Colonies were maintained at 27 ± 2°C, 60 ± 10% RH, under a photoperiod of LD 12:12. Light was provided by two fluorescent lamps (70W) suspended 30cm above the cages. Emerged parasitoids were collected after 20-25 days. Two-day old, mated females were used for experiments. Female parasitoids were considered as experienced because they had been exposed to different stages of mealybugs in the rearing cages for a maximum of 12 hours after emergence.

In all experiments with the nine mealybug species we used third instars and early adult females (preovisac). All experiments were conducted in the laboratory at 27 ± 2°C, 60 ± 10% RH.

### Host Species Susceptibility (No-Choice Experiments)

Ten mealybugs from each species were collected and placed in groups in separate Petri

dishes. One female parasitoid was introduced into each dish. The parasitoid and the mealybugs were observed continuously for 30 min to record number of encounters, and host probing with the ovipositor. Twenty groups of each species were exposed to different individual *A. kamali* females in this manner. The parasitoids were removed after 30 min. All exposed mealybugs were dissected in a drop of ethanol (70%), and the number of parasitoid eggs in each individual host was recorded.

The number of encounters, ovipositor probing, hosts parasitized by each *A. kamali* female, the total number of eggs laid per parasitoid female, and the number of parasitoid eggs per accepted host were used as the criteria for determining host species susceptibility. Paired Chi-square frequency analysis was used to determine if the number of encounters, ovipositor probing, hosts parasitized by each *A. kamali* female, total number of eggs laid per parasitoid female, and number of parasitoid eggs per accepted host was dependent on the mealybug species at  $p = 0.05$  (Systat 7.0 for Windows 95).

#### Host Species Preference (Two-Choice Experiments)

In two-choice experiments, five *M. hirsutus* were placed in a Petri dish with five specimens of one of the eight other mealybug species, and a single female parasitoid was introduced into each experimental arena. The parasitoid and mealybugs were observed continuously for 30 min to record the number of encounters, and probing with the ovipositor for each mealybug species. The parasitoid was then removed. We dissected each host in a drop of ethanol (70%), and the number of eggs in each host was recorded for each species. Twenty batches of each of the eight host combinations were exposed to individual *A. kamali* females in this manner.

The number of encounters, probing, hosts parasitized per parasitoid, the total number of parasitoid eggs laid per replicate, and the number of parasitoid eggs per accepted host were used as the criteria for determining preference between *M. hirsutus* and the other host species. Paired Chi-square frequency analysis was done to determine if the number of encounters, ovipositor probing, hosts parasitized by each *A. kamali* female, total number of eggs laid per parasitoid female, and number of parasitoid eggs per accepted host was dependent on the mealybug species at  $p = 0.05$  (Systat 7.0 for Windows 95).

#### Host Species Suitability

Thirty individuals of each mealybug species were transferred onto sprouted potatoes and placed in a cage made of a transparent plastic cylinder (20 × 12 cm diameter) with the top covered with nylon mesh. Three adult female parasitoids

were introduced into the cage for a period of 24h. The mealybugs were observed on a daily basis to record parasitoid emergence. Parasitoid progeny was collected, sexed and the size (i.e., fitness) of emerged parasitoids was estimated by measuring the length of the left hind tibia (Ghose 1971). The total body length (base of head to tip of abdomen) and the length of the hind tibia were measured for 35 adult *A. kamali* and fitted to a linear regression model (Systat 7.0 for Windows 95). The coefficient of determination ( $r^2$ ) indicated that 84.2% of the variation in the body length of the wasps can be explained by the length of the hind tibia of the parasitoid in a linear relationship, therefore providing an accurate and rapid means of assessing of the overall size of the parasitoid. The host suitability experiment was replicated five times for each mealybug species. The criteria used to determine host suitability were the number of emerged parasitoids per replicate, the secondary sex ratio (number of males divided by total progeny number, as suggested by Godfray 1994; Van Alphen & Jervis 1996), the development time and the size of the parasitoids.

## RESULTS

#### Host Species Susceptibility (No-Choice Experiment)

In no-choice experiments, *A. kamali*'s encounter rate, ovipositor probing, number of hosts parasitized, and number of eggs oviposited was significantly higher on *M. hirsutus*, compared to the other mealybug species (Table 1). Among the eight other mealybug species, the parasitoid's encounter rate for *P. citri*, *P. halli*, and *P. elisae* were significantly ( $P < 0.05$ ) higher than for *D. brevipes*, *N. nipe*, *P. barberi*, *P. neotropicus*, and *S. sachari*. The latter species did not induce searching responses by *A. kamali* females, which remained on the Petri dish cover during most of the 30 min. One species, *P. barberi*, defended itself against parasitoid encounter by violent torsions of its abdomen. The number of ovipositor probings of mealybugs was not significantly different ( $P < 0.05$ ) among the eight tested hosts. Probing of *M. hirsutus* was significantly ( $P < 0.05$ ) greater than for the other mealybug species. *D. brevipes*, *N. nipe*, *P. barberi*, *P. neotropicus*, and *S. sachari* were not probed, whereas few individuals of *P. citri*, *P. halli*, and *P. elisae* were probed.

*Planococcus citri*, and *P. halli* were the only species, other than *M. hirsutus*, that were parasitized by *A. kamali*, but at a significantly ( $P < 0.05$ ) lower level than *M. hirsutus* (Table 1).

In all three attacked species, superparasitism occurred, the number of eggs oviposited being greater than the number of hosts parasitized. Signs of egg encapsulation were observed in these three species.

TABLE 1. ENCOUNTER, PROBING AND OVIPOSITION OF *A. KAMALI* ON NINE SPECIES OF PSEUDOCOCCIDAE (NO-CHOICE TEST). FOR EACH SPECIES, TEN INDIVIDUALS WERE EXPOSED TO ONE FEMALE PARASITOID FOR 30 MIN (N = 20).

| HMB species                     | Average ( $\pm$ SD) |                     |                   |                   |
|---------------------------------|---------------------|---------------------|-------------------|-------------------|
|                                 | Hosts encountered   | Ovipositor probing  | Hosts parasitized | Eggs oviposited   |
| <i>Maconellicoccus hirsutus</i> | 23.9 $\pm$ 12.45 a* | 13.4 $\pm$ 11.05 a* | 4.5 $\pm$ 2.04 a* | 6.4 $\pm$ 3.65 a* |
| <i>Planococcus citri</i>        | 10.7 $\pm$ 6.20 b   | 3.3 $\pm$ 3.55 b    | 1.1 $\pm$ 1.23 b  | 1.2 $\pm$ 1.47 b  |
| <i>Planococcus halli</i>        | 8.1 $\pm$ 5.99 b    | 1.3 $\pm$ 2.49 b    | 0.8 $\pm$ 1.41 bc | 1.0 $\pm$ 1.91 b  |
| <i>Dysmicoccus brevipes</i>     | 0.5 $\pm$ 0.83 c    | 0.0 b               | 0.0 c             | 0.0 b             |
| <i>Pseudococcus elisae</i>      | 7.7 $\pm$ 5.93 b    | 0.4 $\pm$ 1.05 b    | 0.0 c             | 0.0 b             |
| <i>Saccharococcus sacchari</i>  | 0.2 $\pm$ 0.49 c    | 0.0 b               | 0.0 c             | 0.0 b             |
| <i>Puto barberi</i>             | 2.0 $\pm$ 1.91 c    | 0.0 b               | 0.0 c             | 0.0 b             |
| <i>Nipaeococcus nipe</i>        | 0.2 $\pm$ 0.41 c    | 0.0 b               | 0.0 c             | 0.0 b             |
| <i>Plotococcus neotropicus</i>  | 1.8 $\pm$ 1.07 c    | 0.0 b               | 0.0 c             | 0.0 b             |

\*Within columns, pairs of means followed by the same letters are not significantly different (Chi-square test,  $P < 0.05$ ).

#### Host Species Preference (Two-Choice Experiments)

In choice experiments, *M. hirsutus* was significantly preferred to all other mealybug species in terms of encounter rate, ovipositor probing, number of hosts parasitized, and number of eggs oviposited. *Dysmicoccus brevipes*, *N. nipe*, *P. barberi*, *P. elisae*, *P. neotropicus*, and *S. sacchari* were not utilized by female parasitoids, and were not parasitized when exposed to *A. kamali* simultaneously with *M. hirsutus*. Low numbers of *P. citri*, and *P. halli* were parasitized ( $0.3 \pm 0.39$ , and  $0.1 \pm 0.21$  hosts parasitized respectively) when compared to *M. hirsutus* ( $3.9 \pm 0.95$  hosts parasitized) in the 30 min assay. This level of parasitism in *P. citri*, and *P. halli* was not significantly different ( $P = 0.05$ ) from the other six mealybug species tested.

#### Host Species Suitability

Of the nine mealybug species tested, successful development of parasitoid progeny occurred only in parasitized *M. hirsutus*, with an average progeny emergence of  $14 \pm 1.6$  individuals out of the 30 hosts exposed to the three female parasitoids. The average progeny sex-ratio was  $0.49 \pm 0.107$ .

#### DISCUSSION

Of the eight tested mealybug species, *D. brevipes*, *N. nipe*, and *S. sacchari* did not induce searching behavior by *A. kamali*. *Puto barberi*, and *P. neotropicus* induced searching behavior by *A. kamali*, but host rejection occurred after antennation, prior to ovipositor probing. *Pseudococcus elisae* was rejected after being probed with the ovipositor. *Planococcus citri* and *P. halli* were recognized by *A. kamali* as potential hosts and parasitized, but these two host species were not suitable for parasitoid progeny development. *M. hirsutus* was the most suitable host, allowing the complete

development of *A. kamali*. The parasitoid discriminated among different host species and selected the most suitable host for the development and survival of its progeny.

*A. kamali* has been recorded as a parasitoid of *M. hirsutus* (Moursi 1948), but also on several Pseudococcidae host species like *Pseudococcus* sp. on *Citrus limonium* (Agarwal 1965) and cocoa (Shafee et al. 1975), *Ferrisia virgata* (Cockerell) and *Ferrisia* spp. on an avenue tree (Subba Rao & Rai 1970), and *Nipaeococcus viridis* (Newstead) on Citrus (Meyerdirk et al. 1988). *A. kamali* has also been recorded from *Nipaeococcus* spp. on *Acacia* sp. (Shafee et al. 1975). Except for *Ferrisia virgata*, none of these mealybug species are present in Trinidad. In addition no emergence of the parasitoid from this host was obtained in laboratory experiments (M. Hoy 1999, pers. comm.). The members of the genera *Pseudococcus* and *Nipaeococcus* reported from Trinidad (i.e., *Nipaeococcus nipe* and *Pseudococcus elisae*) were not susceptible to parasitization by *A. kamali*.

The absence of alternative hosts for *A. kamali* development in Trinidad might reduce the efficiency of the parasitoid as a biological control agent since other alternative host species will not be available for *A. kamali* when *M. hirsutus* is at low densities. However, this is an advantage from a point of view of preservation of native mealybug biodiversity, as the introduction of this new natural enemy should not disturb indigenous species because *A. kamali* is relatively specific to the HMB pest, and therefore not competing with indigenous species of natural enemies.

From a mass production perspective, it was observed that contamination of *M. hirsutus* cultures with *Planococcus* sp. led to a decrease of the parasitoid progeny emergence (L. Sagarra, unpublished data). This can be explained by the fact that parasitoid eggs allocated to *Planococcus* will not develop into adults. Parasitization of *P. citri*

and *P. halli* by *A. kamali* will require keeping the *M. hirsutus* cultures for *A. kamali* mass-production free from these two other mealybug species.

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## CAYMANIS, A NEW GENUS OF ANTILLOCORINI FROM THE CAYMAN ISLANDS (HEMIPTERA: LYGAEIDAE)

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### ABSTRACT

*Caymanis bracensis*, a new genus and species of Antillocorini from the Cayman Islands, is described and illustrated. This species differs from other antillocorines by the reticulated texture of the surface of the third and fourth abdominal sternites in both sexes and by the presence of a large curved basal spine found ventrally on the fore femur in the males.

Key Words: Hemiptera, Lygaeidae, Antillocorini, new genus, new species, *Caymanis bracensis*, Cayman Brac, West Indies

### RESUMEN

Se describe e ilustra el género *Caymanis* y la especie nueva *C. bracensis* de Antillocorini de la isla de Cayman Brac. Esta especie se distingue de otros antillocorinos por la textura reticulada de la superficie ventral de los segmentos abdominales tercero y cuarto en ambos sexos y por una espina curvada presente ventralmente en el primer par de fémures en los machos.

Baranowski and Slater (1998) reported specimens collected in the Cayman Islands as apparently representing an undescribed species of the *Botocudo* complex. These specimens are not congeneric with the type species of *Botocudo*. They differ from *Botocudo* and most other antillocorines by the presence of all trichobothria of the fifth abdominal sternite located anterior to the spiracle of the same segment, fore femora incrassate and armed ventrally with spines in both sexes, and the males with a large distally curved spine at the base of the fore femur.

"We are using "Lygaeidae" in the traditional sense rather than "Rhyparochromidae" as used in the Henry (1997) classification. This is done to conform to the conservative course we will follow in a large manuscript on the Lygaeidae (sensu lato) of the West Indies, but is not to be interpreted as basic disagreement with the Henry classification."

All measurements are in millimeters.

### *Caymanis* Baranowski and Brambila, **New Genus**

Type species: *Caymanis bracensis* Baranowski and Brambila. Monobasic.

This genus is characterized by the incrassate fore femur with one row of spines on the anterior margin of its ventral surface; males with a basal, ventral, large, distally curved spine, (Fig. 2) on fore femur; pronotum with lateral margins slightly sinuate and carinate, but not explanate; metapleuron with scent gland auricle sessile and curved posteriorly (Fig. 4); trichobothria on abdominal sternite 5 slightly dorsoventral to each

other and anterior to spiracle (Fig. 3). Abdominal sternites 3 and 4 reticulate in both male and female.

Etymology. Named for the island, Cayman Brac, on which it was found.

*Caymanis* has the following typical antillocorine characters: 1) inner half of apical corial margin concave, 2) three sets of gland scars dorsally on the abdomen between terga 3-4, 4-5, and 5-6, 3) innerlatero tergites present, and 4) all abdominal spiracles ventral. The spiracles on abdominal segments 3, 4 and 5 are below the sternal shelf (Fig. 3).

The femoral spine of the male is considered apomorphic for *Abroxis* since no other Antillocorini has this character; *Paradema* has a large spine ventrally on the fore femur, but it is not curved and it is placed distally rather than proximally.

The position of the trichobothria on the fifth abdominal sternite, both located anterior to the spiracle, is also apomorphic since the plesiomorphic condition is the position of these trichobothria below (or behind) the spiracle (Slater 1980). Species of *Caymanis* also have the trichobothria anterior to the spiracle, but in *Abroxis* they are slightly dorsoventrad to each other rather than linear, the latter a highly derived condition according to Slater (1980); furthermore, *Cligenes* differs from *Caymanis* by the presence of a prosternal groove.

No other antillocorine has been found with abdominal sternites 3 and 4 reticulate in both male and female; this character might prove to be an apomorphic generic character rather than a specific character. Sexual dimorphism occurs in the

genus *Caymanis*, males having on the fore femora a large spine near the base, on the ventral surface, not present in females.

*Caymanis bracensis* Baranowski and Brambila,

**New Species**

(Figs. 1-8)

Chiefly dark reddish brown. Scutellum reddish brown with brownish black at base and sides, with apex yellowish white. Clavus and corium brownish white (bronze-like in appearance); corium with two dark brown markings, one laterally at center and one at apex. Membrane opaque brownish white. Abdominal sternites 5-7 dark brown. Antennae, labium and legs yellowish brown, antennal segment IV darker brown. Tylus reaching middle of antennal segment I. Gula narrow, deep, and with apex of bucculae meeting in a round arc, reaching anterior margin of prosternum (Fig. 1). Surface of thorax, clavus and corium not polished but dull; pronotum, scutellum, clavus, and corium punctate; pronotum and scutellum with short decumbent setae. Corium with anterior half of lateral margin slightly sinuate. Male genital capsule Fig. 5 posterior view, Fig. 6 lateral view, Fig. 7 dorsal view and Fig. 8, right paramere. Thoracic sterna keeled, keel on prosternum only between coxae. Metapleuron with evaporative area triangular with apex ventral. Antennal segment II somewhat clavate. Fore femur incrassate with a row of small straight spines on anterior margin of ventral surface, an irregular row of small knobs on the posterior margin, and with a large curved spine near base (Fig. 2).

Head length total 0.35, width across eyes 0.43, interocular distance 0.26. Pronotal length 0.46, width at humeral angles 0.82. Scutellar length 0.46, width 0.50. Length claval commissure 0.12. Total wing length from base of corium 1.28. Length antennal segments I 0.22, II 0.24, III 0.20, IV 0.26. Length labial segments I 0.16, II 0.34, III 0.16, IV 0.14. Total body length 1.86.

Holotype: ♂ CAYMAN BRAC, W.I., Major Donald Road, 17-19-X-1995, under *Ficus* sp., H. V. & R. M. Baranowski (RMB).

Paratypes: 7 ♂♂, 2 ♀♀ same data as holotype (RMB); 1 ♂, 1 ♀ same locality as holotype, 7-VII-1997 (R. M. Baranowski). In R. M. Baranowski and J. A. Slater collections.

**Etymology.** This species was named for the island, Cayman Brac, on which it was found.

Females similar to males in size, texture, and coloration, except fore femur without large ventral spine. Some specimens with posterior pronotal lobe darker than anterior lobe, membrane translucent, or membrane opaque white; nearly all specimens with scutellum lacking dark brown areas; one specimen small and yellowish brown. Range of total body length: 1.65 to 1.88 mm. All specimens macropterous.

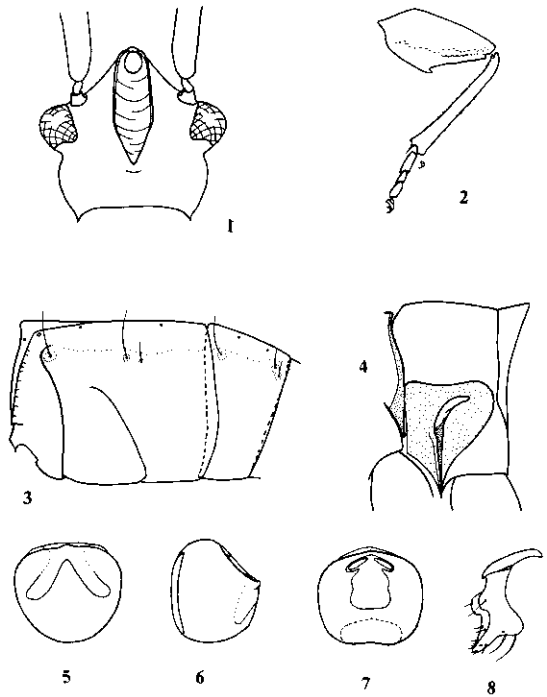


Fig. 1. Head, ventral view.

Fig. 2. Male fore leg, anterior view.

Fig. 3. Male abdominal sternites 3-6, lateral view.

Fig. 4. Metathorax with scent gland auricle and evaporative area.

Figs. 5-7. Male genital capsule, posterior, lateral and dorsal views.

Fig. 8. Right paramere.

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THE LARVAL HABITAT OF *CEDUSA INFLATA*  
(HEMIPTERA: AUCHENORRHYNCHA: DERBIDAE) AND ITS RELATIONSHIP  
WITH ADULT DISTRIBUTION ON PALMS

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ABSTRACT

Adults of Derbidae (Hemiptera: Auchenorrhyncha) are common on foliage of Palmae in many tropical localities; their larvae are believed to develop in decaying debris. The larval stage of *Cedusa inflata* (Ball), a derbid common on palms in Florida and the Caribbean Region, was observed and is figured for the first time, and its habitat, decaying organic debris, was documented. In plantings of coconut palm, a mean of 56.1 larvae of *C. inflata* solitary or in aggregations of up to 13 individuals were found in each of 10 piles of organic debris from the palms. The larvae were in moist places in the interior of debris piles usually near fungal mycelia, their presumed food resource. Sparse numbers ( $x = 6.06$ ) of *C. inflata* adults were observed on foliage of each of 10 palms adjacent to piles of organic debris, but were virtually absent from 10 palms >15 m from debris piles. These observations have implications for decaying debris as the assumed larval habitat of derbid species found as adults on palms in many tropical countries.

Key Words: Homoptera, fungivorous insects, *Cocos nucifera*, planthopper, organic debris, tropical plantation crops

RESUMEN

Adultos de Derbidae (Hemiptera: Auchenorrhyncha) son comunes sobre el follaje de Palmae en muchas localidades tropicales; se suponen que sus larvas se desarrollan sobre detrito orgánico podrido. La larva de *Cedusa inflata* (Ball), un dérbido común sobre las palmeras en Florida y la Región Caribeña, fue observada e ilustrada por primera vez, y su hábitat, detrito orgánico podrido, fue documentado. En plantíos de palma de coco un promedio de 56.1 larvas de *C. inflata*, o solitario o en agregaciones de hasta 13 individuos, fueron encontrados en cada uno de 10 montones de detrito orgánico de las palmas. Las larvas estaban en lugares húmedos en el interior de los montones y usualmente estaban cerca de micelios de hongos, su recurso alimenticioso presumido. Números esparcidos ( $x = 6.06$ ) de los adultos de *C. inflata* fueron observados sobre follaje de cada una de 10 palmeras adyacentes a montones de detrito orgánico, pero fueron virtualmente ausentes de 10 palmeras >15 m de distancia de montones de detritos. Estas observaciones tienen implicaciones para el detrito podrido como el hábitat presumido de las larvas de especies de dérbidos que se encuentran sobre las palmeras en muchos países tropicales.

The adults of Derbidae are found on diverse host plants but are exceptionally well represented on palms (Lepesme 1947; Wilson 1987). Little is known of their bionomics. One species, *Cedusa inflata* (Ball), was described from Hispaniola, and is reported from Puerto Rico, Cuba and Florida (Flynn & Kramer 1983). In a survey of auchenorrhynchous insects on palms grown as ornamental plants in mostly urban areas of southern Florida, *C. inflata* was found on 21 species of palms, second in number of "apparent palm hosts" only to *Myndus crudus* Van Duzee (Cixiidae) which was found on 26 species of palms (Howard

& Mead 1980). However, while in this survey *M. crudus* was found on palms at many sites, *C. inflata* was found at only at 6 of the 112 sites where auchenorrhynchous insects were sampled (F. W. H., unpublished). The presence of this insect on diverse palm species reflected the diversity of the palms themselves at the sites where it was collected, which included a large collection of living palms at Fairchild Tropical Garden in Miami, and two additional sites with unusually large palm collections. A notable feature of the palm collections was that they were under particularly conscientious horticultural maintenance,

which included periodic watering and the use of wood chips and other organic debris as a mulch around the base of each palm.

Both *C. inflata* and *M. crudus* occur as adults on palm foliage but do not complete their life cycle on it. *Myndus crudus* larvae develop in the root zone of grasses (Howard & Villanueva-Barradas 1994). The larvae of species of *Cedusa* were unknown but were assumed to occupy cryptic habitats such as rotting organic debris (Flynn & Kramer 1983). Larvae of the derbid *Omolicna cubana* Myers, the adults of which feed on palms in the Caribbean Region, were reared on an *in vitro* culture of the fungus *Rhizoctonia solani* Kuhn (Eden-Green 1973). Derbidae in general are thought to feed on fungi in such habitats (Carver et al. 1991; Wilson et al. 1994).

