

A NEW QUILL MITE (ACARINA: SYRINGOPHILIDAE)
FROM THE GROUND DOVE

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ABSTRACT

Peristerophila mucuya sp. n. (Acarina: Syringophilidae) is described from a Ground Dove, *Columbina passerina* (Columbiformes: Columbidae) collected at Millett, La Salle County, Texas.

RESUMEN

Peristerophila mucuya sp. n. (Acarina: Syringophilidae) se describe de una Mucuya, *Columbina passerina* (Columbiformes: Columbidae), conseguido de Millett, La Salle County, Texas.

INTRODUCTION

The anatomical terminology and setal designations used in the descriptions follows Kethley (1970; 1973). All measurements are in microns. The range and mean of selected metric characters for paratypes follow in parentheses those of the holotype and allotype. The descriptions are based on the study of the female holotype, male allotype, 10 female paratypes and 10 male paratypes. Nomenclature for the type host follows the thirty-second supplement to the American Ornithologists' Union checklist of North American birds.

It was previously reported (Casto 1976) that an undescribed species of the genus *Peristerophila* (Acarina: Syringophilidae) occurs within the quills of the Ground Dove, *Columbina passerina* (Columbiformes: Columbidae). The study of additional specimens of this form has resulted in the descriptions which follow.

PERISTEROPHILA MUCUYA SP. N.

All type specimens were taken from the primary and secondary coverts, alulars and tail coverts of a Ground Dove, *Columbina passerina* (Columbiformes: Columbidae), collected May 27, 1974, at Millett, La Salle County, Texas, by S. D. Casto. The species name *mucuya* (fem. sing.) is the Spanish common name for the type host.

FEMALE (holotype). Length 940 (920 to 1020, 955); propodosomal width 130 (110 to 130, 123). Gnathosoma: Hypostomal apices ornamented; two pairs of median protuberances (Fig. 1b). Each lateral branch of peritremes (Fig. 1c) with 1-2 chambers; each longitudinal branch with 4-5 chambers. Chelicerae 127 (118 to

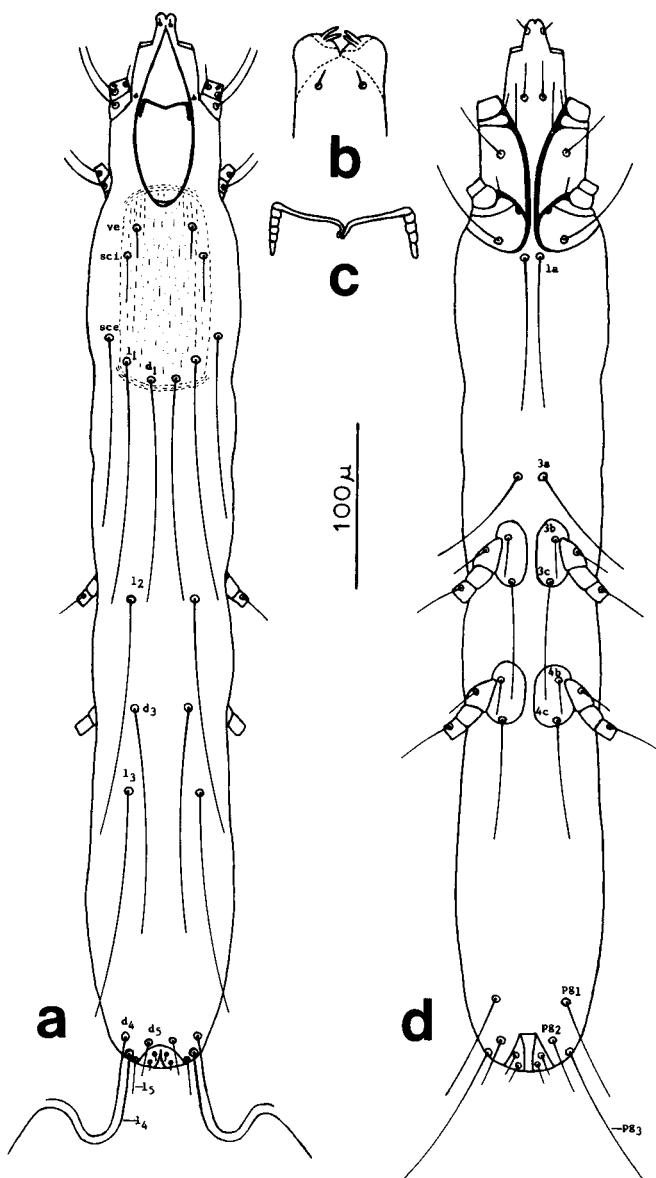


FIG. 1. -- *Peristerophila mucuya* sp. n. Female. a. Dorsum. b. Dorsal view of hypostome. c. Peritreme. d. Venter.