In June 1996 we noticed that adults of *C. inflata* were consistently present on a small coconut palm (*Cocos nucifera* L.) adjacent to a pile of decaying wood, but absent from palms elsewhere in the vicinity. After a 1½-hour search in the pile, an auchenorrhynchous larva was found. This was reared to adult and identified as *C. inflata*. This is a report of a study to determine whether organic debris consistently served as a larval habitat for *C. inflata*, and whether adults of this species were more abundant on palms near this type of habitat.

#### MATERIALS AND METHODS

The study was conducted on the Fort Lauderdale Research and Education Center, Florida, in 5 plantings, each of which consisted of about 100-380 coconut palms (total = ca. 1000). In each planting, 2 "treatment" palms (total = 10) were selected at random, young coconut palms 4-5 m tall to the tip of the tallest leaf being preferred so that they could be easily examined from the ground or a ladder. Beginning in June 1996, debris that fell from coconut palms (fronds, inflorescences, etc.) was periodically gathered and piled so that large quantities were consolidated immediately adjacent to the 10 treatment palms. For each treatment palm a similar palm in the same planting and about 15-50 m from the nearest debris pile was selected as a control.

Treatment and control palms were examined about a year later during the period July 29-August 11, 1997. On each of 10 days the numbers of adult *C. inflata* on the foliage were counted. On the first day, observations were made from about 0730 hrs-0830 hrs and from 1300-1400 hrs. Since there was a negligible difference between the numbers of *C. inflata* in the early morning compared to midday, observations were made in the early morning on all other days.

During the period August 11-August 26, 1997, the debris piles were searched for insects. Counts were made of *C. inflata* adults and larvae. Specimens collected as adults and 20 adults reared

from larvae captured in the debris were identified. Differences in mean numbers of adults on foliage of treatment and control palms were tested by Student's t-test (SAS 1985).

#### RESULTS

There was a mean of 6.1 (SD = 4.43) *C. inflata* adults on the foliage per treatment palm compared to a mean of 0.02 (SD = 0.03) per control palm. This difference was statistically significant (df = 109, prob > F = 0.0000). This indicated that sparse numbers of these insects were on foliage of palms adjacent to piles of debris, and were virtually absent from palms greater than 15 m from debris piles.

A mean of 56.1 larvae (SD = 41.0, range 22-151) of *C. inflata* (Fig. 1) were found in the 10 debris piles. Callow adults were found in this same habitat. Specimens collected as adults or reared from larvae were positively identified as *C. inflata*.

The larvae of *C. inflata* are reddish purple in the early instars. Later instars are a dark, dull purple and about 1.8-2.0 mm. long. No *C. inflata* larvae

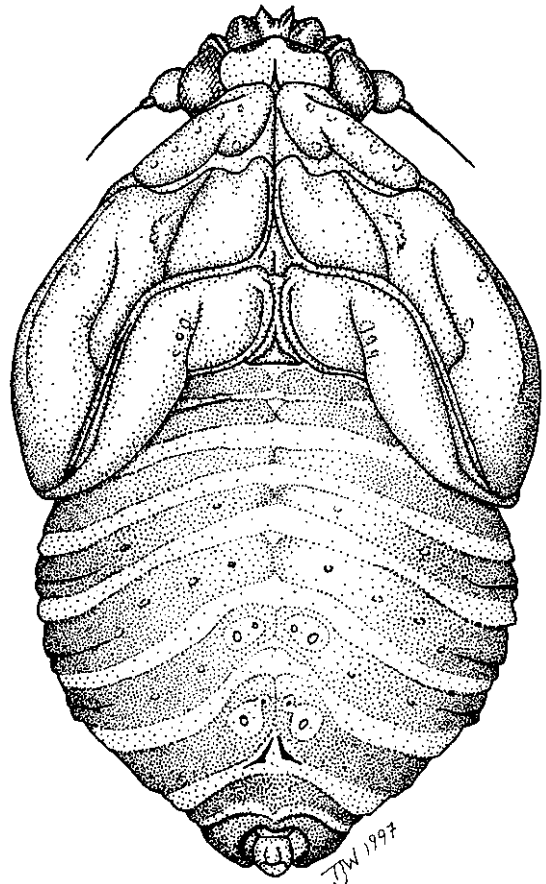


Fig. 1. *Cedusa inflata*, late instar larva.



were observed in the dry outer layers of debris piles. Larvae were solitary or in aggregations of up to 13 individuals on moist debris from about 10 cm below the pile surface to the ground surface. They, along with occasional adults, were most often found near rich growth of fungal mycelia (Fig. 2). We did not attempt to culture and identify the fungi.

Like other Auchenorrhyncha on palms, the adults of *C. inflata* tend to remain motionless on the foliage for long periods. When disturbed, they jump and escape from the foliage more quickly than other Auchenorrhyncha that we have observed in this habitat. However, their presence on palms near debris piles, but virtual absence from palms more than 15 m away from piles, suggests that they do not readily disperse from the vicinity of their larval habitat. Like the adults, the larvae of *C. inflata* become extremely active when their habitat is disturbed, running rapidly and frequently jumping.

#### DISCUSSION

These results are consistent with our hypothesis that the presence of adults of *C. inflata* on palms at certain sites in Florida (Howard & Mead 1980) was related to the presence of organic debris.

Populations of *C. inflata* are typically sparse and patchy both in Florida and in the Caribbean

(Howard et al. 1981; Howard & Mead 1980; F. W. H., unpublished observations). In this study, abundant larval habitats were created by consolidating debris near palms. Even so, only a mean of 6.1 adults of the species were seen on adjacent palms. The numbers of larvae relative to the size of the debris piles indicate that the species tends to occur in low populations even when conditions would appear to be optimal. In contrast, in coconut plantations in Mexico, Central America and the Caribbean, *Omolicna* spp. (Derbidae), which are rare on palms in Florida (Howard & Mead 1980) are among the most consistently found auchenorrhynchous insects on fronds. In Jamaica, debris in petiole axils was searched exhaustively without finding derbid larvae (Wilson 1997). Fungi, ferns and seed plants commonly grow in debris in petiole axils, implying that moisture levels are fairly stable there. However, abundant debris on the ground may be the principal habitat of larvae of *Omolicna* spp. and other derbids in palm plantations in various tropical countries.

#### ACKNOWLEDGMENTS

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Fig. 2. Habitat of larva of *Cedusa inflata*: decaying palm tissue with fungal mycelia. Arrow indicates larva.

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DEVELOPMENT OF *METAMASIVS CALLIZONA*  
(COLEOPTERA: CURCULIONIDAE) ON PINEAPPLE STEMSJORGE SALAS<sup>1</sup> AND J. H. FRANK<sup>2</sup><sup>1</sup>FONAIAP, Centro de Investigaciones Agropecuarias del Estado Lara  
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## ABSTRACT

In the laboratory at 26°C and 14:10 L:D, female *Metamasius callizona* (Chevrolat) oviposited in pocket-shaped slits that they cut in pineapple leaves. Eggs were 1.98 × 0.97 mm and incubation averaged 8.3 d at 82% RH. On a diet of pineapple stem, 5 larval instars averaged 37.4 d to develop to the pupa. The pupal stage averaged 11.8 d, and the pupal weight averaged 0.12 g. Development from oviposition to adult emergence took about 8 wk.

Key Words: Florida, bromeliads, pest weevil, larval instars

## RESUMEN

En el laboratorio a 26° y bajo regímenes de luz de 14 y 10 h respectivamente, hembras de *Metamasius callizona* (Chevrolat) ovipositaron en escisiones con forma de bolsillos que ellas hicieron en hojas de piña. El tamaño de los huevos fue de 1,98 × 0,97 mm y la incubación tomo 8,3 días (promedio) a una HR de 82%. Con una dieta de tallos de piña, los 5 instares larvales tomaron 37,4 días para convertirse en pupa. El estado pupal duro 11,8 días y la pupa peso 0,12g. El desarrollo desde la oviposición hasta el adulto tomó 8 semanas.

*Metamasius callizona* (Chevrolat), a bromeliad-eating weevil native to southern Mexico and some countries of Central America, was first detected in Florida (USA) in 1989. It was reported attacking several native bromeliad species of the genus *Tillandsia*, being most abundant on *T. utriculata* (L.) in southern Florida (Frank & Thomas 1994). It also attacks 12 other genera of bromeliads (including *Ananas*) grown in Florida as ornamental or food plants (Frank & Thomas 1994). Native epiphytic bromeliads are considered highly desirable elements of the flora, and most of them are protected by law (Florida Administrative Code 1998).

A University of Florida project to control *M. callizona* by biological methods (importation of a specialist parasitoid—a tachinid fly of the genus *Admontia*) is in progress. Because of this, a comprehensive knowledge of the life cycle, behavior, and climatic requirements of this weevil is necessary.

## MATERIALS AND METHODS

Research was conducted in a rearing room of the Biological Control Laboratory of the Entomology & Nematology Department, University of Florida, Gainesville, with a photoperiod of 14:10 L:D. A microenvironment for rearing eggs, larvae, and pupae was provided in several 140 mm diam and 25 mm high, large plastic Petri dishes, each having a circular hole cut through its lid to allow introduction of a battery-powered thermo-hy-

grometer probe (RH82, Omega, Stamford, CT). The hole was stoppered when the thermo-hygrometer was not in use. Within the Petri dish, humidity was maintained at 82 ± 6.7% RH (by moistening absorbent paper in the dish), and temperature at 26 ± 0.5°C.

Adult weevils were taken from a greenhouse colony in which numbers are reared for experimental purposes on pineapple (*Ananas comosus* (L.)) crowns rooted in potting soil in plant pots. Pineapple crowns were used as oviposition sites. Two to 3 crowns were planted in soil in pots, or placed vertically in 19 × 14 × 11 cm plastic boxes with a 1-cm sheet of water, and then placed inside a 30 × 30 × 30 cm metal rearing cage. Four to 6 previously sexed couples were placed on the leaves of each crown to obtain eggs. Plants were removed on the second day after adults were introduced. All leaves were separated from the plant and checked under a dissecting microscope for eggs. Eggs were laid individually in slits cut into leaves by adult females. Each egg found was removed and placed in a 55 mm diam × 15 mm high plastic Petri dish with a circular piece of moist paper towel on the bottom. These small Petri dishes were placed within the above-mentioned large ones. Four groups of eggs (n = 14, 14, 13 and 10) were followed through hatching to determine their incubation period. Eggs collected may have been anywhere from 0 to 48 h old; we considered them 24 h old, and adjusted the incubation time accordingly.

After hatching, each larva was fed with sections of pineapple stem sized in relation to the larval size. Each piece of pineapple stem was impregnated with 1 ml of methyl-p-hydrobenzoate (1g/l) to reduce fungal growth. Every day, each larva was observed for development and presence of exuviae after molting. When a larval molt was detected, the exuviae with head capsule were transferred to a labelled vial containing 70% isopropanol, permitting subsequent measurement of head capsule width. Larval food was replaced after each molt. All larvae under observation were followed through each molt to determine the duration of each instar. These observations lasted until pupation. The duration of the pupal stage was recorded. Sex of adults was determined by presence of a depression (concavity) of the 1st and 2nd ventral abdominal segments of males, in many specimens accompanied by brownish coloration. In contrast, these same segments in females are flat and black. The sex ratio was observed in 116 newly emerged adults.

## RESULTS

Eggs were laid individually, in horizontal slits cut in pineapple leaves. Slits were pocket-shaped, and most were on the lower sides of the inner (younger) leaves. Eggs were white when newly oviposited, then yellow, then light brown as they matured. Mean length was  $1.98 \pm 0.02$  mm, width  $0.97 \pm 0.01$  mm ( $n = 35$ ).

Five exuviae were collected from each larva reared from egg to pupa ( $n = 41$ ), indicating that *M. callizona* has 5 instars at least under the test conditions. The mean head capsule width ( $n = 30$  for each instar) for the first instar was 0.92 mm; second 1.21 mm; third 1.69 mm; fourth 2.10 mm; and fifth 2.73 mm (Table 1). Size classes for instars 1-3 were discrete, but the smallest instar 5 heads were narrower than the largest instar 4 heads (Table 1). Discrimination of field-collected specimens of these instars would be difficult.

The incubation period for *M. callizona* eggs ( $n = 51$ ) averaged 8.3 days (Table 2). The larval stage lasted 37.4 days ( $n = 41$ ). The first 4 instars were

TABLE 1. HEAD CAPSULE WIDTH (MM) FOR LARVAL INSTARS OF *M. CALLIZONA* REARED ON PINEAPPLE STEMS (N = 30 SPECIMENS).

| Instar | Width (mm)      |           | Growth Ratio |
|--------|-----------------|-----------|--------------|
|        | Mean $\pm$ SD   | Range     |              |
| 1      | $0.92 \pm 0.07$ | 0.80-1.00 | —            |
| 2      | $1.21 \pm 0.29$ | 1.10-1.30 | 1.32         |
| 3      | $1.69 \pm 0.08$ | 1.50-1.90 | 1.40         |
| 4      | $2.10 \pm 0.15$ | 2.00-2.50 | 1.24         |
| 5      | $2.73 \pm 0.21$ | 2.40-3.00 | 1.30         |

TABLE 2. MEAN DURATION OF THE DEVELOPMENTAL STAGES OF *M. CALLIZONA* REARED IN THE LABORATORY (26°C, 82% RH, 14:10 L:D).

| Stage        | Number reared | Duration (Days)  |       |
|--------------|---------------|------------------|-------|
|              |               | Mean $\pm$ SD    | Range |
| Egg*         | 51            | $8.27 \pm 1.04$  | 7-10  |
| Larva        |               |                  |       |
| 1st instar   | 50            | $5.00 \pm 0.72$  | 4-6   |
| 2nd instar   | 48            | $4.51 \pm 0.73$  | 3-6   |
| 3rd instar   | 47            | $4.43 \pm 0.67$  | 3-6   |
| 4th instar   | 43            | $5.67 \pm 0.62$  | 5-7   |
| 5th instar   | 41            | $17.80 \pm 1.48$ | 15-24 |
| Pupa         | 40            | $11.82 \pm 1.53$ | 9-15  |
| Egg To Adult | 40            | 57.49            |       |

\*Assumes eggs were 1-day old at time of collection.

of similar duration: first 5.0 days, second 4.5 days, third 4.4 days, and fourth 5.7 days, whereas the last was much longer and more variable 17.8 days. The pupal stage ( $n = 40$ ) lasted 11.8 days. The pupa weighed on average  $0.12 \pm 0.03$  g ( $n = 20$ ). The mean development time from egg to adult for *M. callizona* ( $n = 40$ ) was 57.5 days (8 weeks). Among 116 newly-emerged adults (40 reared individually in this study, and 76 obtained from pupae taken from the greenhouse culture), 66 were females and 50 males, resulting in a ratio of 1:1.32 (male:female), but not significantly different from the expected 58:58 ( $\chi^2 = 1.10, P = >0.2$ ).

## DISCUSSION

Development time (8 weeks through all immature stages) was shorter than the 11 weeks obtained when larvae were reared on small *Tillandsia utriculata* bromeliads in the laboratory (Frank & Thomas 1994). This probably is due to better physical and/or nutritional conditions. The same room temperature and light regimen were used for the earlier rearing, but small *T. utriculata* plants were placed into small cages for convenience. In nature in Florida, *M. callizona* typically develops in large *T. utriculata* plants (Frank & Thomas 1994). Plants of that species but under an undefined size (suggested here to be roughly the size that will fit easily within a cylinder measuring 13.7 cm in height and 8.5 cm in diameter) provide inadequate nutrition for larvae. Current rearings were made at high humidity, whereas earlier rearings were made in small plants in screen-covered cages in an air-conditioned room. Although those plants were misted occasionally with water to keep them alive, they and the insects may have been stressed by desiccation. The difference in nutritional value be-

tween pineapple stems and stems of small *T. utriculata* plants is unknown.

Calculation of minimum generation time simply adds  $x$  days (pre-ovipositional period of females) to the development time of the immature stages. However, calculation of mean generation time adds  $y$  days (the mean time from emergence of an adult female to the oviposition of her median egg) to the development time of the immature stages. We do not know either  $x$  or  $y$ .

All stages seem to be present throughout the year in southern Florida (Frank & Thomas 1994). Although cool winter temperatures must increase development time, they do not seem to induce diapause or synchronize generations. Minimal generation time in Florida might approach 10 weeks (5 generations per year), but mean generation time is more likely to be 13-17 weeks (3-4 generations per year) in part because of slower development in the cooler months. At all events, the generations are not discrete in nature in Florida.

The remaining discussion (below) concentrates on contrasts between *M. callizona* and other *Metamasius* species of the weevil subfamily Rhynchophorinae, which also includes *Cosmopolites*, *Rhynchophorus*, and *Sphenophorus*. The principal contrast is with the best-studied *Metamasius* species, *M. hemipterus* (L.), a widespread, polyphagous, Neotropical weevil, best known for its damage to sugarcane and bananas, which has been present in Florida since at least 1984 (Woodruff & Baranowski 1985). Its adults are of similar size to those of *M. callizona*. Another studied species is *M. ritchiei* Marshall, which is known from Jamaica and Cuba. Its natural hosts presumably are native bromeliads, but in Jamaica it has been reported to attack cultivated pineapple, which is not native (Gowdey 1923).

#### Oviposition

The style of oviposition, in which an egg is placed in a pocket cut into a leaf by the adult female, is known in other Curculionidae. It was seen in *Cionus* and *Cleopus* (Cioninae) and *Eugnamptus* (Rhynchitinae) by Howden (1995) who called it "Category 4" (among various oviposition behaviors of weevils) and noted that the pocket is cut by the female's mandibles.

#### Number of Larval Instars

Unlike many insect families such as Carabidae and Culicidae, the Curculionidae do not share a fixed number of larval instars. Not only is there interspecific variation, but even intraspecific variation. Restrepo et al. (1982) indicate variation within *M. hemipterus* as 7-9 larval instars. The indication by Risco (1967) of only 3 larval instars within the same species is devoid of data. For these reasons, we cannot guarantee that the 5

instars that we observed in *M. callizona* are immutable under all circumstances. *Rhynchophorus palmarum* (L.) has 11-13 instars (Restrepo et al. 1982).

#### Fecundity and Incubation Time

Risco (1967) specified that larvae of *M. hemipterus* in Peru hatched after an incubation period of 7-10 days. Without further discussion, Restrepo et al. (1982), using a sugarcane substrate in the laboratory in Colombia, stated that female *M. hemipterus* had a pre-mating period of 1 day, then laid 544 eggs during their ovipositional period of 34 days, 90% of eggs were viable, and egg incubation time was 2-3 days (remarkably shorter!). The incubation time that we obtained for *M. callizona* was 8.3 days. *Rhynchophorus palmarum* laid a maximum of 880 eggs, and its incubation time was 2-3 days (Restrepo et al. 1982), but *R. cruentatus* (F.) females laid  $26 \pm 15$  eggs, which had an incubation time of  $69 \pm 17$  hours (Giblin-Davis et al. 1989).

#### Pupal Duration

Frank & Thomas (1994) obtained a pupal duration of 8-24 days for *M. callizona*, with mean  $\pm$ SD of  $15.6 \pm 6.4$  days for the first 15 pupae reared at 26°C with uncontrolled humidity. Current observations at high humidity reduced the range to 9-15 days with mean  $11.8 \pm 1.5$  days. Pupal duration of *M. ritchiei* is reported to have a range of 18-24 days (Gowdey 1923), and that of *M. hemipterus* is reported as 10 days (Woodruff & Baranowski 1985), 14 days (Restrepo et al. 1982), and 22-24 days (Risco 1967). It is likely that lower temperature prolongs the pupal period, and lower humidity may do the same. The pupal duration of *Rhynchophorus palmarum* was 20-38 days (Restrepo et al. 1982).

#### Life Cycle

For *M. callizona* in Florida, we suggest a minimal generation time of 10 weeks with mean generation time of 13-17 weeks. *Metamasius ritchiei* is reported to have a larval developmental period of 8-10 weeks, and a pupal period of 18-24 days, thus a minimal developmental period of 11+ weeks (Gowdey 1923). Data from Restrepo et al. (1982) suggested a minimal generation time of 9 weeks for *M. hemipterus* in Colombia, with mean generation time of 11+ weeks (this obtained by adding half the duration of the 34-day ovipositional period). Risco (1967) gives a minimum development time of 12-18 weeks (egg + larva + pupa) for *M. hemipterus* in sugarcane in Ecuador. It is not clear to what extent these differences are due to interspecific differences and to what extent to different rearing conditions.

## ACKNOWLEDGMENTS

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## A NEW *HYDROMETRA* SPECIES FROM ARGENTINA (HETEROPTERA: HYDROMETRIDAE)

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### ABSTRACT

A new Hydrometridae species, *Hydrometra quadrispina*, is described here. The most distinctive characteristics are: Male VIIIth abdominal segment bears 4 ventral spines, a dorsal white stripe runs all along the body in both sexes, and the anteclypeus is broad and slightly pointed. Several other differences were found between this species and the sympatric *H. argentina* Berg and *H. sztolemani* Jaczewski. *Hydrometra quadrispina* seems to have a very restricted distribution in a relict of the marginal forest in Punta Lara (Buenos Aires, Argentina). This area is threatened by a future bridge project to Uruguay.

Key Words: Neotropical, new species, systematics

### RESUMEN

Una nueva especie de Hydrometridae, *Hydrometra quadrispina* es aquí descrita. Los caracteres más distintivos son: Segmento VIII del macho con 4 espinas ventrales, una banda blanca corre dorsalmente a lo largo de todo el cuerpo en ambos sexos, y el anteclepeo es ancho y apenas puntiagudo. Varias otras diferencias fueron encontradas entre ésta especie y las simpátricas *H. argentina* Berg y *H. sztolemani* Jaczewski. *Hydrometra quadrispina* parece tener una distribución muy restringida en un relicto de la selva marginal en Punta Lara (Buenos Aires, Argentina) estando este ambiente amenazado por el proyecto de un puente a Uruguay.

According to Bachmann (1976, 1998), three Hydrometridae species are found in Argentina: *H. argentina* Berg, 1879, *H. sztolemani* and *H. fruhstorferi* Hungerford & Evans, 1934. The latter was recorded only from the Northeast of the country, quite near Brazil (Misiones). *H. sztolemani* has a wide distribution, being recorded from Brazil and Paraguay to Buenos Aires, Argentina (35°S). *H. argentina* also has a broad range throughout the Neotropical region, from Panama and Trinidad and Tobago Islands to Argentina (where it is the most common species); it is the southernmost of Neotropical Hydrometridae species (Rio Negro province, 37°S, according to Bachmann, 1976).

The Neotropical Hydrometridae were revised by Hungerford & Evans (1934) together with those of the world. Scattered information was provided by Drake (1950, 1953, 1954). A later complete checklist and description of some new species was given by Drake & Lauck (1959). Bachmann published a short paper with brief descriptions of the Argentinean species (1976). The latest information concerning this family in Argentina is summarized by Bachmann (1998).

*Hydrometra quadrispina* Perez Goodwyn, **New Species**

Terminology of morphology follows that of Polhemus & Polhemus (1995) and Polhemus and Lansbury (1997) based in Andersen (1982).

Color: Dorsal ground color when alive dark green (fuscous when fixed), females lighter than males, with white stripe running all along mid dorsal line of body, in both sexes (Fig. 1). Ventral ground color yellowish brown, fuscous narrow stripe, more evident in females, runs along mid line from little behind third pair of coxae approximately to intersegmental line between VIIth and VIIIth abdominal sternite. Body stippled by tiny brown tubercles, except abdominal tergites as well as the dorsal thorax mid line, which are bare, following white stripe. Thin bright setae scattered over entire body.

Head following described pattern, white stripe being wider immediately after antennal tubercles, almost reaching sensory bristles. Antennal tubercles shining dark brown, as well as base of sensory bristles. Anteclypeus white, with its border dark brown in dorsal view. Maxillary plate white, gular lobe translucent (Figs. 2 and 3).

Pronotum lighter than thorax, posterior boundary delineated by even lighter color.

Legs yellowish brown, with distal portion of femora and tibiae darker. Tarsi almost black.

External and internal boundaries of connexivae dark brown, forming 4 lines along abdomen in dorsal view, except for VIIIth segment. Lateral dark brown stripe running along ventral boundary of connexiva. VIIth tergite, all VIIIth segment, and proctiger clothed with long hairs. Male VIIIth sternite bearing rounded depressions, one

on each side of ventral mid line, both bare with hairs, lighter than ground color (Figs. 4 and 5).

Structure: all measurements are given in mm.

Overall length ♂:  $9.90 \pm 0.30$ ; ♀:  $11.10 \pm 0.40$ .

Head long ( $2.57 \pm 0.25$ ), wider at antennal tubercle ( $0.38 \pm 0.02$ ). Maxillary plate quadrangular, not prominent but distinctly colored. Gular lobe large, rounded, and covering base of rostrum. Rostrum exceeding ocular line, about  $0.625$  of postoculus. Antecolus to postoculus ratio  $1.72 \pm 0.15/0.88 \pm 0.20$ . Interoculus slightly narrower than width of eye ( $0.13/0.16$ ) bearing longitudinal furrow, more evident in females. Anteclypeus moderately large, conical, blunt. Antennal formula I, II, III, IV ( $0.43 - 1.05 - 2.10 - 1.10$ ).

Prothorax with row of pits the anterior lobe, forming collar (Fig. 1), few irregularly scattered pits on posterior lobe. Pronotum length  $1.45 \pm 0.20$ , remainder of thorax  $1.25 \pm 0.15$ . Distance between coxae I and II:  $0.70$ , between coxae II and III:  $1.35$ . Anterior and middle acetabulae each with one pit on anterior and posterior parts. Proportions of legs as follows: Femur, tibia, tarsal I, tarsal II, tarsal III. Fore leg:  $2.37/2.65/0.07/0.22/0.22$ . Middle leg:  $2.95/3.0/0.07/0.27/0.26$ . Hind leg:  $3.40/4.25/0.07/0.24/0.23$ .

Abdomen length. ♂:  $4.40 \pm 0.20$ ; ♀:  $5.50 \pm 0.20$ . Male posterior boundary of VIIth segment lined by setae, setting off bristle collar (Fig. 5). Female VIIth and VIIIth tergites covered by long hairs, VIIth sternite bearing pair of tufts of slender, faint hairs on both sides of mid line (Figs. 6 and 7).

Male terminalia as shown in Figs. 4 and 5. The VIIth sternite bears "spines" (tufts of setae resembling spines, following cited authors), all about same size, projected caudad; the external ones nearer proximal edge of VIIth segment (Figs. 4 and 5).

Distribution: Known only from type locality. Argentina, Buenos Aires, Ensenada, Punta Lara marginal forest, pond by the road, shaded by trees, and most of the year covered by pleuston. ( $34^{\circ}47', 05'S$ ;  $58^{\circ}00', 49'W$ ) about  $0.5$  km away from the Rio de La Plata estuary.

Examined material: all micropterous, no brachypterous or macropterous forms were found. Holotype ♀ 28/XII/97, allotype ♀ 28/XII/97 (MLP). Paratypes 3 ♀ 28/XII/97 P. P. Goodwyn (MLP); 1 ♂ 19/XII/97, Ellenrieder; 2 ♂ 17/XII/97; 4 ♀ 3/I

98 P. P. Goodwyn (MACN), 1 ♂ 12/IV/98 (ZSM) P. P. Perez Goodwyn.

The holotype, allotype and 6 paratypes are deposited in the Museo de La Plata (MLP). 6 paratypes in the Museo Argentino de Ciencias Naturales "Bernardino Rivadavia" (MACN), and 1 paratype in the Zoologische Staatssammlung München, Deutschland.

#### DISCUSSION

*Hydrometra argentina* is found over a wide range, but was almost absent in these small shaded ponds of the forest. Nevertheless it was the only species present just  $1$  km. away, in sunny ponds, shaded only by bushes. Occasionally, *Hydrometra sztolcmani* was found in both kinds of environments.

*Hydrometra quadrispina* specimens are conspicuous in the field by their white mid-dorsal line. Males are easily distinguished from both sympatric species by the presence of four spines.

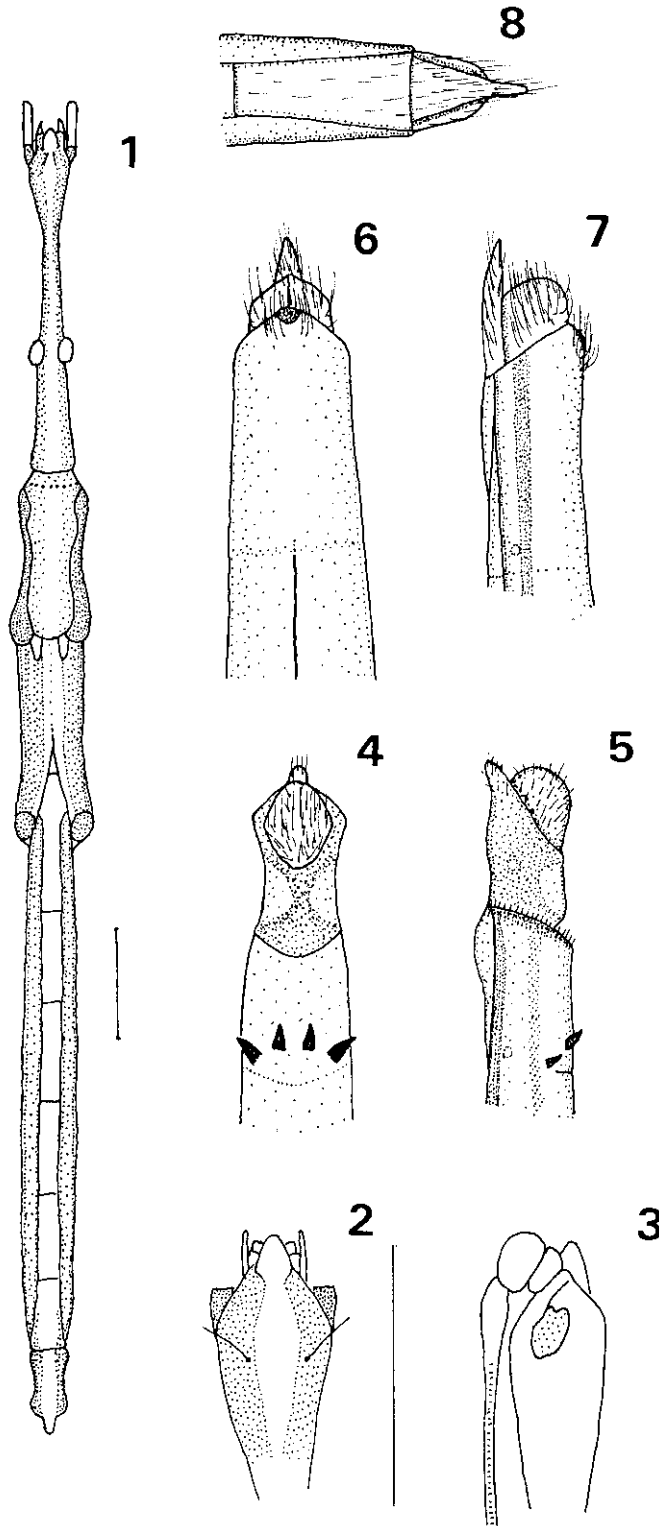
Females of *H. argentina* and *H. quadrispina* are very similar, but they can be distinguished because the terminal process of *H. argentina* is slightly longer and more pointed, and the distal midventral boundary of the VIIIth segment is pointed, whereas in *H. quadrispina* it is blunt. There are slight differences in the furrow in the interoculus; that of *H. quadrispina* is more developed, particularly in females. The anteclypeus of these 2 species is distinctive; it is more slender in *H. argentina*. The maxillary plate of *H. argentina* is larger and darker. Measurements did not reveal significant differences between *H. argentina* and *H. quadrispina*, except for the tarsus II of the middle and hind legs, both are longer in *H. argentina* ( $0.38$  and  $0.40$ , respectively).

*Hydrometra quadrispina* was only found only a few times in a restricted area during 2 years of intensive sampling in a place that had been sampled by former investigators for many years; thus it can be considered "rare". This environment is a relict of a previously extensive marginal forest now endangered in its southern limit by human activities. The exact collection site probably will be destroyed soon by a huge bridge that will join Argentina with Uruguay.

#### KEY TO SPECIES OF *HYDROMETRA* IN ARGENTINA

- 1 Longer than  $12.5$  mm, dark overall color, males bearing 2 semilunar tufts of hairs in the VIIth sternite. Misiones Province. . . . . *H. fruhstorferi*
- 1' Not longer than  $12$  mm, males never bearing 2 semilunar tufts of hairs in the VIIth sternite . . . . . 2
- 2 Male VIth segment with 2 large lateral processes overlapping the posterior margin of the Vth segment. Female terminal process blunt . . . . . *H. sztolcmani*
- 2' Male terminalia without processes, only bearing spines. Female terminal process pointed. . . . . 3
- 3 Both sexes with a mid-dorsal pale line, male VIIth sternite with 4 spines . . . . . *H. quadrispina*
- 3' Uniform dorsal color, male VIIth sternite with 2 spines . . . . . *H. argentina*





Figs. 1-8: *Hydrometra quadrispina* Perez Goodwyn. 1: Male dorsal habitus, legs omitted. Scale 1 mm. 2-3: Head: 2, dorsal view; 3, lateral view, antennae omitted. 4-5: Male terminalia: 4, ventral view; 5, lateral view. 6-8: Female terminalia: 6, ventral view; 7, lateral view; 8, dorsal view. Figures 2-8 scale 1 mm.

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*MELANOPLEUROIDES DOMINICANUS*, A NEW LYGAEINE GENUS AND SPECIES FROM THE DOMINICAN REPUBLIC (HETEROPTERA: LYGAEIDAE)

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ABSTRACT

*Melanopleuroides dominicanus* is described from the Dominican Republic. Characteristics to separate it from similar taxa, genitalic drawings, and a brief discussion of its phylogenetic relationships are provided.

Key Words: West Indies, new genus, new species, Heteroptera, Lygaeinae

RESUMEN

*Melanopleuroides dominicanus* es descrito de la Republica Dominicana. Los caracteristicas que lo separan de taxa similar, dibujos genitales, y una discusión breve de sus relaciones filogenéticas son proveidas.

The Western Hemisphere members of the lygaeid subfamily Lygaeinae were revised and their phylogenetic relationships discussed at the genus level by A. Slater (1992). Specimens of this species failed to key to any genus in the key provided in that paper. As the general appearance is very similar to members of the genus *Melanopleurus* Stål it was first thought that a revision of the key might be necessary. Inspection of the genitalia shows sufficient differences from those of *Melanopleurus* and other described genera to warrant treatment as a new genus. There are over 270 species of Lygaeidae known from the West Indies with 112 from the Dominican Republic including 18 Lygaeinae of which 7 are endemic.

All measurements are given in millimeters.

*Melanopleuroides* A. Slater and Baranowski,

**New Genus**

Type species: *Melanopleuroides dominicanus* A. Slater and Baranowski. Monobasic.

Moderately robust; impunctate except immediately before and behind callus and on propleuron and sternum; hairs short, longest on hemelytra, semidecumbent, moderately dense except nearly absent at base of corial disc. *Head* fairly strongly declivent; vertex convex; ocellus small, slightly raised above surface, distance between ocelli about 10 times distance from ocellus to eye; bucculae moderately produced, ventral margin slightly convex. *Pronotum* with posterior width about one and one half times anterior width; medial length slightly less than anterior width; anterior margin concave, very slightly raised laterally, not beaded, separated from callus by coarsely punctate but not depressed area; pos-

terior margin slightly convex, with shallow but distinct depression laterally; lateral margin slightly sinuate; callus indistinct, defined primarily by punctate areas immediately before and behind, callar impressions unbranched, sinuate, angled towards anterior pronotal angles; depression behind callus shallow, coarsely punctate, interrupted medially by indistinct carina, reaching lateral margin. *Scutellum* slightly more than half as long as wide; stem and arms of median carina broad; lateral fovea distinct basally becoming indistinct apically, bottom grooved. *Hemelytron* slightly surpassing apex of abdomen; corial veins indistinct; membrane opaque. *Propleuron* divided into three parts by dorsoventral impressions, anterior and posterior parts coarsely punctate, median part impunctate, slightly convex. *Meso-pleuron* divided into anterior and posterior parts but shallow dorsoventral impression, impunctate. *Metapleuron* impunctate; posterior margin distinctly concave; ostiolar peritreme with anterior and posterior margins about equally high and with apical button distinctly separated from surrounding pleuron dorsally and posteriorly; evaporative area restricted to base of peritreme. *Abdomen* without anterolateral scars; sternum II fully exposed; sternum VI of female about as long medially as laterally; sternum VII of female cleft to base.

*Clasper* (Fig. 1c) with blade at slightly oblique angle to shank, curved, flattened, inner face only slightly ventrally directed; posterior projection distinct, conical; shank flattened, without rooflike interior projection; pair of sharply defined ridges separated by groove on interior side at base of blade. *Aedeagus* (based on very incomplete inflation) bent before telescoping into phallosome; con-

junctiva with lateral lobe on each side bearing broad flat sclerite; at least two large lobes bearing single heavy apical spines either apical on conjunctiva or on vesica; gonoporal process short, thick, without secondary process; phallosome without apical flange, process, process a short c-shaped flange broadest dorsally. *Spermatheca* (Fig. 1d) with basal tube short, without distinct swollen area; apical tube swollen, without distinct bulb, continued as very thin tube apically. *Ovipositor* short; first valve (Fig. 1a) with connecting membrane extending beyond apex of valvula, median cleft reaching almost to base, without Y-sclerite, strongly pigmented or lightly sclerotized area from membrane apex almost to base parallel to valvula; second valve (Fig. 1b) with connecting membrane cleft almost to base, heavily pigmented or lightly sclerotized mesad of valvula.

This species keys to *Melanopleurus* in A. Slater's (1992) key. The general appearance is similar to some of the smaller members of that genus. The two genera can be easily separated by the presence in *Melanopleurus* of a distinct pale macula on the vertex of the head and by the distinctly concave posterior metapleural margin of *Melanopleuroides* which contrasts with an almost straight posterior margin in *Melanopleurus*.

In view of the very incomplete inflation of the aedeagus exact placement on A. Slater's (1992) proposed phylogenetic tree seems inappropriate at this time, some characteristics of the genitalia are worthy of mention. The general development of the conjunctival and vesical lobes and spines places this genus in the group containing the genera *Latochrinnus*, *Lygaeospilus*, *Melacoryphus*, *Dalmochrinnus*, *Achlyosomus*, *Craspeduchus*, and *Melanopleurus*. Within that grouping the apparent development of the conjunctival and vesical spines and the shape of the phallosome flange are similar to those of *Melanopleurus* and *Craspeduchus*. The lack of a Y-sclerite on the first valve of the ovipositor is shared with *Latochrinnus* and *Achlyosomus*. The ovipositor is similar in general fascies to that of *Latochrinnus* but that genus lacks sclerotized or heavily pigmented areas on the membrane. The position of the interior ridges and groove on the clasper is unique, they may be homologous with the ridges and grooves present on the shank of the clasper in other lygaeines. The long thin apical tube on the spermatheca is also unique though there is some resemblance to the spermathecae of *Dalmochrinnus* and *Achlyosomus* in which the apical tube is much thicker at its base than at its apex. It could be argued that there is a progression from the distinct basal swelling in *Melanopleuroides* through a basal thickening in *Dalmochrinnus* and *Achlyosomus* to a completely thin apical tube in *Craspeduchus* and *Melanopleurus*.

Known only from the Dominican Republic.

**Etymology:** Named to show the external similarity to *Melanopleurus*.

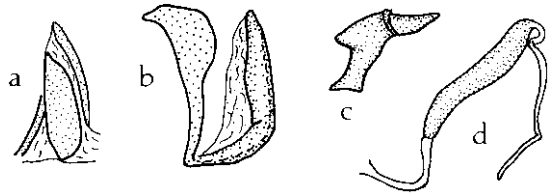


Fig. 1. a. Ovipositor, 1st valve, ventral view; b. Ovipositor, 2nd valve, lateral view; c. Right clasper, interior view; d. Spermatheca.

*Melanopleuroides dominicanus*

A. Slater and Baranowski, **New Species**

Head black except for pale yellow ventral margin of bucculae. Pronotum and scutellum black. Clavus and corium dark brown to black except for base and outer two thirds of apical margin orange yellow. Membrane dark brown with a pale macula on lateral margins. Thoracic pleura black except margins pale yellow. Posterolateral metapleural angle orange yellow. Abdomen dark brown to black. Legs and antennae black except for antennal segment IV blackish brown.

Body, legs and antennae with silvery appressed hairs.

Length head 0.80, width 1.13, interocular space 0.70. Pronotum trapezoidal, anterior margin concave. Length pronotum 0.88, width 1.55. Scutellum with T-shaped callosity. Length scutellum 0.43, width 0.86. Length claval commissure 0.48. Midline distance apex clavus-apex corium 1.03. Midline distance apex corium-apex membrane 1.03. Labium extending between metacoxae. Length labial segments I 0.45, II 0.55, III 0.50, IV 0.25. Length antennal segments I 0.30, II 0.63, III 0.53, IV 0.65. Total body length 4.60.

**Holotype:** Male. Dominican Republic: Pedernales, Parque Nacional del Bahoruco, 10 July 1996, R. Turnbow. In Florida State Collection of Arthropods.

**Paratypes:** 1 ♂, 3 ♀♀, DOMINICAN REPUBLIC: Monte Cristi, 8.2 km N Villa Elisa, 31 May 1994, R. Turnbow; 2 ♀♀, Monte Cristi, 8.6 km N villa Elisa, 26 May 1992, R. Turnbow; 1 ♂, 1 ♀, La Vega Prov. Hotel Montana, 25 May 1992, R. Turnbow; 1 ♂, 1 ♀, Pedernales, 23-24 km N Cabo Rojo, 535 m, 1 July 1996, R. Turnbow. In University of Georgia, James A. Slater and Richard M. Baranowski collections.

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REFERENCES CITED

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## A NEW GENUS AND FIVE NEW SPECIES OF NEOTROPICAL LETHAEINI (HETEROPTERA: LYGAEOIDEA: RHYPAROCHROMIDAE)

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### ABSTRACT

*Neopetissius*, new genus, with type species *Neopetissius slaterorum*, new species, from Brazil, British Honduras (Belize), Colombia, Ecuador, El Salvador, Guatemala, Mexico, Panama, Surinam, Trinidad and Venezuela, is described. Four additional species are described and figured: *Neopetissius froeschneri*, new species, from Bolivia, Brazil, Panama and Peru; *Neopetissius immanis*, new species, from Brazil; *Neopetissius perplexus*, new species, from Bolivia and adjacent Brazil; and *Neopetissius variegatus*, new species, from the West Indies. Morphological characters are discussed in a phylogenetic context.

Key Words: Rhyparochromidae, Lethaeini, Neotropics

### RESUMEN

El nuevo genero, *Neopetissius*, con especie tipo *Neopetissius slaterorum*, nueva especie, de Brasil, Honduras Inglesa (Belice), Colombia, Ecuador, El Salvador, Guatemala, México, Panamá, Surinam, Trinidad y Venezuela, es descrita. Cuatro especies adicionales son descritas e ilustradas: *Neopetissius froeschneri*, nueva especie, de Bolivia, Brasil, Panamá y Perú; *Neopetissius immanis*, nueva especie, de Brasil; *Neopetissius perplexus*, nueva especie, de Bolivia y Brasil adyacente; y *Neopetissius variegatus*, nueva especie, de las antillas caribeñas. Características morfológicas son descritas en un contexto filogenético.

This paper describes new Neotropical Lethaeini in order to make names available for a large paper on the West Indian lygaeid (sensu lato) fauna in preparation by J. A. Slater and R. M. Baranowski. It is surprising that these large, apparently widespread bugs have not yet been described, given the amount of material in museum collections. They have long been an enigmatic group, as evidenced by the various determination labels attached to specimens over the years by respected students of the (then) Lygaeidae. Examples of these include: "*Lethaeus*, det. Barber;" "*Lethaeini* nr. *Petissius*, det. Ashlock;" "*Petissius assimilandus*, det. Ashlock;" "*Petissius* sp., det. Barber;" "*Gonatoides* n.sp. #1 and #3, det. Sweet;" and "*Cistalia* sp.???", det. Ashlock." This confusion over identity is also a testament to the difficulty in establishing generic limits in the Lethaeini, despite my earlier attempt (O'Donnell 1986) to do so.

My decision to describe a new genus for these new species, even though generic limits are ambiguous, is based on the fact that they show a number of features that seem to preclude them from inclusion in any existing genus. Of the four described genera listed above, *Lethaeus* is easy to eliminate. It is strictly an Old World genus, with a double, striated iridescent spot on the top of the head. The new species described herein all have a single, non-striated spot, as do species in the other three genera listed above. They are all part of the

"one-spot clade" a monophyletic unit within Neotropical lethaeines that includes *Cryphula*, *Paragonatas* and *Rhaptus* in addition to *Cistalia*, *Gonatoides* and *Petissius* (O'Donnell 1986). *Rhaptus* is a monotypic genus with autapomorphies (somewhat dorsoventrally flattened, greatly enlarged fore femora) that easily eliminate it from further consideration as a congener of the new species.

It is not as easy to dismiss placement of the new species in some of the other "one-spot" genera. There are several reasons. First, phylogenetic relationships among genera of this clade remain unsatisfactorily resolved: *Paragonatas* is polyphyletic, and *Petissius* and *Gonatoides* paraphyletic, in my (1986) phylogenetic analysis. Second, polarization of the defining synapomorphy (possession of a single median iridescent spot as apomorphic) is itself equivocal. Third, although the single spot occurs only in the New World, some of the characters that appear apomorphic in the one-spot clade also occur in taxa from other zoogeographic regions—a clear indication that revision of generic limits should proceed at the world level.

Therefore, *Neopetissius*, new genus, is circumscribed by the following putative synapomorphies: 1. Broadly explanate lateral pronotal margins. The margins characteristic of *Neopetissius* are flared upward slightly, and are expanded nearly equally for their entire lengths; in addi-

tion, a prominent transverse pronotal impression imparts a partially concave, often even sinuate, lateral edge (see below). The explanate lateral pronotal margins of *Petissius*, by contrast, are narrowed anteriorly and become almost obsolete posteriorly. The lateral edge is smoothly convex, hardly, if at all, indented between the anterior and posterior pronotal lobes. The lateral pronotal margins of *Gonatoidea*, while broadly expanded, are not strongly differentiated from the remainder of the pronotum and are also smoothly convex. *Paragonatas* and *Cistalia* have carinate, but not explanate, lateral pronotal margins. 2. Transverse impression. Species of *Neopetissius*, in contrast to all other neotropical Lethaeini of the one-spot clade, have a prominent transverse impression that divides the pronotum into distinct anterior and posterior lobes. Some species also have a marked longitudinal impression, an unusual feature not only for lethaeines but for rhyparochromids in general. 3. Pronotal collar. *Neopetissius* has a prominent, triangular pronotal collar, set off from the remainder of the pronotum by a groove of punctures. This condition is found in several apparently otherwise unrelated Old World genera, but is not found elsewhere in the one-spot clade.

The following additional features may be of generic significance. 1. Tuberculate femoral hairs. These are short hairs set on oblique tubercles, covering the postero-ventral surface of at least one femur. They are not as apparent on females. 2. Swollen venter of head. The underside of the head is swollen on either side of the labium, especially in males. This swelling takes various forms, but again, evaluation of character states needs to proceed across hemispheres because Old World taxa also exhibit several states, and homology is uncertain.

The male genitalia, in general so useful for establishing generic limits in the Lethaeini, are of limited value in the one-spot clade, and especially among the new species. They are among the most complicated and asymmetrical of any in the tribe, and seem to combine features of several genera (complete arcuate extension; corrugations, sleeve and wings, all present). In fact, the male genitalia of one species (*N. immanis*, n.sp.) are different enough that status as a separate genus may be warranted in the future. This group is yet another case where insect species quite alike in external appearance have very different male genitalia. It is likely that additional species will be discovered as more material becomes available for dissection and zoogeographic analysis.

#### MATERIALS AND METHODS

Specimens were borrowed from and/or deposited in the following collections: American Museum of Natural History, New York, NY (AMNH); The

Natural History Museum, London, UK (BMNH); Carnegie Museum, Pittsburgh, PA (CARN); California Academy of Sciences (CAS); Instituto Nacional de Pesquisas de Amazonia, Manaus, Brazil (INPA); Institut Royal des Sciences Naturelles de Belgique, Brussels, Belgium (IRSN); James A. Slater Collection, University of Connecticut, Storrs, (JAS); Rijksmuseum van Natuurlijke Historie, Leiden, Netherlands (LEID); National Museum of Natural History, Smithsonian Institution, Washington, DC (NMNH); David Rider, North Dakota State University, Fargo, ND (RIDER); Museu Nacional, Rio de Janeiro, Brazil (RIO); Richard M. Baranowski, University of Florida, Homestead, FL (RMB); Snow Entomological Museum, University of Kansas, Lawrence, KS (SNOW); Merrill Sweet, Texas A&M University, College Station, TX (SWEET); Texas A&M University, College Station, TX (TAMU); University of Missouri, Columbia, MO (UMC); University of California, Davis (UCD); University of Michigan, Ann Arbor, MI (UMAA); University of Connecticut, Storrs, CT (UCMS); Universidad Central de Venezuela, Maracay, Venezuela (VENZ).

Techniques for dissecting genitalia follow O'Donnell (1991), with the following modifications: specimens were relaxed in boiling water instead of relaxing fluid; spermathecae were stained with BioQuip's double stain to render lightly sclerotized areas more readily visible; and dissected genitalia were positioned on a bed of 0.25 mm glass beads immersed in 70% ethanol, which held the pieces steady for drawing. Names of colors follow Smithe (1975, 1981). All measurements are in millimeters.

#### *Neopetissius* O'Donnell, **New Genus**

Medium to large, elongate oval (males) to broadly oval (females). Surface dull to subshining. Head with one basal median iridescent spot dorsally, presumably composed of pegs. Lateral pronotal margins explanate, carinate, with trichobothrium on anterior third; pronotal collar triangular, well-defined, set off from remainder of pronotum by groove of closely spaced punctures; collar impunctate (*N. slaterorum* n. sp., *N. variegatus* n. sp.) or punctate (other species); transverse pronotal impression well developed, longitudinal impression variable. Field of tuberculate hairs present posteroventrally on femora. Clasper usually compressed on outer surface of outer projection. Sperm reservoir variable, with sleeve prominent and separate from vesical seminal duct (except *N. immanis*, n. sp.). Spermatheca with broad duct.

Type species: *Neopetissius slaterorum*, new species.

Etymology. "Neo-" meaning "new" with "*Petissius*" its close relative. Masculine.

Distribution. Widely in the Neotropics.

KEY TO SPECIES OF *NEOPETISSIUS*

- 1 Underside of head of male swollen (Fig. 1) into a forward-projecting tubercle (only males are known) . . . . . *immanis*, n. sp.
- 1' Underside of head of male not swollen into a forward-projecting tubercle . . . . . 2
- 2 Labium extending posteriorly beyond hind coxae, onto second abdominal segment in males, or third abdominal segment in females . . . . . *variegatus*, n. sp.
- 2' Labium not extending posteriorly beyond hind coxae . . . . . 3
- 3 Evaporative area on mesopleuron extending toward dorsal margin as a wide "tongue" (Fig. 2) . . . . . *slaterorum*, n. sp.
- 3' Evaporative area on mesopleuron extending toward dorsal margin as a narrow tongue (Fig. 3) . . . . . 4
- 4 Pronotal calli with fine punctures . . . . . *perplexus*, n. sp.
- 4' Pronotal calli impunctate . . . . . *froeschneri*, n. sp.

*Neopetissius froeschneri* O'Donnell, **New Species**  
Figs. 3, 4, 10, 11, 18

Medium size. Total length 6.4. Maximum width, at level of apex of clavus, 2.5. Dorsal surface subshining. Head, anterior pronotal lobe, first antennal segment, most of scutellum and all of femora very dark grayish brown. Most of remainder of dorsum, antennal segments II-IV and tibiae chestnut. Third antennal segment slightly paler distally but without a distinct pale annulus. Dorsum marked with buff yellow as follows: pronotal collar except laterally; lateral pronotal margins; anterior half of posterior pronotal lobe except at middle; humeri; small spot at posterior

pronotal margin midway from midline to humeral angle; elongate dash on clavus near apex of scutellum; lateral corial margins; corial veins proximally and distally (somewhat darkened across middle of corium); and spot at basal angle of Cu and R + M. Two obscure pale spots present between M and Cu. Inverted-heart shaped distal corial macula, veins and obscure apical macula on membrane cream color. Venter nearly uniformly dark grayish brown, shining on thorax, subshining on abdomen. Labium buff yellow.

Head porrect, flat across vertex; eyes large. Length head 0.75; preocular length 0.42. Width head 1.10; interocular width 0.58. First antennal segment incrassate, diameter greater than that of other segments, exceeding apex of tylus by half its length; 2 setae present proximally on inner surface. Antennal segments II-IV terete, with scattered upstanding hairs shorter than diameter of segment in addition to decumbent pubescence. Length antennal segments I 0.72; II 0.92; III 0.80; IV 1.00. Venter of head strongly swollen from level of apex of antenniferous tubercle to middle of eye; heavily and evenly coarsely punctate. Labium just reaching metacoxae; first segment reaching level of middle of eye. Length labial segments I 0.80; II 0.70; III 0.87; IV 0.57.

Pronotum with anterior margin shallowly concave; posterior margin essentially straight; lateral margins sinuate, broadly explanate, notched at posterior corner of humeri. Trichobothrium level with posterior edge of punctures defining collar at meson. Anterior lobe with prominent triangular collar that broadens mesally, set off by a row of closely-spaced punctures laterally and a broader band of punctures mesally. Anterior lobe with calli impunctate except for scattered punctures along inner margin of explanate lateral margins; transverse impression well-developed, especially laterally; longitudinal furrow very prominent along middle third of meson; posterior lobe with coarse, widely-spaced punctures at transverse impression grading to finer, more closely-

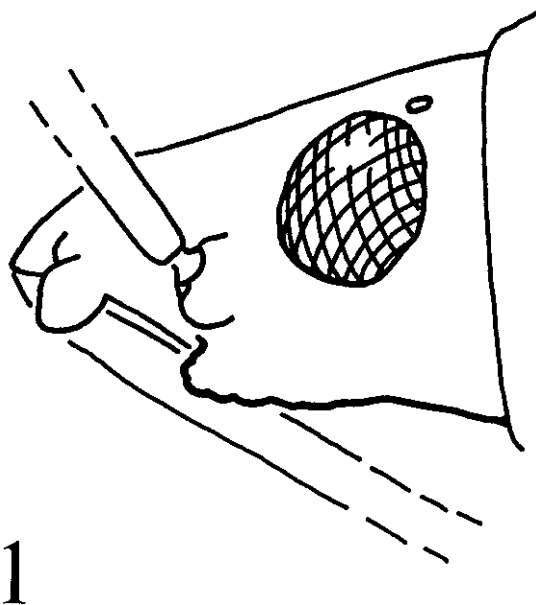


Fig. 1. *Neopetissius immanis*, n. sp., head, lateral view. Scale line equals 0.10 mm.

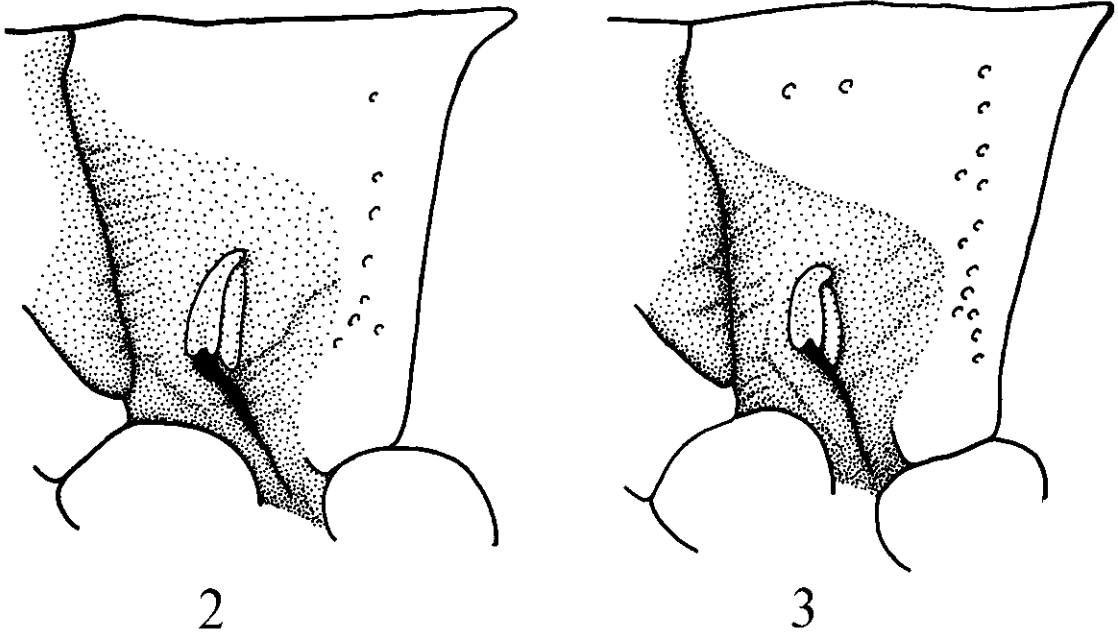


Fig. 2. *Neopetissius slaterorum*, n. sp., metathorax, lateral view. Fig. 3. *Neopetissius froeschneri*, n. sp., metathorax, lateral view. Scale line equals 0.10 mm.

spaced punctures posteriorly; humeri raised, prominent, impunctate, extending nearly to transverse impression. Length pronotum 1.30; posterior width 2.22; width across trichobothria 1.50. Scutellum elevated anteriorly and along lateral margins, depressed mesally; impunctate on lateral elevations, otherwise shallowly punctate anteriorly, more coarsely mesally and along lateral margins. Length scutellum 1.20; width 1.20. Hemelytron macropterous; clavus with 3 regular and 2 irregular rows of punctures. Corium with lateral margins explanate, sinuate; Cu bent sharply laterad just posterior to level of apex of scutellum; corial fracture mesal to R + M, ending at level of claval apex; R + M strongly raised to end of corial fracture; membrane slightly exceeding apex of abdomen, veins distinct; one cross-vein each between Sc and R and R and M. Length claval commissure 0.75; midline distance apex clavus-apex corium 1.30. Metathoracic scent gland with ostiolar peritreme slightly raised above metapleuron, remote from dorsal margin of evaporative area; long axis of peritreme paralleling meso-metapleural junction, apex bent at 45 degree angle; elevated lobe present on posterior side of auricle. Evaporative area rugose, covering all of mesoepimeron, extending narrowly along meso-metapleural suture nearly to dorsal margin of mesopleuron; occupying ventral  $\frac{2}{3}$  of metapleuron, also extending dorsally but not as far as on mesopleuron; dorsal margin on metapleuron sloping strongly ventrad posteriorly. Legs with all

femora densely covered with decumbent hairs, set on tubercles postero-ventrally on profemur; these tubercles inconspicuous on mesofemur. Fore femur the most swollen, armed below with 4 (right leg, 3 on left leg) stout spines distally and 3 elongate hair-spines in same row proximally; hind femur with row of widely-spaced short, semi-decumbent spines on upper surface and one longer spine near distal end ventrally. Fore tibia spinose on posterior surface only; tibiae and tarsi covered with decumbent pubescence.

Sterna covered with long, decumbent, silvery, widely-spaced hairs, somewhat denser along dorsal margin. Male clasper (Fig. 10) with inner projection very broadly rounded; outer projection indented on dorsal aspect; shank very short, nearly obsolete, with flange. Sperm reservoir (Fig. 11) with sleeve only moderately sclerotized; vesical seminal duct strongly sclerotized, coiling thickly and asymmetrically distally as it exits sleeve before becoming flat, with a thickened edge at transition point; wings, large, quadrate in lateral view; arcuate extension complete but faint across middle of bulb; corrugations present; holding sclerites absent. Spermatheca (Fig. 18) mushroom-shaped, with bulb sitting directly on narrow distal flange; duct diameter about  $\frac{1}{3}$  diameter of bulb; proximal flange lightly sclerotized, especially on distal edge, very asymmetrical, split on proximal side, flaring bell-like opposite split; duct offset. (A small sclerite proximal to proximal flange may be part of the spermatheca itself, or a



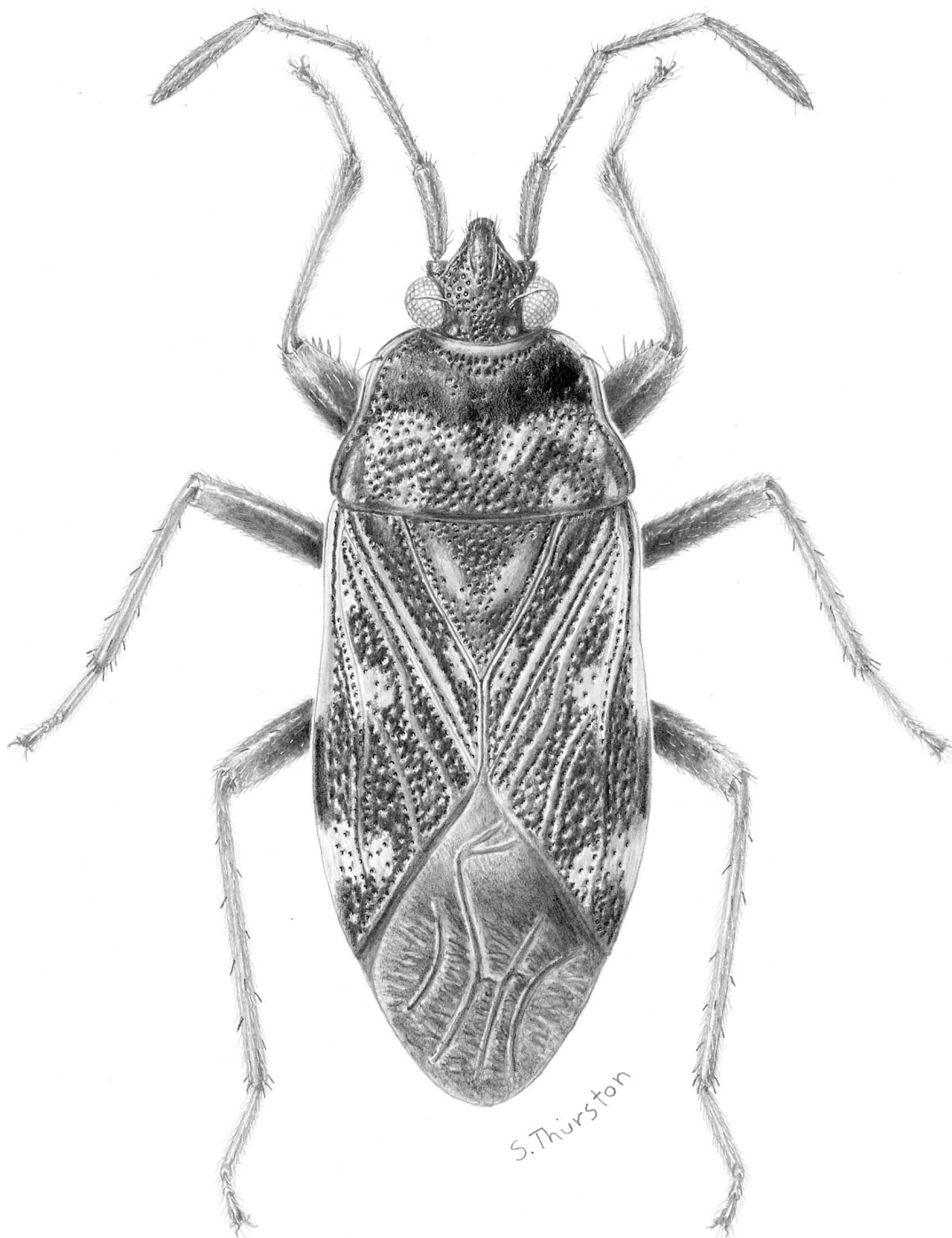


Fig. 4. *Neopetissius froeschneri*, n. sp., dorsal view. Scale line equals 0.10 mm.

piece of male genitalia that has broken off inside the female.)

Holotype: ♂, BRAZIL, Piracicaba, S. P., 13-III-1966, C. A. Triplehorn, blacklight (AMNH).

Paratypes: 1 ♂, same data as holotype except 25-XI-1965 (JAS); 2 ♂♂, 3 ♀♀, same data as holotype except 2-XII-1965 (one of each sex dissected and illustrated) (JAS,UCMS); 1 ♂, same

data as holotype except 19-XII-1965 (JAS); 3 ♂♂, 1 ♀, same data as holotype except 3-II-1966 (JAS, UCMS).

**Additional Material Examined:** BOLIVIA: 3 ♂♂, 3 ♀♀, Puerto Suarez, J. Steinbach, 150 m (CARN); BRAZIL: 3 ♂♂, 10 ♀♀, Bahia, Encruzilhada, XI-1972, M. Alvarenga, 960 m (AMNH); 1 ♂, 2 ♀♀, Santa Catarina, Nova Teutonia, 27°11'N, 52°23'W, 11-X-1961, Fritz Plaumann (SWEET); 2 ♂♂, 6 ♀♀ same except 11-X-1960 (JAS); 1 ♂, 1 ♀, same except III-1972 (VENZ); 2 ♀♀, same except II-1973 (VENZ); 1 ♀, same except 8-X-1963, no coordinates given (JAS); 1 ♀, same except 6-VII-1957 (JAS); 1 ♀, same except 6-X-1962 (JAS); 1 ♂, 1 ♀, same except 27-IX-1957 (JAS); 1 ♂ same except VI-1977, 300-500 m, 27°11'B, 52°23'L (UNAM); COSTA RICA: 2 ♀♀, Puntarenas, nr. Monteverde, 31 May 1988, J. O'Donnell, at light (UCMS); PANAMA: 3 ♂♂, 4 ♀♀, Canal Zone, Barro Colorado Island, XII-1946-II-1947, J. Zetek (NMNH); 1 ♂, same except I & II 1945 (NMNH); 1 ♂, El Real, 8-VIII-1952, F. S. Blanton (NMNH); 1 ♀, Las Cumbres, 7-I-1975, L. B. O'Brien, at night (JAS); PERU: 1 ♀, Loreto, km 3 Tournavista Rd., 34 km W Pucallpa, 17-XII-1971, R. T. & J. C. Schuh, 300 m, at light (AMNH); 1 ♀, same except 23-XII-1971 (AMNH).

**Etymology:** Named for Dr. Richard C. Froeschner, Curator Emeritus at the National Museum of Natural History, Smithsonian Institution, in recognition of his long and productive career as a heteropterist.

*Neopetissius immanis* O'Donnell, **New Species**

Medium sized, ovoid. Total length 7.60. Maximum width, at level of apex of clavus, 2.70. Body surface subshining, clothed above with short, cream color hairs arising from punctures. Head, anterior pronotal lobe, scutellum, first antennal segment and all femora dusky brown to dark grayish brown; posterior pronotal lobe, most of hemelytra, and remaining antennal segments chestnut; explanate lateral pronotal and corial margins buff yellow; tibiae buff yellow with tinges of chestnut at both ends. Hemelytron marked with cream color as follows: elongate dash along inner half of claval margin; two spots between R + M and Cu, and veins adjacent to posterior of these; small spots on apical corial margin at junction with clavus and at Cu; and inverted, subapical heart-shaped macula. Membrane almost uniformly chestnut, veins slightly paler. Venter dusky brown, becoming chestnut at posterior margins of pro- and metapleura and last 2 abdominal segments. Labial segment 1 predominantly cream color but tinged with chestnut; segments 2 and 3 cream color; segment 4 chestnut.

Head quadrate, correct, tylus reaching slightly less than half way to end of first antennal segment; area just ventrad of antenniferous tubercle

expanded into a cone-shaped protruberance (Fig. 1), extending as far anteriorly as end of antenniferous tubercle. Length head 1.10; width 1.32; interocular 0.65; preocular 0.70. Pronotum with anterior margin concave; triangular collar well defined by deep punctures; calli raised, with sparse, shallow punctures; transverse impression pronounced; longitudinal furrow deep; posterior lobe evenly and coarsely punctate, humeri prominent; posterior margin straight; lateral margins broadly explanate, sinuate, notched at humeri; trichobothrium located one-third of the way along lateral margin. Length pronotum 2.08; width 2.60. Scutellum with impunctate v-shaped elevation; length scutellum 1.20; width 1.25. Length claval commissure 0.88. Hemelytron macropterous, with widely explanate lateral corial margin tapering at level of claval apex. R + M strongly elevated, carinate. Clavus with 3 regular and 2 irregular rows of punctures; membrane reaching end of abdomen, with 4 prominent longitudinal veins and 2 cross veins between Sc and R. Midline distance apex clavus-apex corium 1.55. Metathoracic scent gland auricle hook-shaped; evaporative area covering ventral half of metapleuron, extending narrowly along meso-metapleural junction nearly to dorsal margin; dorsal margin of evaporative area sinuate. Labium extending between hind coxae, first segment just reaching base of head. Length labial segments I 1.30; II 1.20; III 1.18; IV 0.62. Antennae with first segment abruptly thickened beyond apex of tylus. Length antennal segments I 1.00; II 1.40; III 1.10; IV 1.40. Fore femur with a row of 5 short stout subdistal spines and several elongate hair-spines proximally; outer surface of all femora with tuberculate, distally-directed hairs. Upper surface of all femora with a row of erect spines about the size of tibial spines. Spines on hind tibiae separated by a distance greater than length of a spine.

Sterna covered with long, cream color decumbent hairs. Male clasper (Fig. 8) with large, pointed inner projection; inner point of projection recurved toward reduced shank; outer projection truncate, not prominently indented on outer (dorsal) surface; area of attachment flanged. Sperm reservoir (Fig. 15) with vesical seminal duct broad, loosely coiled, not encased by sleeve; holding sclerites long, curving, prominent and heavily sclerotized; wings arrow-shaped, prominent; arcuate extension forming a complete, wide bridge across bulb; area of insertion of vesical seminal duct into bulb rotated so that it appears opposite wings; corrugations absent.

**Holotype:** ♂, SURINAM, Zanderij, 31.VII-3.VIII-1964, DCG, (LEID).

**Paratypes:** BRAZIL, 1 ♂, Para, 3-26-IX-1962, W. L. Brown (AMNH); 1 ♂, Para, Jacareacanga, V-1969 F. R. Barbas (dissected and illustrated) (UCMS); 1 ♂, Amazonas, BR-174, KM 45, 2-IV-1982, E. L. Oliveira (INPA).

**Etymology.** From the Latin "immanis," an adjective meaning "immense" or "monstrous," in reference to the large protrusions on the venter of the head.

*Neopetissius perplexus* O'Donnell, **New Species**

Figs. 6, 13, 17

Total length 5.70. Maximum width, at claval commissure, 2.40. Dorsal surface glabrous, subshining. General coloration dusky brown, becoming burnt umber on posterior pronotal lobe, clavus and corium. Marked with cream color as follows: explanate pronotal margins and anterior half of corial margins, anterior pronotal margin, spot on posterior pronotal margin near junction of clavus and scutellum, elongate dash on clavus near apex of scutellum, 2 spots on corium between R + M and Cu, subapical inverted heart-shaped macula on corium, small irregular spots along veins of membrane, and small macula at apex of membrane. Antennal segment I burnt umber, segment II, proximal  $\frac{3}{5}$  of III and all of segment IV chestnut; distal  $\frac{2}{5}$  of segment III cream color. Venter uniformly dusky brown; femora except extreme distal ends burnt umber; labium, tibiae and tarsi cream color. Ventral surfaces of explanate pronotal and corial margins a strongly contrasting pale cream color.

Head moderately declivent, with numerous small, shallow punctures. Length head 0.80; preocular length 0.45; width 1.10; interocular 0.60. Tylus reaching  $\frac{1}{3}$  of way to apex of first antennal segment. First antennal segment gradually broadening distally, widest before distal end; segments II-IV terete, with upstanding hairs shorter than diameter of segment in addition to decumbent pubescence. Length antennal segments I 0.80; II 1.05; III 0.92; IV 1.07. Venter of head slightly raised and acinose on either side of midline. Labium reaching metacoxae, with first segment attaining base of head. Length labial segments I 0.85; II 0.72; III 0.90; IV 0.50.

Pronotum with anterior margin concave; collar area broad, delimited posteriorly by a groove of closely spaced punctures and otherwise punctate on posterior  $\frac{2}{3}$ ; lateral pronotal margins broadly explanate, sinuate; trichobothrium situated slightly anterior to level of collar at midline; posterior margin straight; transverse impression prominent, deepest near lateral margins and at midline where it meets longitudinal furrow. Pronotal calli sparsely and shallowly punctate; lateral margin of anterior lobe adjacent to explanate margin and entire posterior lobe coarsely and evenly punctate, with punctures becoming smaller toward posterior margin. Length pronotum 1.35; width across trichobothria 1.40; posterior width 2.20. Scutellum with depressed medial area and V-shaped elevation; with small shallow punctures anteriorly and deeper, coarser punctures mesally

and laterally. Length scutellum 1.22; width 1.25. Hemelytron macropterous, with explanate lateral margins as wide as those of anterior pronotal lobe. Clavus with 2 regular and 3 irregular rows of punctures; length claval commissure 0.70; corium with R strongly raised, corial fracture extending along R to level of claval apex. Midline distance apex clavus-apex corium 1.30; membrane extending slightly beyond end of abdomen and with two prominent cross-veins, one between Sc and R and one between R and M. Metathoracic scent gland peritreme curving gently posteriorly, removed from dorsal margin of evaporative area by less than its length; evaporative area rugose, extending dorsally as a narrow tongue closer to dorsal margin on mesopleuron than on metapleuron; prominent elongate lobe present opposite auricle; dorsal margin of evaporative area on metapleuron sloping sharply ventrad posteriorly. Fore femora moderately incrassate, each armed below with 4 stout spines distally, 3 longer, more slender spines mesally, and several elongate hairspines proximally (from paratype). All femora appearing "bumpy" but actually covered with silvery decumbent hairs set obliquely onto small tubercles. Tibial spines reduced in number and shorter than diameter of tibia.

Male clasper (Fig. 6) with a large, indented outer lobe, shank reduced; sperm reservoir (Fig. 13) with vesical seminal duct broad, strongly coiled, with a longitudinal split on distal half; sleeve prominent, extending to strong distal bend of sclerotized portion of vesical seminal duct; arcuate extension forming a complete, narrow bridge; wings quadrate, heavily sclerotized; corrugations apparent. Female genitalia with spermatheca (Fig. 17) mushroom-shaped, with bulb sitting directly on narrow distal flange; duct diameter about  $\frac{1}{3}$  bulb diameter; proximal flange strongly asymmetrical, flaring like a bell proximally; duct proximal to flange offset.

Holotype ♂, BRAZIL, Mato Grosso: Vila Vera, 55°30'long., 12°46'lat., IX-1973, M. Alvarenga (AMNH) (dissected).

Paratypes: BOLIVIA, 2 ♀♀, Santa Cruz, Prov. of San Esteban, Muyurina, 49 km N. of Santa Cruz. 1120 ft. elevation, 26-X-1959, R. B. Cumming, Blacklight Trap (1 dissected and illustrated) (RMB, UCMS); 1 ♀ same except 27-XII-1959 (RMB); 1 ♀, Santa Cruz, Saavedra, Dept. Santa Cruz Agr. Exp. Sta., R. B. Cumming, 27-XII-1959, Blacklight trap (RMB).

Additional Material Examined: BOLIVIA, 1♂, Santa Cruz, San Esteban Muyurina, 49 km N Santa Cruz, 1120 ft., 26-X-1959, R. B. Cumming, Blacklight Trap (dissected and illustrated) (UCMS).

**Etymology.** From the latin "perplexus" an adjective, in reference to the confusing, entangled nature of the species relationships in the genus.

The only male from Santa Cruz has prominently expanded antenniferous tubercles and ap-

parently represents an aberrant phenotype. This specimen otherwise appears normal.

This species appears to be closely related to *N. froeschneri* on the basis of the shared, unique configuration of the spermatheca, despite the fact that the sperm reservoirs are quite different. These two species are also very similar in external appearance. *N. froeschneri* has an overall reddish cast to the dorsum, and a narrower, less distinct light annulus on the third antennal segment.

*Neopetissius slaterorum* O'Donnell, **New Species**

Figs. 2, 5, 9, 12, 16

Dorsum mottled, subshining, glabrous. Total length 6.10. Nearly parallel-sided, maximum width, at level of apex of clavus, 2.30. Head dusky brown; anterior pronotal lobe, most of scutellum, and dark markings on corium, especially distal half, sepia; irregular markings on posterior pronotal lobe and apex of scutellum brick red; 2 elongate dashes along raised area of scutellum and light parts of posterior pronotal lobe buff yellow. Dorsum cream color as follows: spot on either side of midline on pronotal collar; lateral pronotal margins; postero-lateral corners of humeri; ground of clavus and corium; veins of membrane and obscure lighter areas basally, laterally, and apically. Distal third of antennal segment III and prominent subapical corial spot almost white. Remainder of antenna sepia. Venter shining, deep maroon on head, becoming gradually lighter posteriorly, with abdominal sternum VII amber. Labium cream color. Femora amber, lighter distally; tibiae and tarsi buff yellow. Explanate pronotal and corial margins cream color beneath.

Head impunctate, subshining except for prominently shining juga; eyes large. Length head 0.75; preocular length 0.40. Width head 1.15; interocular 0.68. Antennae terete; segment I gradually expanded distally, with stout hairs basally and another  $\frac{2}{3}$  of distance from base; all segments covered with decumbent pubescence; segments III and IV with additional scattered upstanding hairs shorter than diameter of segment. Length antennal segments I 0.80; II 1.02; III 1.00; IV 1.10. Venter of head not swollen, with a few inconspicuous hair-bearing punctures. Labium reaching metacoxae, with first segment reaching base of head. Length labial segments I 0.90; II 0.85; III 0.98; IV 0.50. Pronotum with anterior margin very shallowly concave; posterior margin shallowly concave across scutellum; lateral margins broadly explanate, slightly sinuate at level of transverse impression, set off by a row of punctures; trichobothria set at level of middle of collar; anterior lobe with distinct collar that broadens mesally, set off by a row of punctures that are closely-spaced laterally but more widely-spaced mesally; calli impunctate, raised; transverse impression well-developed, especially laterally and

mesally; posterior lobe except humeri and posterior margin evenly and coarsely punctate. Longitudinal furrow weakly developed on anterior lobe and at posterior margin, deeper and prominent at transverse impression. Length pronotum 1.35; posterior width 2.30; width across trichobothria 1.45. Scutellum elevated anteriorly, and with 2 raised, impunctate areas along lateral margins that join a weakly elevated, narrow, impunctate midline ridge at apex; otherwise finely and evenly punctate. Length scutellum 1.22; width 1.20. Hemelytron macropterous. Clavus with 2 straight, regular rows of punctures outlining edges, and 2 additional irregular rows. Corium with lateral margins explanate on proximal half; veins distinct, not strongly raised except for R; corial fracture mesal to R, extending to level of claval apex; Cu abruptly divergent from claval-corial suture at level of apex of scutellum. Membrane with cross veins present between Sc and R and R and M. Length claval commissure 0.70; midline distance apex clavus-apex corium 1.30. Fore femur slightly more swollen than either mid or hind femur; fore femur armed below with 4 short stout spines distally and strong hair-spines proximal to these; postero-ventral surface with a field of short hairs set on oblique tubercles; hind femur with 2 semidecumbent spines on upper surface and one longer spine on lower surface distally. Fore tibia with spines reduced but with short, erect, pale hairs present in addition to decumbent pubescence. Metathoracic scent gland (Fig. 2) with ostiolar peritreme not strongly elevated above evaporative area, curving evenly posteriorly to end in a blunt point; elongate lobe posterior to auricle narrow, ridge-like. Evaporative area covering all of mesoepimeron, extending broadly along meso-metapleural junction to dorsal margin of mesopleuron. Evaporative area covering ventral  $\frac{2}{3}$  of metapleuron, extending broadly dorsally but not to dorsal margin of metapleuron. Dorsal margin of evaporative area sinuate, broadly curved postero-dorsally.

Abdomen covered with long hairs ventrally. Male clasper (Fig. 9) with outer projection slightly indented, extending further along shank than inner projection; area of attachment with flange. Sperm reservoir (Fig. 12) with large bulb area, arcuate extension forming a complete, narrow bridge; wings and corrugations prominent; sleeve enclosing an asymmetrical, convoluted vesical seminal duct that is quite elaborate distad to sleeve, consisting of a flattened, thick curve and additional separate sclerites of uncertain homology that are unlike anything else in the tribe. Spermatheca (Fig. 16) with bulb sitting directly on broad distal flange; duct diameter about half the diameter of bulb; proximal flange with prominent perpendicular ring.

Holotype: ♂, PANAMA, Barro Colorado Island, 1-9-V-1964, WD and SS Duckworth. (NMNH).

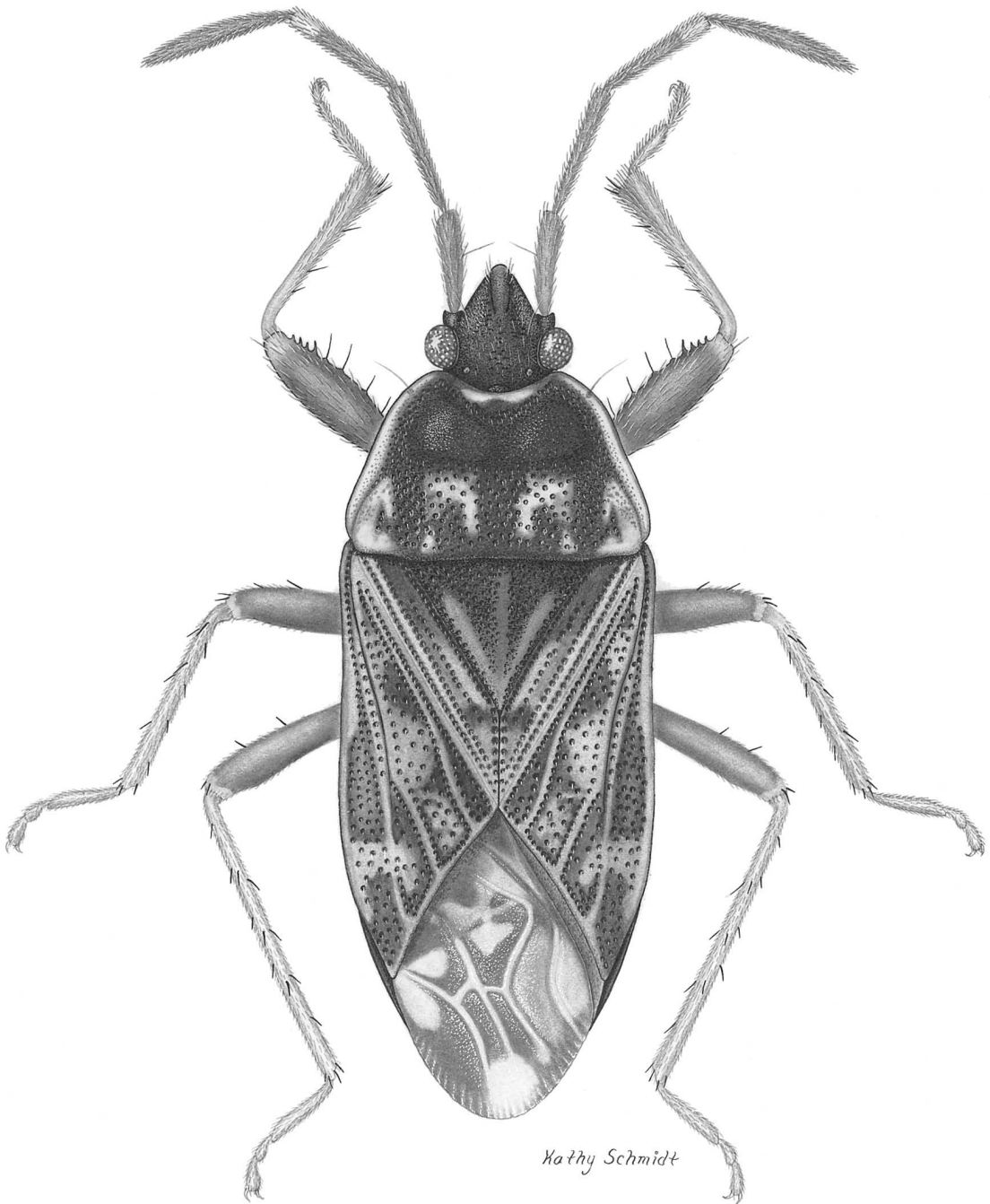


Fig. 5. *Neopetissius slaterorum*, n. sp., dorsal view. Scale line equals 0.10 mm.

Paratypes: 3 ♂♂, 6 ♀♀, same data as holotype (1 ♂ dissected and illustrated) (NMNH, UCMS); 1 ♀, same except 10-17-V-1964 (NMNH); 1 ♂, 1 ♀ same except 5-10-IV-1965 (♀ dissected and illustrated) (NMNH, UCMS); 1 ♀, same except 7-VIII-1967, C. W. & L. O'Brien, at light (JAS); 1 ♂, 1 ♀,

same except 18-28-IV-1964 (NMNH); 2 ♂♂, 2 ♀♀, same except 28-30-IV-1964 (NMNH); 1 ♂, 1 ♀, same except 14-III-1956, Carl W. and Marian E. Rettenmeyer (SNOW); 1 ♀, same except 18-V-1973, D. Engleman (JAS); 1 ♀, same except 9-I-1929, C. H. Curran (AMNH); 1 ♂, same except

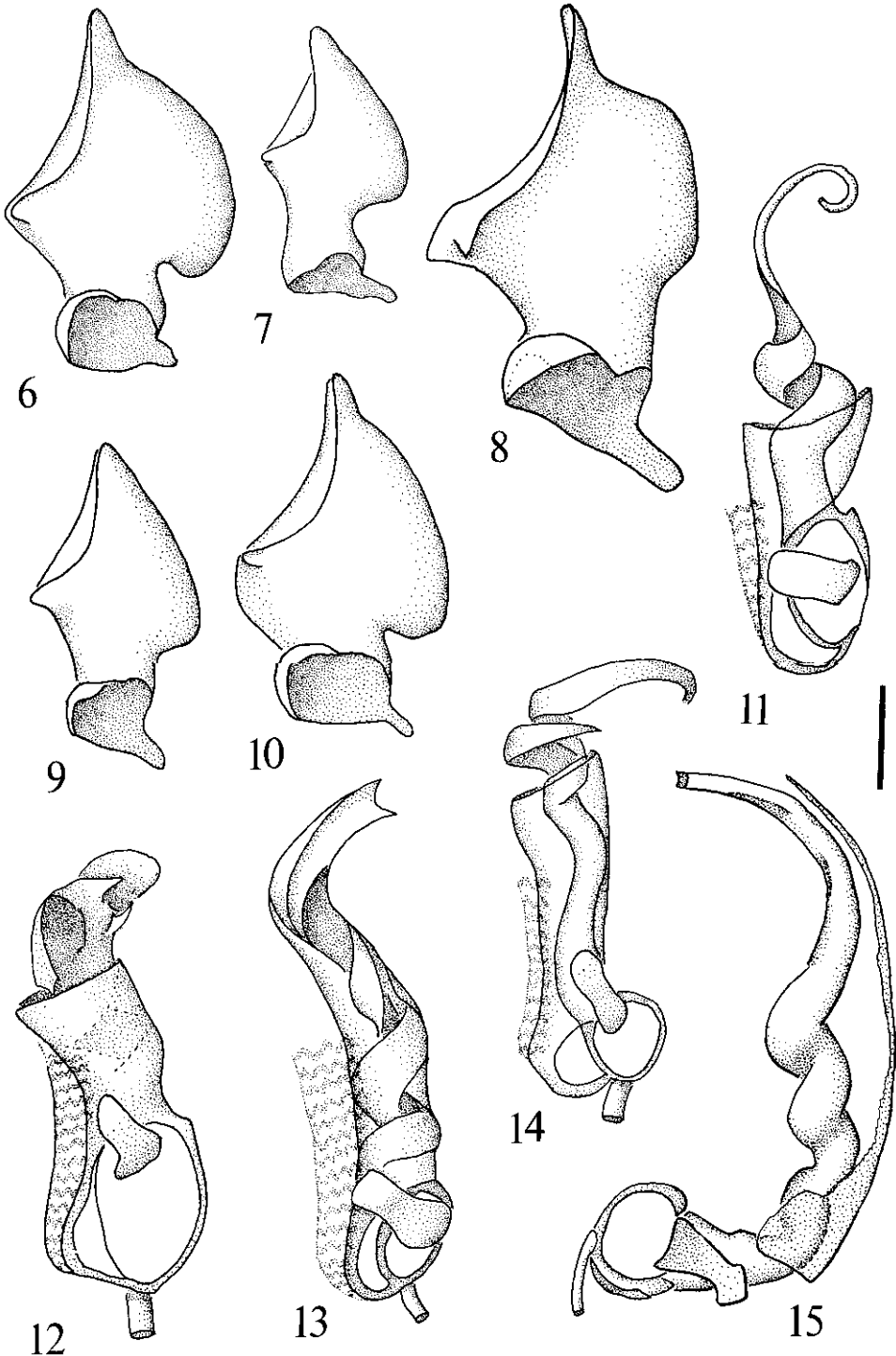


Fig. 6. *Neopetissius perplexus*, n. sp., ventral view left clasper. Fig. 7. *Neopetissius variegatus*, n. sp., ventral view left clasper. Fig. 8. *Neopetissius immanis*, n. sp., ventral view left clasper. Fig. 9. *Neopetissius slaterorum*, n. sp., ventral view left clasper. Fig. 10. *Neopetissius froeschneri*, n. sp., ventral view left clasper. Fig. 11. *Neopetissius froeschneri*, n. sp., lateral view, sperm reservoir. Fig. 12. *Neopetissius slaterorum*, n. sp., lateral view, sperm reservoir. Fig. 13. *Neopetissius perplexus*, n. sp., lateral view, sperm reservoir. Fig. 14. *Neopetissius variegatus*, n. sp., lateral view, sperm reservoir. Fig. 15. *Neopetissius immanis*, n. sp., lateral view, sperm reservoir. Scale line equals 0.10 mm for all drawings except Fig. 15, where it equals 0.13 mm.

Griswald (no date) (AMNH); 2 ♂♂, same except VIII-IX-1949, Zetek, Berlese funnel (NMNH); 1 ♀, same except IV-1945 (NMNH); 4 ♂♂, same except XII-1946-II-1947 (NMNH); 2 ♂♂, same except I-III-1944, Zetek (NMNH); 1 ♂, same except IX-X-1940, at light (NMNH); 1 ♂, same except 24-V-1940, at light (NMNH); 1 ♂, 1 ♀, same except V-1941 (NMNH); 1 ♂, 1 ♀, same except VII-VIII-1942 (NMNH); 1 ♀, same except X-XI-1941 (NMNH); 1 ♂, same except 14-IV-1937, S. W. Frost (NMNH); 1 ♀, same except 21-VI-1924, W. M. Wheeler (AMNH); 1 ♂, same except 24-VII-1924, N. Banks (AMNH); 1 ♂, same except 30-VIII-1974, H. Hespeneheide (JAS); 1 ♂, same except 5-XI-1973, H. Wolda, light trap (JAS); 1 ♂, same except Snyder Molinos, XI-1973, H. Wolda (JAS); CANAL ZONE: 1 ♂, La Campana, II, III-1938, Jas Zetek, Fruit Fly Trap; 1 ♂, Coco Solo Hospital, 8-VI-1973, D. Engleman, light trap (JAS); 1 ♀, same except 15-VI-1973 (JAS); 1 ♂, same except 23-V-1972, 9°21'N, 79°37'W (JAS); 1 ♂, Fort Kobbe, 1-VII-1976, E. G. Riley (UMC); 1 ♂, same except 9-VI-1985, E. Riley, D. Rider (RIDER); 1 ♂, Gatun Spillway, 24-IV-1974, D. Engleman (JAS); 1 ♀, same except 16-I-1974, Slater & Harrington (JAS); 4 ♀♀ (one illustrated in dorsal view), Madden Forest Reserve, 9-I-1974, J. A. Slater, J. Harrington, adults in *Ficus* sp. litter (JAS); 1 ♂, 1 ♀, same except 17-I-1974 (JAS); 1 ♀, same except mi 2.5, 4-V-1973, 9°05'N, 79°37'W, H. Stockwell (JAS); MEXICO: 1 ♂, 2 ♀♀, Quintana Roo, 20 km N Felipe Carrillo Puerto, 12-14-VI-1983, E. Riley (RIDER); 1 ♀, Yucatan, Chichen Itza, 10-11-VI-1983, E. Riley (RIDER).

Additional Material Examined: TRINIDAD: 7 ♂♂, 4 ♀♀, Caura Valley Recreational Site, 5.6 mi post, 24-VIII-1982, J. A. & E. Slater, R. Clayton, M. Hassey (JAS); 2 ♂♂, Simla Biological Station (no date) M. Emsley, at light (JAS); 1 ♂, 1 ♀, Diego Martin, 3-IX-1941, E. McG. Callan, in cave (NMNH); 1 ♂, 1 ♀, St. George Co., Aripo Valley, Rapsey, 1-8-VIII-1978, R. M. Baranowski, malaise trap (RMB); 1 ♀, same except 4-11-X-1978 (JAS); 1 ♀, same except no date (JAS); 1 ♂, same except 7-14-VIII-1978 (JAS); 1 ♂, same except 27-IX-4-X-1978 (JAS); 1 ♂, Simla, Arima-Blanchisseuse Rd., 22-VII-1975, J. Price, blacklight trap, elev. 600 ft (RMB); 1 ♀, same except 21-IX-1983, R. M. Baranowski (JAS); 1 ♀, 14-VII-1902, Chipman (CAS); 1 ♀, Waller Field, 5 mi E Arima, 14-VI-1973, R. Baranowski, F. O'Rourke, V. Picchi, J. Slater (JAS); 1 ♂, Toco Rd., 20.74 mi post, 13-VIII-1975, R. M. Baranowski (JAS); 1 ♂, St. George Co., Curepe, Santa Margarita Circular Road, 10-VII-1978, F. D. Bennett (JAS); 1 ♀, same except 29-X-1975 (JAS); 2 ♂♂, 1 ♀, Simla, Arima Valley, 24-VIII-1978, M. Ramla, blacklight trap (JAS); 1 ♂, same except 20-IX-1978 (JAS); 1 ♂, same except 18-VIII-1978 (JAS); 2 ♀♀, Maracas Valley, 1 mi N St. Joseph, 9-IV-1979, L. Du Bruijn, blacklight trap (JAS); BRAZIL: Bahia: 23 ♂♂, 21

♀♀, Encruzilhada, XI-1972, M. Alvarenga, 960 m (AMNH); 1 ♂, 1 ♀, same except 900 m (AMNH); 1 ♂, same except XI-1974, 960 m (AMNH); 1 ♀, Para, Jacareacanga, XII-1968, M. Alvarenga, at light (AMNH); 2 ♂♂, Pernambuco, Caruaru, V-1972, J. Lima, 900 m (AMNH); 1 ♂, Mogajuba, Mangabiera, IV-1953, Orlando Rego (RIO) 1 ♂, Amazonas, V-8, 19-V-1982, J. A. Rafael, malaise (INPA); 1 ♀, Santarem (F.A.O.), Diamantina 15-XII-1963, G. Marlier (IRSN); BRITISH HONDURAS (BELIZE): 1 ♂, Punta Gorda, 1931 (NMNH); 1 ♂, same except II-1932 (NMNH); 1 ♀, same except III-1931, J. J. White (NMNH); 1 ♂, San Antonio, VI-1931, J. J. White (NMNH); COLOMBIA: 1 ♂, Guajira, Manaure, 19-20-IX-1968, B. Malkin (AMNH); ECUADOR: 1 ♂, Pichincha, Rio Palenque, 29-IV-5-V-1987, B. Brown, L. Coote, malaise trap, rainforest, 120-160 m (UCMS); EL SALVADOR: 1 ♀, Rosario, 23-III-1955, M.S.V. (NMNH); GUATEMALA: 1 ♂, Altav. Paz., Cacao Trece Aguas, (no date), Barber & Schwartz (NMNH); 1 ♂, Peten Tikal, 8-IV-1956, Hubbell-Cantrell (UMAA); HONDURAS: 1 ♂, Guimas, 4-V-1923, T.H. Hubble (NMNH); MEXICO: Veracruz: 2 ♂♂, 5 ♀♀, 4 mi NW Sonte Comapan, 9-VI-1965, Burke, Meyer, Schaffner, at light (SWEET); 2 ♀♀, Rio Quezalapan, 2 mi E. Lago Catemaco, 12-VII-8-VIII-1964, J. R. Meyer, (SWEET); 1 ♂, near Montepio, UNAM Field Station los Tuxtlas, 10-16-VI-1981, W. R. Dolling, B. M. 1981-411, tropical rainforest, general collecting (BMNH); 1 ♂, Catemaco, 20-VII-1980, Schaffner, Weaver, Friedlander, at light (TAMU); 1 ♂, 1 ♀, 38 mi S. Acayucan, 17°57', 94°54', 2-III-1976 (no collector) (AMNH); 1 ♀, Lake Catemaco, 24-XI-1962, C. & P. Vaurie (AMNH); 1 ♂, Catemaco, 20-V-1964, J. C. & D. Pallister (AMNH); 1 ♀, same except 30-V-1964 (AMNH); 2 ♀♀, same except 31-V-1964 (AMNH); Yucatan: 2 ♀♀, Colonia Yucatan, 12-VII-1952, J. & D. Pallister (AMNH); 1 ♀, Chuminopolis, 6-VIII-1964, J. C. & D. Pallister (AMNH); 1 ♂, Chichen Itza, 24-V-1956, T. H. Hubble, at light (UMAA); Oaxaca: 1 ♂, Tehuantepec, 11-VI-1964, J. C. & D. Pallister (AMNH); Tamaulipas: 1 ♂, Boca Toma, 7 km SSE Gomez Farias, 5-7-I-1981, E. G. Riley (UMC); 1 ♂, Sotano de Gomez Farias, 1-VI-1964, J. Reddell et al. (NMNH); Campeche: 1 ♀, Escarcega, 3-VI-1962, F. Islas S., light trap (NMNH); 1 ♂, same except 5-VI-1962 (NMNH); 1 ♂, same except 20-VI-1962 (NMNH); 2 ♀♀, same except 22-VI-1962 (NMNH); 1 ♂, 1 ♀, same except 24-VI-1962; 3 ♀♀, Escarcega, Forestry Research Station El Tormento, 17-21-VI-1981, W. R. Dolling, tropical rainforest at light (BMNH); SURINAM: 1 ♂, 1 ♀, P. H. v. Doesburg, Jr. (LEID); 1 ♀, Republiek, 10-V-1963, P. H. v. Doesburg, Jr. (LEID); 1 ♀, Brokopondo, 10-XII-1965, G. F. Men. (LEID); VENEZUELA: Aragua, El Limon, 14-V-1970, A. Namirez, 450 m (VENZ); 3 ♀♀, same except 17-18-II-1973, C. J. Rosales, 480 m, en trampa malaise (VENZ);

1 ♂, same except 22-II-1973 (VENZ); 1 ♀, same except 21-III-1973 (VENZ); 1 ♀, same except 28-III-1973 (VENZ); 1 ♀, same except 26-V-1976 (VENZ); 1 ♀, same except 20-VI-1973, 450 m (VENZ); 4 ♀♀, same except 25-VI-1973 (VENZ); 1 ♂, 1 ♀, same except 24-VI-1974 (VENZ); 1 ♂, same except 4-VII-1983, F. Fernandez Y., 450 m, lua negea (VENZ); 1 ♀, same except 30-V-1976, 450 m, luz de mercurio (VENZ); 1 ♂, same except 23-VII-1976 (VENZ); 1 ♂, same except 6-V-1977 (VENZ); 1 ♀, same except 16-V-1977 (VENZ); 1 ♀, same except 5-IV-1978 (VENZ); 1 ♀, same except 5-IV-1978 (VENZ); 1 ♀, same except 26-IV-1978 (VENZ); 1 ♂, La Isleta Choroni, 14-15-VII-1975, J. Salcedo, F. Fernandez, 200 m, en la luz (VENZ); 1 ♂, 1 ♀, Trujillo, Agua Viva, 9-VII-1977, E. Osuna (VENZ); 1 ♂, 1 ♀ Falcon, Las Dos Bocas, 7-VI-1969, R. Casares, J. B. Teran, M. Gelbez, 200 a 500 m (VENZ); 1 ♀, Falcon, Bocade Aroa, 1-3-IX-1976, C. Michelangelli, J. A. Clavijo, Luz de Mercurio (VENZ); 1 ♂, Monages, Uverito, 22-VI-1978, C. J. Rosales, en trampa malaise (VENZ); 1 ♀, same except 17-X-1979, en luz de neon (VENZ); 1 ♂, same except 19-VI-1978, trampa neon (VENZ); 1 ♀, same except 16-X-1978, C. J. Rosales and J. A. Gonzalez (VENZ); 1 ♂, same except 25-I-1979 (VENZ); 1 ♂, Monagas, Jusepianagas, 17-XI-1967, E. Osuna, A. Osutia (VENZ); 1 ♂, same except 10-IX-1965, F. Fernandez, C. J. Rosales (VENZ); 1 ♀, Cojedes, Galeras del Pao, 27-VII-1967, C. J. Rosales, R. Poole (VENZ); 1 ♀, Anzoategui, Clarines, 6 km N, 25-VIII-1975 (VENZ); 1 ♀, Aragua, Cagua, 27-XI-1957, E. Dosonte, 450 m (VENZ); 1 ♀, Zulia, Rio Ariguia, 20-VIII-1979, E. Osuna (VENZ); 1 ♂, Zulia, Kasmerario, Yaaa Sierra de Perija, 22-IX-1961, C. J. Rosales, F. Fernandez, 250 m (VENZ); 1 ♀, Barinas, Calderas, 8-V-1972, J. & B. Bechyne, 1000 m (VENZ); Apure, Hato El Frio, Fundo Ceibote, 20-V-1975, C. J. Rosales, 100 m (VENZ); 1 ♂, Bolivar, Jabilla, Rio Cavra, 25-XI-1978, A. Chacon, 100 m (VENZ); 1 ♂, Bolivar, Rio Guaniamo, 25-28-V-1979, J. Clavillo, A. Chacon, G. Yopez, 160 m, N6°45', O66°01' (VENZ); 1 ♀, Bolivar, Guri, 16-XI-1966, J. & B. Bechyne, E. Osuna (VENZ); 1 ♀, Guasipati, 20-V-1975, B. Bechyne (VENZ); 1 ♂, Bolivar, Macagua, Gran Sabana, 17-XI-1966, J. Bechyne, E. Osuna, (VENZ); 1 ♀, Dto. Federal, Chichiriviche, Colonia, Tovar, 28-I-1977, C. J. Rosales, L. J. Joly, 10 km carret. (VENZ); 1 ♂, T. F. Amazonas, Pto. Ayacucho, 22-IV-1967, J. Anduce (VENZ);

Etymology: Named for James A. and Elizabeth A. Slater, in recognition of their many years together.

*Neopetissius variegatus*, O'Donnell, **New Species**

Dorsum variegated, covered with short, up-standing, silvery hairs, dull except for subshining head. Total length 5.40. Nearly parallel-sided; maximum width, just anterior to level of apex of

clavus, 2.00. Head and anterior pronotal lobe except for collar and lateral margins dark grayish brown; posterior pronotal lobe, clavus and corium mostly chestnut, marked with buff yellow as follows: anterior pronotal collar on either side of middle; kidney-shaped spot near middle of posterior pronotal lobe and 2 equally-sized spots along posterior margin, one at humerus and one nearer meson; narrow, elongate, curving spot on clavus between anterior rows of punctures; linear spot between two posterior rows, connecting with previous spot mesally, and a small triangular patch along claval commissure; lateral margin of corium for  $\frac{2}{5}$  its length. 2 irregularly shaped spots between R + M and Cu in basal half of corium, and large, inverted-heart shaped spot subapically on corium; and 2 ovoid dashes laterally on scutellum. Membrane translucent raw umber, with veins, small indistinct spots between veins, and faint macula at apex cream color. First, second, basal  $\frac{2}{3}$  of third, and fourth antennal segments chestnut. Distal  $\frac{1}{3}$  of third antennal segment strongly contrasting cream color. Venter shiny maroon, becoming chestnut on coxal cavities, at posterior edges of thoracic segments, and distally on abdomen. Femora chestnut; tibiae buff yellow, suffused with chestnut on fore tibia and distally on mid and hind tibiae. Labium chestnut, becoming paler distally.

Head slightly declivent; tylus reaching middle of first antennal segment. Venter of head rugose, only slightly swollen on either side of midline. Length head 0.80; preocular length 0.50; width head 0.92; interocular 0.52. Antennae with first segment thickest, with inward curve and with a stout hair  $\frac{1}{3}$  of way along inner margin. Length antennal segments I 0.70; II 1.00; III 0.78; IV 0.90. Labium extending onto 3rd abdominal sternum, first segment slightly surpassing base of head. Length labial segments I 0.92; II 0.92; III 0.92; IV 0.48.

Anterior pronotal margin concave, with a well-defined collar set off by indistinct groove of small punctures. Posterior margin straight; lateral margin only slightly sinuate at junction of anterior and posterior lobes; explanate lateral margins narrower than width of collar at midline. Trichobothrium level with anterior-most punctures of collar groove at midline. Anterior lobe impunctate, slightly swollen, calli confluent; transverse impression deepest at sides, prominent even across middle; posterior lobe evenly and sparsely punctate. Width across trichobothria 1.22; posterior width pronotum 1.75; length 1.08. Scutellum with small shallow punctures in slightly depressed mesal area, a few larger punctures midway along lateral margin. Length scutellum 0.90; width scutellum 0.92. Clavus with 3 regular and 2 irregular rows of punctures. Length claval commissure 0.60. Corium with lateral margins explanate on anterior half, about as



wide as widest part of lateral pronotal margins, veins not strongly elevated; membrane with two cross-veins, one between Sc and R and one between R and M. Midline distance apex clavus-apex corium 1.05. Length apex corium-apex membrane 0.85. Scent gland peritreme elongate, narrow, gradually sloping posteriorly, not strongly elevated above evaporative area; evaporative area rugose, covering all of mesoepimeron and extending as a narrow tongue nearly to its dorsal margin, and covering more than half of metapleuron, extending as a wider tongue towards dorsal margin. Dorsal margin of metapleuron with a series of parallel vertical ridges. Fore femur moderately incrassate, armed below with 2 ranks of spines, an inner rank with several elongate hair-spines proximally, one longer, thicker tuberculate spine distad of these, and one large and three small, strong, stout, tuberculate spines; outer row, on postero-ventral surface of fore femur, with 5-6 hairs set on oblique, distally-directed tubercles; mid and hind femora only slightly swollen, with similar but less prominent rows of obliquely tuberculate spines. Hind femur with a spine near distal end ventrally.

Abdomen with venter shiny, sparsely clothed with long silvery hairs; sternum 4 with more numerous shorter appressed hairs. Male clasper (Fig. 7) stout, inner projection further from area of attachment than outer projection; outer surface of outer projection compressed. Sperm reservoir (Fig. 14) with a prominent, distally coiled sleeve; arcuate extension complete, broad in dorsal view; vesical seminal duct smoothly curving inside sleeve until spiral starts, then flattening and forming a thickened ridge at sharp distal bend; wings elongate, prominent, directed distally; holding sclerites absent; corrugations distinct. Spermatheca (Fig. 19) mushroom-shaped, with bulb sitting directly on very narrow distal flange; duct about  $\frac{1}{3}$  diameter of bulb, but nearly doubling in width proximally; proximal flange very lightly sclerotized except for heavy point on one side.

Holotype: ♂, BAHAMAS, Mayaguana Isl. 24-VIII-1963, C. Murvosh, Blacklight trap (AMNH).

Paratypes: 1 ♂, same data as holotype except 27-VIII-1963; 3 ♂♂, 2 ♀♀, same data as holotype except 28-VIII-1963; 1 ♀, same data as holotype except 26-VIII-1963; 2 ♀♀, same data as holotype except 3-VIII-1963 (JAS, RMB, UCMS).

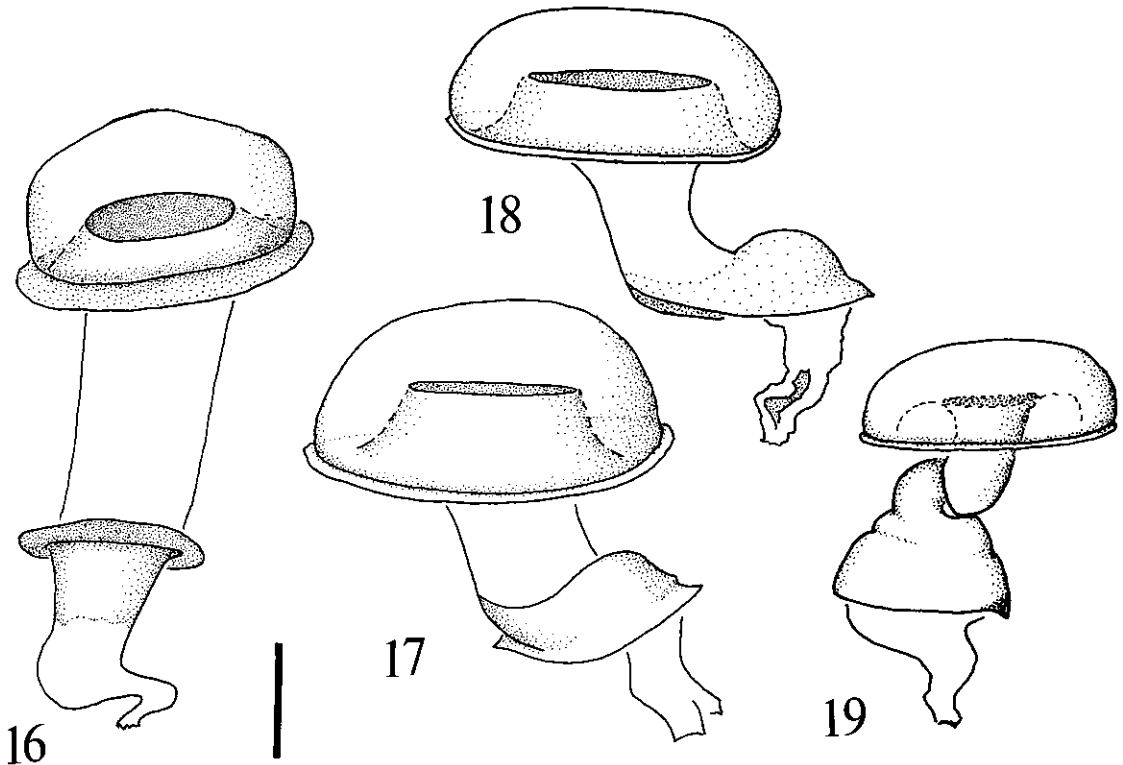


Fig. 16. *Neopetissius slaterorum*, n. sp., spermatheca. Fig. 17. *Neopetissius perplexus*, n. sp., spermatheca. Fig. 18. *Neopetissius froeschneri*, n. sp., spermatheca. Fig. 19. *Neopetissius variegatus*, n. sp., spermatheca. Scale line equals 0.10 mm.

Additional Material Examined: BAHAMAS: 2 ♂♂, 4 ♀♀, Eleuthera, Rainbow Bay, 1-9-VI-1984, R. & D. Wiley, blacklight trap (RMB); 1 ♀, Andros Is. Nicholls Town, 28-VI-1994, R. M. & H. V. Baranowski, blacklight trap (RMB); CUBA: Coast below Pico Turqueno, 26-30-VI-1936, Darlington (AMNH); DOMINICAN REPUBLIC: 1 ♂, 1 ♀, Dajabon, 9 km S Loma de Cabrera, 19-21N, 71-37W, 12-VII-1992, J. Rawlins, S. Thompson, C. Young, R. Davidson, 620 m, disturbed pastures in mesic woodland (CARN); La Altagracia: 1 ♀, Nisibon (Papagallo), 23-VI-1998, R. M. Baranowski and R. E. Woodruff, blacklight trap (RMB); 1 ♀, same except 16-19-VI-1998, R. E. Woodruff and P. H. Freytag (RMB); 1 ♀, 5 km W Nisibon, 17-VI-1998, R. E. Woodruff and P. H. Freytag (RMB); 2 ♀♀, La Romana, 3 km Casa de Campo, 15-VI-1998, R. E. Woodruff and P. H. Freytag (RMB); 1 ♂, El Seibo, Loma de Chivo, 7 km N Pedro Sanchez, 20-VI-1998, R. E. Woodruff and P. H. Freytag, 5000 ft., blacklight trap (RMB); 1 ♂, Monsenor Noel, nr. Bona, Jacaranda Hotel, 1-VII-1999, R. E. Woodruff and R. M. Baranowski (RMB); Pedernales: 1 ♂, 24.5 km N Cabo Rojo, 5-VII-1998, R. M. Baranowski and R. E. Woodruff, 3200 ft., blacklight trap (RMB); 1 ♂, 23.5 km N Cabo Rojo, 18-06N, 71-39W, 13-19-VII-1990, L. Masner, J. Rawlins, C. Young, 540m deciduous forest, intercept trap (CARN); 1 ♀, 9.5 km N Cabo Rojo, 18-02N, 71-39W, 19-VII-1990, J. Rawlins, C.W. Young, S. A. Thompson, 35 m (CARN); 1 ♀, 13 km N Pedernales, Along Rio Mulito, 18-09N, 71-46W, 17-VII-1992, J. Rawlins, S. Thompson, C. Young, R. Davidson, 230 m, riparian woodland (CARN); 1 ♀, Hato Mayor, Parque Los Haitises, 3 km W Cueva de Arena, 18-04N, 69-29W, 7-9-VII-1992, R. Davidson, J. Rawlins, S. Thompson, C. Young, 20m (CARN); 1 ♂, Monte Cristi, 5 km NNE Botoncillo, 19-06N, 71-24W, 29-30-XI-1992, R. Davidson, M. Klinger, S. Thompson, J. Rawlins, 50 m, arid thornscrub (CARN); 1 ♂, La Toma, N of San Cristoban, 9-10-VI-1969, Flint & Gomez (NMNH); 1 ♂, Puerto Plata, 23-VIII-1967, L. H. Rolston (SWEET); 1 ♂, Santo Domingo, 12-VIII-1967, J. C. Schaffner, at black light (SWEET); 2 ♂♂ (one dissected and illustrated), 2 ♀♀ (one dissected and illustrated), 1 immature, Duarte, 6 km N Castillo. R. D. Schuster, 8-VIII-1978 (JAS, UCD, UCMS); HAITI: 1 ♀, Etang Lachaux, SW

Peninsula, 26-27-XI-1934, Darlington, under 1000 ft (AMNH); JAMAICA: 1 ♂, Portland, near Millbank, along Rio Grande River, 18-V-1969, R. E. Woodruff, blacklight trap (RMB); 1 ♀, St. Andrew, Vi-1973, C. Griffith (JAS); PUERTO RICO: Rio Abajo, Forest Rd. #621, K.S.2., 1000 ft., 18°18' N, 66°04' W (NMNH); VIRGIN ISLANDS: St. Croix: 1 ♂, Hams Bluff, 27-I-1979, M. A. & L. L. Ivie (UCMS); St. John: 3 ♀♀, Estate Carolina, NW Coral Bay, 18-V-1984, W. B. Muchmore, 250 ft., litter (RMB); 1 ♀, Estate Adrain ruins, 25-II-1984, W. B. Muchmore, along walls (RMB); 1 ♂, top Bordeaux Mt., 15-V-1984, W. B. Muchmore, liter (RMB); TURKS AND CAICOS: 1 ♀, M. Caicos, Bambarra, 4-XII-1993, B. M. Riggs, blacklight trap (RMB); 1 ♂, same except 12-XII-1993 (RMB); 1 ♂, North Caicos Is., Pelican Beach Hotel, 31-V-1991, H. V. & R. M. Baranowski, blacklight trap (RMB).

Etymology: From the Latin, "variegat-", marked variously, an adjective in reference to the variegated dorsal coloration.

Females lack the prominent tuberculate hairs on the fore femur, and have longer beaks. Some specimens have three cross-veins in the membrane.

#### ACKNOWLEDGMENTS

I wish to thank the curators of the institutions and the owners of the personal collections listed in the Materials and Methods section for the loan of material and for their extreme patience over the extended period that I have had their specimens on loan. I also thank the illustrators, Kathy Schmidt and Steven Thurston, both formerly of the University of Connecticut, for the drawings that so superbly capture the essence of the insects. The University of Connecticut Research Foundation supported field work in Ecuador in 1987 and Costa Rica in 1988.

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## EFFECT OF ENZONE™ AS A SOIL FUMIGANT ON SURVIVAL OF VARIOUS DEVELOPMENTAL STAGES OF *DIAPREPES ABBREVIATUS* (COLEOPTERA: CURCULIONIDAE) IN CONTAINER-GROWN CITRUS

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*Diaprepes abbreviatus* L., a root weevil native to the Caribbean region (Woodruff 1985), has become an important pest of citrus and ornamental plants in Florida since its introduction over 30 years ago (McCoy 1999). Both the adult and larval stages of *D. abbreviatus* are polyphagous, feeding on the leaves and roots of about 270 plant species (Simpson et al. 1996). Plants that support larval development to pupation, a period of 5-15 months, include citrus, sugarcane, various woody ornamentals, and several agronomic crops (Schroeder et al. 1979). Injury to citrus by the adult is characterized by a notching of the leaf margin while larval feeding destroys fibrous roots and the bark of the tap, lateral, and crown roots (Quintela et al. 1998). *D. abbreviatus* is a univoltine species with the adult, egg, and neonate stages appearing on the host plant aboveground, whereas all larval stages, pupae, and teneral adults reside belowground (Wolcott 1933).

The potential economic impact of *D. abbreviatus* on commercial growers of citrus and ornamental plants in nurseries and the field is significant. An estimated loss of \$75 million annually has been reported from tree decline and lost production in open forum among citrus growers (Diaprepes Task Force 1997). About 100 commercial plant nurseries are infested throughout Florida. Sale of infested trees from nurseries offer one of many ways by which the weevil can be disseminated throughout the state. It is imperative that these nurseries do not sell liners infested with weevil larvae. All weevil-infested nurseries operate under a compliance agreement with the Florida Department of Agriculture and Consumer Services that regulates the movement of nursery stock (McCoy 1999). Larval control in infested nurseries must be performed using an approved chemical or mechanical treatment of potting media in containers. Currently, bifenthrin, formulated as Talstar® 10 WP and Talstar T & O granular are the only approved regulatory chemical treatments (McCoy et al. 1995). Talstar® is a soil barrier treatment applied to prevent neonate invasion of the soil. It is, however, ineffective against later instars already infesting the plant roots.

To find a soil treatment effective against all developmental stages of *D. abbreviatus*, two greenhouse studies were initiated to determine the

effect of sodium tetrathiocarbonate (Enzone™ 31.8%) as a soil fumigant on all developmental stages of *D. abbreviatus*. The active ingredient decomposes in the soil environment to release carbon disulphide, a broad spectrum biocide of plant parasitic nematodes, grape phylloxera and some soil fungi (Hinds 1902, Young 1990, Weber et al. 1996). The formulation exhibits very low phytotoxicity and is environmentally benign.

In both tests, 3-year-old Marsh grapefruit (*Citrus × paradisi* Macfad) trees grafted to Swingle Citrumelo rootstocks (*Citron cirus* 'Swingle') were bare rooted and pruned lightly for transplanting into 15 liter plastic containers. Each tree was planted in sieved Candler soil (Entisol type; 92% sand, 2.9% clay, 2.0% silt) at a maximum soil depth of 26.7 cm (soil volume/pot = 0.0179 m<sup>3</sup>, soil surface area = 670 cm<sup>2</sup>). Trees were placed on a bench in an air-conditioned greenhouse maintained at 25.5-26.5°C, where they received regular watering and 60 ml of liquid fertilizer (8:4:8) per tree every 2 weeks. Any weeds growing on the soil surface were removed by hand. In test 1, 100 neonate *D. abbreviatus* (48 hours old) were scattered on the soil surface next to the trunk of each containerized tree each week for 6 consecutive weeks prior to treatment. In addition, five, 6<sup>th</sup> instars, were placed in each pot on four different occasions beginning at the 6<sup>th</sup> week after neonate inoculations were begun. In test 2, 100 neonates of the above age were added to the containerized trees twice at 5 and 6 weeks prior to treatment. Ten, 7<sup>th</sup> instar larvae, 3 pupae and 3 teneral adults were buried in the soil at a depth of about 7.6 cm of each pot 1 week prior to treatment.

Prior to treatment, infested and non-infested containerized trees were randomized according to treatment and replicate and removed from the greenhouse to a shaded out-of-doors site for treatment. After 20 hours, they were returned to the greenhouse. In test 1, Enzone™ was applied as a drench at rates of 500 (4.6 ml in 3.78 liters H<sub>2</sub>O) and 1000 ppm. Initially, all containers were watered using a sprinkling can to achieve soil saturation (3.78 liters/unit).

The entomopathogenic nematodes, *Heterorhabditis bacteriophora* Poinar and *H. indicus* Poinar, Karunaker, & David supplied on a sponge (Integrated BioControl Systems, Aurora, IN), were

applied in equal number to each container as a standard at rates of 22 and 54 infective juveniles (IJ's)/cm<sup>2</sup> or 14,740 and 36,180 IJ's/unit. For application, the required number of IJ's were pipetted into one liter of water and sprinkled on the soil with a watering can. Prior to inoculation, nematode viability was estimated microscopically by counting the number of motile and dead IJ's in 10 fields of view at 60× mag. Both species had viabilities of 82%. Treatments including an infested and non-infested control were replicated 6 times. In test 2, Enzone™ was applied in the manner described for test 1, except five rates were tested ranging from 100 to 2000 ppm and nematodes were not applied. Treatments included an infested and non-infested control and were replicated 5 times.

From 3 to 5 days post-treatment, each tree from the infested control and Enzone treatments was removed from its container and the soil carefully washed from the roots. Soil from the container and from the roots was wet sieved (2.0 mm mesh) to recover surviving and dead larvae detectable to the naked eye. Dead larvae were diagnosed as having no movement and color change.

In test 1, trees treated with nematodes and the uninfested control were processed at 14 days post-treatment in the manner described above. Many dead larvae exhibited red color typical of nematode infection or partial cadaver disintegration upon diagnosis. All live and dead larvae recovered from the soil in the Enzone treatments and the infested control had head capsule measurements performed microscopically to determine approximate instar at the termination of the test. In both tests, fibrous roots were examined for symptoms of Enzone™ phytotoxicity. None was observed.

In test 1, 203 live and dead larvae recovered from soil of the infested control had completed numerous molts according to head capsule measurements; 8.2% were categorized as 4<sup>th</sup> instar, 40.4% as 5<sup>th</sup> instar, 36.9% as 6<sup>th</sup> instar and 14.5% as 7<sup>th</sup> instar. These findings suggest that larval development from neonate to 7<sup>th</sup> instar occurred within 60 days, which agree with Quintela et al. (1998). By comparison to the uninfested control, larval injury to infested trees was uniform, with virtually no fibrous root survival and excessive bark loss (Fig. 1). Obviously, root injury occurred before Enzone™ treatment were applied to containers.

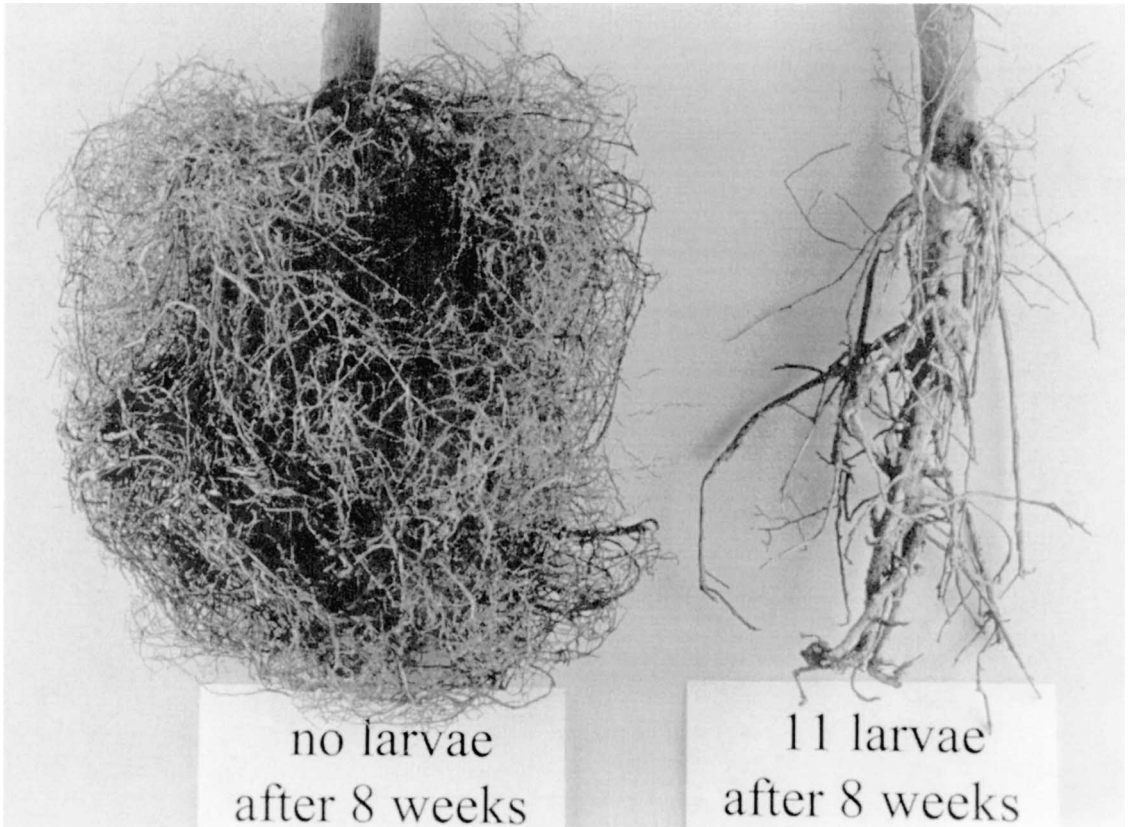


Fig. 1. A comparison of Swingle citrumelo root masses with and without larval feeding of *Diaprepes abbreviatus* after 8 weeks exposure.

TABLE 1. EFFECT OF TWO RATES OF ENZONE™ COMPARED TO ENTOMOPATHOGENIC NEMATODES ON THE SURVIVAL OF LARVAL INSTARS OF *DIAPREPES ABBREVIATUS* INFESTING CONTAINERIZED CITRUS TREES IN THE GREENHOUSE.

| Treatment                    | Rate               | No. live and dead larvae recovered | Mean % larval mortality ± SE <sup>b</sup> |
|------------------------------|--------------------|------------------------------------|---|
| Enzone™ (31.8%)              | 500 ppm            | 247                                | 88.2 ± 14.0 a                             |
| Enzone™ (31.8%)              | 1000 ppm           | 202                                | 94.3 ± 8.1 a                              |
| Mixed nematodes              | 22/cm <sup>2</sup> | 83                                 | 19.3 ± 8.9 b                              |
| Mixed nematodes <sup>a</sup> | 54/cm <sup>2</sup> | 64                                 | 28.1 ± 7.8 b                              |
| Infested control             | —                  | 203                                | 11.5 ± 6.1 b                              |

<sup>a</sup>Represents mixed population of *Heterorhabditis bacteriophora* and *H. indicus* infective juveniles of equal density.

<sup>b</sup>Non-infested control free of larvae. Treatments replicated 6 times. Means followed by the same letter are not significantly different at the 5% level of probability via Tukey's Studentized Range (HSD) Test.

Observations made on larval distribution in soil upon tree removal showed that about 80% were within the tree rhizosphere, usually along tap root. Other larvae were found throughout the soil but not in the upper 2.5 to 5.0 cm. Saturated soil found at the bottom of the container had no apparent effect on larval distribution. Treatment means were compared using the Tukey's studentized range (HSD) test after correction by Abbott's formula (SAS Institute 1990).

As shown in Table 1, Enzone at 500 and 1000 ppm killed 88.2 and 94.3% of the 4-7<sup>th</sup> instar larvae of *D. abbreviatus*, respectively, after 72 hours and had significantly higher mortality than the mixed nematode standard and the infested control. Larval mortality from nematode parasitism was not significantly different from control mortality. Larvae were no doubt missed during the soil sieving process in all treatments; however, some nematode-infected cadavers appeared to have decomposed during the 14 days after treatment based on number recovered. If so, larval mortality is likely to have been more in the order of 60%.

In test 2, 990 live and dead larvae were recovered from all treatments. Although head capsule measurements were not made, observation sug-

gests that most were 4<sup>th</sup> instar or older. Many pupae buried in the soil within 5 days of recovery transformed to the adult stage and teneral adult recovery was good (Table 2). The root system of all infested trees were severely damaged by *D. abbreviatus* larvae.

Adult mortality for the range of dosages tested, was not affected by Enzone™ rate and all rates were significantly higher than the infested control ( $F = 21.54$ ,  $P = 0.001$ ) (Table 2). Enzone™ concentrations of 500 ppm or greater killed 100% of the adults recovered from soil. Though the low number tests prevents statistical analysis, pupae were also highly susceptible to Enzone™ with 100% kill at all concentrations. Larval mortality in relationship to rate was positively linear ( $R^2 = 0.671$ ). Mean mortality for all treatments was significantly different from the control ( $F = 75.84$ ,  $P = 0.0001$ ).

Data presented herein suggest that Enzone is an effective fumigant against all developmental stages of *D. abbreviatus* in infesting containerized citrus. It is imperative that the chemical fully saturate the containers from top to bottom at the time the soil is near saturation to assure complete volatilization (Young 1990). This requirement could limit any field use of the fumigant; however, it has

TABLE 2. EFFECT OF DIFFERENT RATES OF ENZONE™ ON THE SURVIVAL OF VARIOUS DEVELOPMENTAL STAGES OF *DIAPREPES ABBREVIATUS* INFESTING CONTAINERIZED CITRUS TREES SWINGLE CITRUMELO ROOTSTOCK IN THE GREENHOUSE.

| Treatment            | Rate (ppm) | Recovery/treatment |       |        | Mean % mortality ± SE <sup>a</sup> |             |               |
|----------------------|------------|--------------------|-------|--------|------------------------------------|-------------|---------------|
|                      |            | Larvae             | Pupae | Adults | Larvae                             | Pupae       | Adults        |
| Enzone™ (31.8%)      | 2000       | 101                | 4     | 18     | 99.0 ± 0.01 a                      | 100.0 ± 0.0 | 100.0 ± 0.0 a |
| Enzone™              | 1000       | 192                | 0     | 18     | 98.7 ± 0.01 a                      | —           | 100.0 ± 0.0 a |
| Enzone™              | 500        | 183                | 4     | 14     | 94.4 ± 0.02 ab                     | 100.0 ± 0.0 | 100.0 ± 0.0 a |
| Enzone™              | 250        | 182                | 1     | 22     | 90.9 ± 0.02 ab                     | 100.0 ± 0.0 | 95.5 ± 0.04 a |
| Enzone™              | 100        | 195                | 2     | 19     | 71.8 ± 0.05 b                      | 100         | 94.7 ± 0.07 a |
| Infested control     | —          | 137                | 0     | 17     | 12.9 ± 0.05 c                      | —           | 47.1 ± 0.08 b |
| Non-infested control | —          | 0                  | 0     | 0      | —                                  | —           | —             |

<sup>a</sup>Arcsin transformed means ± standard errors within a column followed by the same letter are not significantly different by ANOVA followed by Tukey's Studentized Range (HSD) Test ( $P \geq 0.05$ ). Values based on 5 replications.

potential for quarantine use in citrus and ornamental nurseries if phototoxicity does not pose a problem.

We thank Entek Corp. for supplying product for testing and Angelique Hoyte for technical assistance in the study. Florida Agricultural Experiment Station Journal Series No. R-07264

#### SUMMARY

Enzone™ at rates of 250 ppm or greater was highly effective as a soil fumigant for the near eradication of larvae, pupae and adult *D. abbreviatus* infesting container-grown citrus with no phytotoxic effect.

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SPATIAL DISTRIBUTION OF SOUTHERN CHINCH BUGS  
(HEMIPTERA: LYGAEIDAE) IN ST. AUGUSTINEGRASS

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St. Augustinegrass (*Stenotaphrum secundatum* (Walt.) Kuntze) lawns are utilized throughout the southern United States for their climactic adaptation and their ability to tolerate full sun to moderate shade. The southern chinch bug, *Blissus insularis* Barber is the plant's most damaging insect pest (Crocker 1993). The importance of this insect pest is shown by its ability to develop resistance to insecticides (Reinert & Portier 1983) and overcome host plant resistance (Busey & Center 1987; Cherry & Nagata 1997).

Numerous studies have given anecdotal information on the spatial distribution of southern chinch bugs (SCB) in St. Augustinegrass. Watson (1925) described the grass of an infested lawn as turning brown in patches which might die out completely. He found that around the dead brown spot there was a circular marginal area in which the SCB were feeding turning the grass yellow. Wilson (1929) reported that damage is first noticed in small spots which increase in size if uncontrolled. He also reported that few SCB were in the dead brown grass with most SCB being found in a one to 1.5 m wide strip at the margin of the infestation. Likewise, Reinert (1978) reported taking samples along the periphery of grass damaged by SCB where the population would be the highest. Kerr (1966) reported that SCB occur aggregated in scattered patches rather than being evenly distributed in a lawn. The later studies of Reinert and Kerr (1973) and Crocker and Simpson (1981) also reported that SCB occurred in aggregations in St. Augustinegrass.

Previous studies clearly show that there has been a consensus among SCB researchers that the spatial distribution of SCB in St. Augustinegrass is aggregated. However, a thorough analysis of the spatial distribution of the insect determining if the populations are aggregated and if so, how aggregated, has not been reported. The objective of this study is to report the spatial distribution of SCB in St. Augustinegrass. This information will be useful in understanding the basic biology of the insect and developing sound strategies to sample and control SCB.

The spatial distribution of light infestations of SCB was examined during 1998. A light infestation is defined here as an area of St. Augustinegrass which is less than 1 m<sup>2</sup> containing SCB and damaged yellow grass and the infestation is surrounded by green, healthy appearing grass. Ten light infestations were sampled from May to September, 1998 from urban lawns in Palm Beach County, Florida. Infestations were found by looking for small areas

of yellow appearing St. Augustinegrass and then visually examining the area for SCB in the field. If SCB was detected, a 1 × 1 m sample was taken at the infestation and at 5 m and 10 m from the infestation in the healthy appearing green St. Augustinegrass. Each of the three samples was taken by vacuuming the 1 m<sup>2</sup> for nymphs and adults for 5 minutes using a modified Weed Eater® Barracuda blower/vacuum (Poulan/Weedeater, Shreveport, LA). The use of a vacuuming technique for sampling SCB has been described by Crocker (1993). Each infestation was sampled once with the three samples being taken at the same time (i.e., within minutes of each other). Also, all three samples were taken from the same property to ensure that type of grass, fertilizer, insecticidal spraying, etc. were the same between the three samples. After collection, samples were frozen for later counting in a laboratory. Samples were passed through a U.S.A. Standard Testing Sieve #10 (2 mm opening) to remove large debris. Adults and nymphs were counted by microscopic examination. Data from the 10 infestations were pooled and difference in numbers of nymphs, adults, and total SCB between the three sample areas were determined using Tukey's test (SAS 1996).

The spatial distribution of moderate infestations of SCB was examined during 1999 at different infestations than sampled in 1998. A moderate infestation is defined here as an area of St. Augustinegrass which is greater than 4 m<sup>2</sup> containing SCB and damaged yellow to brown dead grass and the infestation is surrounded by healthy appearing green grass. Ten moderate infestations were sampled from May to September, 1999 from urban lawns in Palm Beach County, Florida. Infestations were located as previously described. Three 1 × 1 m samples were taken at each infestation. One sample was taken in the interior area of brown, dead St. Augustinegrass. A second sample was taken at the infestation edge which was yellow grass bordering the interior area. The third sample was the exterior being 5 m from the edge into green, healthy-appearing grass surrounding the infestation. The 10 infestations were sampled by vacuuming as previously described. Each infestation was sampled once with the three samples being taken at the same time (i.e., within minutes of each other). All three samples were taken on the same property to again ensure insecticidal spraying, etc. was the same between samples. After collection, samples were processed as previously described. Statistical analysis was performed as previously described.

Previous samples had been taken by vacuuming different areas for chinch bugs. However, interior and edge areas of moderate infestations often had yellow to brown or dying grass which was thinner and less lush than green grass in exterior areas. Hence, it appeared that vacuuming for chinch bugs may have been more efficient in interior and edge areas and not reflecting true population differences between those areas and the green exterior areas. Thus, flotation samples (Kerr 1966) were also taken since the efficiency of measuring chinch bug populations using this method would not be affected by differences in grass thickness. From June to October, 1999, 25 moderate infestations were sampled in St. Augustinegrass in urban lawns in Palm Beach County, Florida. Infestations were located as previously described and were new infestations from previously sampled infestations. Three samples were taken at each infestation. One sample was taken in the interior, edge, and exterior of each infestation, these areas having been defined previously. Each sample was a 25 cm diameter grass sample dug down 15 cm. Each sample was placed in a plastic bucket and covered with a fine mesh cloth to prevent escape of insects. Samples were taken to a laboratory where water was slowly added to buckets over a two hour period. Chinch bugs surfacing were aspirated and nymphs and adults counted. Statistical analysis was performed as previously described.

Significantly more nymphs, adults, and total SCB were found in suction samples at the light in-

festation (0 m) than at 5 m or 10 m from the infestation (Table 1). There was no significant difference in numbers of nymphs, adults, or total SCB in suction samples between 5 m and 10 m from the light infestation.

Significantly more nymphs, adults, and total SCB were found in suction samples at the infestation edge than the interior or exterior of moderate infestations (Table 1). Significantly more total SCB were found in suction samples in the interior of moderate infestations than the exterior. As in suction samples, significantly more nymphs, adults, and total SCB were found in flotation samples at the infestation edge than the interior or exterior of moderate infestations (Table 1). Also as in suction samples, significantly more total SCB were found in flotation samples in the interior of the moderate infestation than the exterior.

Both suction samples and flotation samples show that SCB are extremely aggregated in yellow damaged grass in light and moderate infestations with much fewer SCB in the green grass 5 m or more away from an infestation. However, the efficacy of SCB control using spot treatments of an insecticide only at SCB infestations versus complete lawn coverage with an insecticide is not known. To compare these two treatments, possible SCB resurgence, control of other lawn pests, and predator mortality also need to be considered and there are no data currently available comparing the two methods. Future research is needed to determine the overall effect on all lawn arthro-

TABLE 1. SCB IN DIFFERENT AREAS OF ST. AUGUSTINEGRASS INFESTATIONS.

|        | Meters from light infestation <sup>1</sup> |                  |              |
|--------|--|------------------|--------------|
|        | 0  | 5                | 10           |
| nymphs | 1780.8 ± 269.1 A                           | 11.0 ± 3.1 B     | 12.0 ± 4.6 B |
| adults | 115.9 ± 28.7 A                             | 2.6 ± 1.5 B      | 0.8 ± 1.1 B  |
| total  | 1896.7 ± 333.2 A                           | 13.6 ± 4.5 B     | 12.8 ± 6.1 B |
|        | Moderate infestation <sup>1</sup>          |                  |              |
|        | Interior                                   | Edge             | Exterior     |
| nymphs | 62.3 ± 20.4 B                              | 1462.9 ± 359.5 A | 1.9 ± 1.0 C  |
| adults | 1.9 ± 1.1 B                                | 94.9 ± 23.4 A    | 0.9 ± 0.3 B  |
| total  | 64.2 ± 21.5 B                              | 1557.8 ± 365.8 A | 2.8 ± 0.8 C  |
|        | Moderate infestation <sup>2</sup>          |                  |              |
|        | Interior                                   | Edge             | Exterior     |
| nymphs | 7.3 ± 1.1 B                                | 73.8 ± 15.3 A    | 1.8 ± 0.7 C  |
| adults | 4.8 ± 0.9 B                                | 49.5 ± 12.3 A    | 0.7 ± 0.3 C  |
| total  | 12.1 ± 2.0 B                               | 123.3 ± 20.1 A   | 2.5 ± 0.9 C  |

<sup>1</sup>Means ± 1 SE SCB in 1 m<sup>2</sup> suction samples. Means in a row followed by the same letter are not significantly different (alpha = 0.05) using Tukey's test (SAS 1996).

<sup>2</sup>Means ± 1 SE SCB in 25 cm diameter flotation samples. Means in a row followed by the same letter are not significantly different (alpha = 0.05) using Tukey's test (SAS 1996).



pods, pests and beneficials, to determine if spot treatment or broad coverage of insecticides should be used for SCB control. Lastly, it should be noted that heavy SCB infestations do occur, although much less frequently than the light to moderate infestations described in this paper. In heavy SCB infestations, areas of SCB damage spread and merge so that SCB may be found throughout a lawn. In heavy SCB infestations, little St. Augustinegrass is remaining and resodding of the lawn may be necessary.

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#### SUMMARY

The spatial distribution of southern chinch bugs, *Blissus insularis* Barber, in St. Augustinegrass was examined. In light to moderate infestations southern chinch bugs are extremely aggregated in small areas of lawns surrounded by large areas of green grass which contain few chinch bugs.

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## COLLECTION OF SOYBEAN LOOPER AND OTHER NOCTUIDS IN PHENYLACETALDEHYDE-BAITED FIELD TRAPS

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Many adult lepidopteran pests are monitored in agricultural systems using sex pheromone-baited traps. However, chemicals other than sex pheromones have been isolated, identified and bioassayed as moth attractants. Field studies have shown that traps baited with phenylacetaldehyde capture various noctuid subfamilies, especially Plusiinae (Smith et al. 1943, Creighton et al. 1973, Cantelo & Jacobson 1979). Enhanced upwind flight and increased trap capture have resulted when phenylacetaldehyde has been combined with sex pheromones (Creighton et al. 1973, Meagher & Mitchell 1998) or blacklights (Cantelo & Jacobson 1979). Phenylacetaldehyde has the potential in agricultural systems to non-specifically attract both female and male moths.

Soybean looper moths, *Pseudoplusia includens* (Walker), have been captured in traps baited with phenylacetaldehyde (Smith et al. 1943, Creighton et al. 1973), but the traps were large and are not in use today. This note describes capture of *P. includens* and other noctuids in traps currently used in agricultural settings.

Two experiments were conducted in northwestern Alachua County, Florida. The first experiment used Unitraps (Great Lakes IPM, Vestaburg, MI) that were placed along roads and edges in an 80-ha field of cotton, *Gossypium hirsutum* L. Traps were baited with lures containing phenylacetaldehyde (0.5 ml) placed in hollow polyethylene stoppers (Kimble, Vineland, NJ, purchased through Thomas Scientific, Swedesboro, NJ, #9713-F28). Traps contained insecticide strips to kill moths that were captured (Hercon® Vaportape II containing 10% 2, 2-dichlorovinyl dimethyl phosphate, Hercon Environmental Co., Emigsville, PA). Trap contents were removed three times per week and soybean looper moths were collected from 21 July to 12 September 1997.

The second experiment was designed to capture noctuid moths during a part of the season when there was no commercial field crop. Unitraps baited with 0.5 ml phenylacetaldehyde in stoppers or unbaited traps were placed in the middle of a 60-ha field of grain sorghum, *Sorghum bicolor* (L.) Moench, that was harvested and regrew prior to the experiment. The experiment was designed as a randomized complete block with three blocks of the two treatments (baited or unbaited), and trap location within a block was randomized weekly. Moths were collected from 4 December 1998 through 17 February 1999.

Moths were identified with the aid of Kimball (1965) and Covell (1984), and comparison with identified specimens in the Florida State Collection of Arthropods, Florida Department of Agriculture and Consumer Services, Division of Plant Industry, Gainesville.

Moth numbers per night from the sorghum field were compared across treatments using a split block analysis of variance (ANOVA), where treatment was the main plot and date was the subplot (Steel & Torrie 1980). To satisfy ANOVA assumptions, counts were  $\log(x + 1)$  transformed before analysis. Treatment means were separated using an LSD mean separation test (PROC GLM, SAS Institute 1996). Untransformed means ( $\pm$ SE) are given in the text, whereas statistical results refer to transformed data.

In the cotton field, soybean looper moths were noticed in traps starting in early August but were not recorded numerically until late August (Fig. 1). Moths were collected in traps through September. In the sorghum field, almost one half of the moths collected were either *Mocis latipes* (Guenée) or *Leucania* sp., although species from six noctuid subfamilies were represented in trap captures (Table 1). Similar numbers of male and female moths were collected in traps baited with phenylacetaldehyde. Baited traps collected more moths than unbaited traps ( $P < 0.05$ ), as only 1 moth was collected in the unbaited traps.

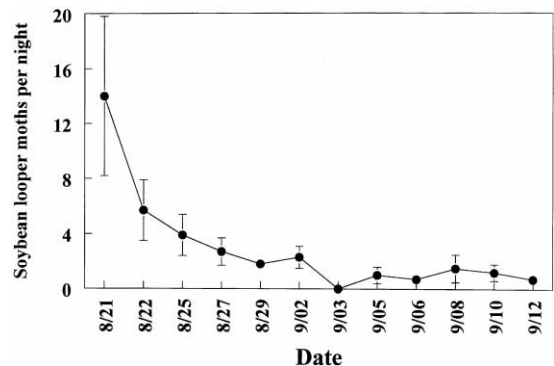


Fig. 1. Collection of soybean looper moths in a cotton field with Unitraps baited with phenylacetaldehyde, Alachua Co., FL, 1997.

TABLE 1. NUMBER OF NOCTUID MOTHS COLLECTED IN A FIELD OF REGROWTH SORGHUM WITH UNITRAPS BAITED WITH PHENYLACETALDEHYDE, 4 DECEMBER, 1998—17 FEBRUARY, 1999, ALACHUA, FL.

| Subfamily/Species                      | Females   | Males     |
|--|-----------|-----------|
| <b>Amphipyridae</b>                    |           |           |
| <i>Platysenta mobilis</i> (Walker)     | 1         | 0         |
| <i>Spodoptera dolichos</i> (F.)        | 0         | 1         |
| <i>S. latifascia</i> (Walker)          | 1         | 4         |
| <b>Catocalinae</b>                     |           |           |
| <i>Mocis disseverans</i> (Walker)      | 2         | 0         |
| <i>M. latipes</i> (Guenée)             | 16        | 14        |
| <i>M. marcidia</i> (Guenée)            | 5         | 5         |
| <b>Hadeninae</b>                       |           |           |
| <i>Leucania</i> sp.                    | 22        | 15        |
| <i>Pseudaletia unipuncta</i> (Haworth) | 1         | 0         |
| <b>Heliothinae</b>                     |           |           |
| <i>Helicoverpa zea</i> (Boddie)        | 1         | 0         |
| <i>Heliothis virescens</i> (F.)        | 1         | 0         |
| <b>Noctuinae</b>                       |           |           |
| <i>Agrotis subterranea</i> (F.)        | 3         | 5         |
| <i>Anicla infecta</i> (Ochsenheimer)   | 1         | 1         |
| <b>Plusiinae</b>                       |           |           |
| <i>Grapha oxygramma</i> (Geyer)        | 3         | 1         |
| <i>Argyrogramma verruca</i> (F.)       | 0         | 3         |
| <i>Autographa biloba</i> (Stephens)    | 2         | 1         |
| <i>Pseudoplusia includens</i> (Walker) | 0         | 2         |
| <i>Rachiplusia ou</i> (Guenée)         | 4         | 6         |
| <b>TOTAL</b>                           | <b>63</b> | <b>58</b> |

## SUMMARY

This research determined some of the economically-important noctuids that can be captured using phenylacetaldehyde as a lure. It was confirmed that phenylacetaldehyde can be used for monitoring (presence or absence) or sampling (numbers per time per area) using commercially available insect traps. These traps are currently used to sample pest noctuids by growers, consultants and county extension workers.

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### THREE FUNGAL SPECIES ISOLATED FROM *COPTOTERMES FORMOSANUS* (ISOPTERA: RHINOTERMITIDAE) BODIES, CARTON MATERIAL, AND INFESTED WOOD

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The Formosan subterranean termite (FST), *Coptotermes formosanus* Shiraki (Isoptera: Rhinotermitidae), is one of the most destructive termites in the U.S., causing millions of dollars in damage annually to wood products and living trees (Beal 1987, Edwards & Mill 1986, La Fage 1987, Su & Tamashiro 1987).

Termite-fungus associations have been extensively studied (Alasoadura 1966, Batra and Batra 1966, Bose 1923, Bottomley and Fuller 1921, Fletcher 1921, Lund 1960a, 1960b, 1962, Roonwal 1960, Sands 1970). The objective of this study was to isolate and identify fungi associated with *C. formosanus* that might help in their nutrition.

Collections were made in the metropolitan New Orleans, LA, area. The search was limited to fungi associated with wood decay, and fungal spores present externally on the body of alates, workers and soldiers, since these can be ingested during grooming of nestmates. Alates were collected by the use of UV (22-watt circle black light, BioQuip, Gardena, CA) light traps placed in three different locations during the 1998 swarming season (May-August). The collected alates were transferred into plastic boxes lined with wet paper and placed into a Percival® environmental chamber at 25°C ± 1°C until they lost their wings. Then, 100 pairs (male and female) were transferred to sterile glass vials containing 3 ml of sterile media composed of yellow pine dust: agar (Difco, Detroit, MI,) (1:3) (King et al. 1974). The vials were closed with sterile cotton and incubated at 27 ± 1°C and 70% RH for 30 d. Twenty mated pairs were randomly selected for extraction of fungi.

Workers and soldiers were extracted from logs and debris found in three locations. Thirty groups of 10 termite workers and two soldiers were transferred to sterile plastic Petri dishes containing 10 g of the media (10 dishes per location) described above. The edges of the dishes were sealed with Parafilm® to maintain humidity. The dishes were kept in the chamber as above.

Samples of wood and carton material from seven wind-fallen FST infested trees were collected following a storm (Hurricane Georges, September 1998). Ten samples from each tree were individually transferred to sterile Petri dishes and humidified with sterile ionized-Q water to favor fungal growth. The dishes were incubated for 30 d and fungi were isolated and identified using the methods described below.

Ten dishes of each group and 20 mated pairs were selected for isolation of fungi. The samples consisted of three small pieces of media collected from the inside of the nuptial chamber of the alates where mycelia growth was evident. Samples were transferred to sterile Petri dishes containing 15-ml sterile media, to favor the isolation of rapid growth fungi, which are more likely to be an abundant source of supplementary food.

Two media were used: 1) PDA (potato-dextrose-agar, Difco, Detroit, MI) prepared according to the label and 2) water: agar: yellow pine dust (97.5:1.5:1). Both media were autoclaved for 20 min at 120°C, cooled to 55°C in a 50°C water bath, and acidified with lactic acid to a 5.6 ± 0.2 pH. The edges of the inoculated dishes were also sealed with Parafilm® to maintain humidity. These dishes were incubated at 27 ± 1°C for 1 and 2 weeks, respectively.

The dishes were then examined under a stereo microscope to find conidia and mycelia. One technique to isolate pure cultures was designed to favor the isolation of fungi associated with wood decay. Seven sterile 2 × 2 × 0.5 cm pieces of woods: sweetgum, *Liquidambar styraciflua* L. (Hamameliaceae), pecan, *Carya illinoensis* (Wangenh.) K. Koch (Juglandaceae), and yellow birch, *Betula alleghaniensis* Britton (Betulaceae) were individually placed into Petri dishes containing sterile PDA. Each piece of wood was inoculated on one of the edges with a small piece of agar and fungal mixture. The dishes were sealed and incubated as above for 7 d. Small pieces of agar with mycelia from each culture was transferred to sterile PDA to obtain pure fungal cultures. This was repeated 2 times for each culture.

The second technique was designed to isolated saprophytic fungi, 1-ml spore suspensions of the mixed fungal cultures were made by scraping culture plates containing 10 ml of 0.01% Triton X-100 (Amresco, Solon, OH). Spore suspensions were enumerated using a Levy hemacytometer (Hausser, Horsham, PA), and diluted with 0.01% Triton X-100 to a final concentration of approximately 1 × 10<sup>8</sup> spores/ml. A 100 µl aliquot of each suspension was spread, with a sterile glass hockey stick onto an agar plate containing 10 ml of the above PDA. Plates were incubated at 25 ± 1°C for 2-4 d, until small colonies were visible. Each colony was individually transferred by sterile loop to one section of a quadrant Petri dish con-

TABLE 1. PERCENT OF SAMPLES OF DIFFERENT SUBSTRATES WITH SPECIES OF FUNGI ASSOCIATED WITH *COPTOTERMES FORMOSANUS* IN NEW ORLEANS, LA.

| Substrate                                    | Species |      |     |
|--|---------|------|-----|
|  | Af      | An   | Cl  |
| Infested trees <sup>a</sup>                  |         |      |     |
| Ulmaceae                                     |         |      |     |
| American elm, <i>Ulmus americana</i> L.      | 80      | 80   | 50  |
| Chinese elm, <i>Ulmus parvifolia</i> Jacq.   | 80      | 80   | 44  |
| Fagaceae                                     |         |      |     |
| Live oak, <i>Quercus virginiana</i> Mill.    | 78      | 78   | 60  |
| Aceraceae                                    |         |      |     |
| Red maple, <i>Acer rubrum</i> L.             | 65      | 65   | 70  |
| Platanaceae                                  |         |      |     |
| Sycamore, <i>Platanus occidentalis</i> L.    | 60      | 60   | 55  |
| Salicaceae                                   |         |      |     |
| Willow, <i>Salix babylonica</i> L.           | 60      | 60   | 80  |
| Taxodiaceae                                  |         |      |     |
| Bald cypress, <i>Taxodium distichum</i> (L.) | 45      | 45   | 50  |
| Termites                                     |         |      |     |
| Cartoon material <sup>b</sup>                | 25      | 25   | 100 |
| Alates <sup>c</sup>                          | 100     | 100  | 100 |
| Workers <sup>c</sup>                         | 100     | 1001 | 100 |

Percentage of successful isolation from 30 samples per substrate. Af = *Aspergillus fumigatus* Fresenius, An = *Aspergillus nomius* Kurtzman, and Cl = *Curvularia lunata* (Wakker).

<sup>a</sup>By *C. formosanus*.

<sup>b</sup>From nests of *C. formosanus* found in . . .

<sup>c</sup>From external body parts.

taining the PDA as above, incubated at 25± 1°C for 7 d until sporulation, and transferred by sterile loop to new PDA plates.

Plates containing pure cultures of the fungi were incubated at 25 ± 1°C for 7 d to induce sporulation. Identification was accomplished after growth on standard media using Ellis (1971) (for *Curvularia*), and Klich and Pitt (1988) (for *Aspergillus*) taxa keys. Identification of *Aspergillus nomius* was confirmed using the methods of: Kurtzman et al. (1987) and Singh et al. (1991).

Species of fungi isolated from the reproductive and worker termites placed in the pine medium, carton material, and infested trees were identified as *Curvularia lunata* (Wakker) Boedijn (Pleosporales: Pleosporaceae), *Aspergillus fumigatus* Fresenius, and *Aspergillus nomius* Kurtzman, Horn, and Hesselstine (Eurotiales: Trichocomaceae) (Table 1).

*Curvularia lunata* is a facultative pathogen of mainly monocotyledonous plants (Bhale et al. 1982, Bisen 1983, Domsch et al. 1980, Gadage & Patil 1977, Kore & Bhide 1981, Pearson & Muki 1982).

*Aspergillus fumigatus* is ubiquitous (Cutler et al. 1996, Dorner et al. 1984, Ekundayo 1983, Klich & Pitt 1988, Pal et al. 1986, Rath et al. 1997).

*Aspergillus nomius* has been isolated from insects (Kurtzman et al. 1987, Ito et al. 1997) and plant substrates (Kurtzman et al. 1987; Feibelman et al. 1998).

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#### SUMMARY

Three species of imperfect fungi (*Curvularia lunata*, *Aspergillus fumigatus* and *Aspergillus nomius*) were isolated from the body of *Coptotermes formosanus* alates and workers from 6 different locations around the Greater New Orleans area. Samples from 7 species of trees infested by *C. formosanus* as well as their carton material also presented these fungi. *C. lunata* growth was favored in the carton material while the *Aspergillus* species growth was favored in the wood. The possibility of a termite-fungi association is discussed.

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A RECORD OF *MICROMALTHUS DEBILIS*  
(COLEOPTERA: MICROMALTHIDAE) FROM CENTRAL AMERICA AND A  
DISCUSSION OF ITS DISTRIBUTION

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While going through a sample from a flight intercept trap set in Belize, I came across a specimen of *Micromalthus debilis* LeConte, apparently the first record from Central America. This species, belonging to the monotypic family Micromalthidae, is recorded as native to eastern North America. While *M. debilis* might be common and is known from several states (Downie & Arnett 1996), individual collecting events appear rare.

In addition to being classified in a monotypic family, this species has an intriguing biology. Larvae develop in moist decaying hardwoods and can go through several stages and shapes in their development, appearing as caraboid, cerambycoid, and curculionoid larval types. The complicated lifecycle involves paedogenesis and several types of parthenogenesis (thelytoky, amphitoky, and arrhenotoky) (for details see Barber 1913a, b; Pringle 1938; Scott 1936, 1938, 1941).

The classification of this species has undergone much discussion and controversy. Although Downie & Arnett (1996) place the family within the Polyphaga based on several shared adult features with the Cantharoidea (or Elateroidea sensu Lawrence & Newton [1995]), overwhelming evidence from larval, wing, and male genitalic characters indicates that placement of the family within the Archostemata is correct (Lawrence & Newton 1995).

It is apparent that *M. debilis* was once much more widespread. Fossil larval records from amber are known from Lebanon (Cretaceous), the Baltic (Oligocene), the Dominican Republic (M. Lviae, pers. comm.) and Chiapas, Mexico (late Oligocene or early Miocene) (Lawrence & Newton 1995; Rozen 1971). Currently, *Micromalthus debilis* is considered native only to the eastern United States. Other North American records are British Columbia and New Mexico (Borror et al. 1986). More distant and overseas localities are South Africa (Scholtz & Holm 1985), Cuba, Brazil, Hong Kong, and Hawaii (Lawrence 1982). Arnett's (1968) record from Europe, based on Silvestri (1941), is in error, as Silvestri only stated that *M. debilis* should be found in Europe due to the import of large amounts of timber from the New World.

Most of these records (outside the eastern U.S.) undoubtedly are the result of introductions via the spread of larvae in dead wood (Lawrence &

Newton 1995). But it can sometimes be difficult to ascertain whether a species is native or introduced, as Whitehead and Wheeler (1990) pointed out.

The Belize specimen was collected in an area of relatively pristine forest (Orange Walk District, Rio Bravo Conservation Area, Well Trail near Research Station, 16-18.IV.1995, flight intercept trap, P. W. Kovarik), distant from any commercial center. This record appears to represent a disjunction, but the fossil record shows that the species was once present in nearby southern Mexico. Individuals are only rarely collected, and the sampling effort for small beetles in southern Mexico and Belize has been minimal, especially compared to that in eastern North America. Also, one of the known hosts, *Quercus*, is present in the Rio Bravo Conservation Area (P. Kovarik, personal communication). Hence, *M. debilis* might have a much more widespread native distribution than previously thought, and this Belize record might not be the result of an introduction. It is even possible, though perhaps less likely, that the Cuban record represents a natural distribution in the West Indies. Regardless of the factors resulting in the present distribution, it remains to be discovered how widespread this beetle family may actually be. I thank Alfred Wheeler and one anonymous reviewer for their comments.

#### SUMMARY

*Micromalthus debilis* LeConte is recorded from Central America for the first time. The possibility that it is native, and not introduced, is discussed.

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## BOOK REVIEWS

COLON-FERRER, M., AND S. MEDINA-GAUD. 1998. Contribution to the systematics of the diaspidids (Homoptera: Diaspididae) of Puerto Rico. Agricultural Experiment Station, University of Puerto Rico. ix + 254 p. (Including 14 color photographs). \$20 plus \$3.00 for postage, from Editor's Office, College of Agricultural Sciences, PO Box 21360, Rio Piedras, Puerto Rico 00928.

A regional publication about armored scale insects is a useful instrument for entomologists concerned with identifications from that region. This serves as a useful tool for extension entomologists, regulatory entomologists, and students of entomology. Many pertinent literature sources are combined generally to make identifications easier without consulting different literature sources. A concise review of the Puerto Rican and other geographical areas literature is given.

In this publication the authors have covered 77 species of armored scale insects. Three genera are recorded for the first time in Puerto Rico. Keys are included for identification of the 11 scale insect families known to occur in Puerto Rico, for 3 tribes, and for the respective species.

Preservation methods and microscope slide mounting techniques are covered; however, with a few limitations. One limitation is the use of Essig's Fluid with the acronym (EAF), which is Essig's Aphid Fluid, without any mention of its contents. Another glaring mistake is the symbol for potassium hydroxide (KOH) is given as KHO (possibly correct but unconventional).

A host plant index is also provided. This is helpful as some armored scale insects are host specific or nearly so. Parasite and predator information, where available, is provided.

Each species has a short field description of female and male armor with a more extensive morphological description of the female body. A line drawing of the typical type used for scale insects is given for each species, although only one drawing appears to be original, and the others are taken from the cited literature sources.

Overall, this is an adequate regional treatment of important plant pests, the armored scale insects. If you can overlook the numerous typographical errors and the egregious mistaken identity of Photo 2b as *Aonidiella orientalis*, when it is obviously *Diaspis boisduvalii* you will be happy with this compilation of information on armored scale insects.

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## A PERSONAL ACCOUNT OF PROGRAMS TO ERADICATE THE SCREWORM IN THE UNITED STATES AND MEXICO WITH SPECIAL EMPHASIS ON THE FLORIDA PROGRAM

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### ABSTRACT

The great saga of the eradication of the screwworm first from Curacao and then from all of North and Central America is recounted with special emphasis on activities in Curacao and Florida from 1951 through 1959. The author, who worked as a research scientist on all aspects of laboratory and field research and operations, brings to light many biological and operational problems along with corresponding solutions, which are not treated in the published accounts of USDA administrators involved in these programs.

Curacao served as a 170 square mile outdoor laboratory for developing the sterile insect technique. This setting permitted quantitative determination of the dynamics of the wild population, and the overflooding ratios and dispersal patterns essential for population suppression. The attack on the wild population during the time of year when it naturally undergoes decline proved to be essential in achieving eradication with minimal resources.

The Florida programs yielded three extremely important findings. The first is that eradication is greatly facilitated by taking advantage of severe weather events which reduce the range and density of the target population. Secondly eradication cannot be readily attained merely by the release of sexually sterile insects, since it is absolutely essential that producers simultaneously attack the immature stages by diligent inspection and treatment of wounds. Thirdly the leadership of the producer clientele is critically important to securing program resources from livestock owners, the State Legislature and the Congress. These lessons were corroborated repeatedly as the program dealt with the southwestern USA, Mexico and the Central American countries.

Key Words: Screwworm, *Cochliomyia hominivorax*, sterile insect technique, eradication, Curacao, history

The full text of this document can be viewed at: <http://www.flaentsoc.org/baumhover1997annmeet.html>

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