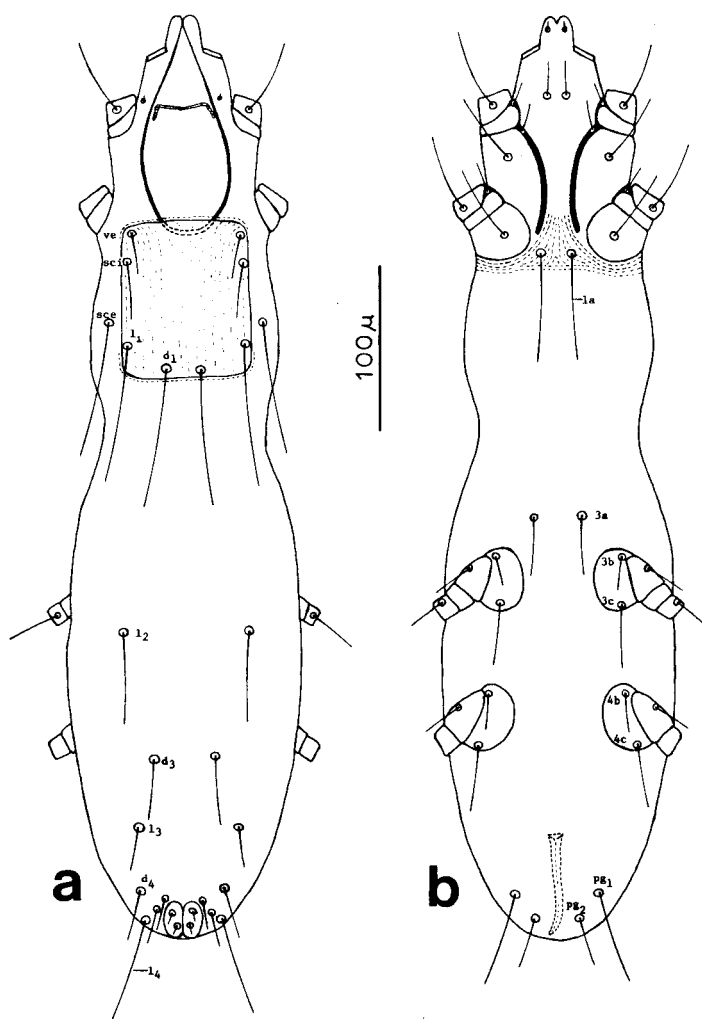


FIG. 2. -- *Peristerophila mucuya* sp. n. Male. a. Dorsum.
b. Venter.

to 131, 123) in length; edentate. Stylophore 160 (155 to 164, 160) rounded posteriorly. *Dorsal Idiosoma* (Fig. 1a): Propodosomal plate obscure; region of plate with less pronounced striae and occasional punctations. Ratios ve:sci:sce, 1:1.6:7.3. Hysterosomal plate absent. d5 and l5 subequal. d4 2/3 length of l4. *Ventral Idiosoma* (Fig. 1d): Setae la extending 2/3 way to 3a. Ratios pg1:pg2:pg3, 2.5:1:3.5. *Legs*: Legs I longer and thicker than legs II; length legs IV, trochanter to tip of claws, 100 (95 to 102, 100). a' and a'' legs I with 7-10 tines. a' and a'' of legs II to IV with 11-14 tines. 3c and 4c subequal. Coxae of legs III and IV almost touching on the ventral midline of some specimens. Chaetotaxy of legs: coxae 2-1-2-2, trochanters 1-1-1-1, femora 2-1-0-0, genua 3-2-1-1, tibiae 5-4-3-3, tarsi 11-7-6-6.

MALE (allotype). Length 540 (500 to 560, 530); propodosomal width 120 (90 to 120, 108). Hypostomal apices slightly roughened but without definite ornamentation. Each lateral branch of peritremes with 1-2 chambers; each longitudinal branch with 3-6 chambers. Chelicerae 100 (100 to 107, 104) in length; edentate. Length stylophore 122 (122 to 131, 126) extending below propodosomal plate. *Dorsal Idiosoma* (Fig. 2a): Propodosomal plate weakly sclerotized with indistinct margins. Ratios ve:sci:sce, 1:1.3:3.7. Ratios d3:l3:d4, 1.5:1:1. d4 1/2 length of l4. *Ventral Idiosoma* (Fig. 2b): Setation as shown. Two pairs of paragenitals. Aedeagus 64, curved. *Legs*: a' and a'' of legs I to IV with 5-7 tines. MCA1 and MCA2 lightly sclerotized; not fused. Coxae of legs III and IV inserted more toward lateral margin of body than on female.

Types: Female holotype, male allotype, 1 female paratype and 1 male paratype USNM Acarology Coll. No. 3965, remaining specimens in collection of author.

REMARKS

The condition of the mesal coxal apodemes in the genus *Peristrophe* is described as "MCA1 parallel, not fused to MCA2" (Kethley 1970). However, the illustrations of *P. columba* (Figs. 32 and 33) which accompany the generic description show MCA1 to be fused to MCA2. My examination of specimens of *P. columba* show the apodemes to be fused in both males and females. MCA1 and MCA2 of *P. zenadoura* females are in contact and often appear to be firmly fused. In contrast, the apodemes of *P. zenadoura* males are somewhat separated with evidence of only a very slight fusion. The observation that MCA1 and MCA2 are fused in *P. mucuya* females, but not in the males thus represents what may be a trend toward a sexual dimorphism of this character.

P. mucuya most closely resembles *P. longisoma* which has been recorded from the White-winged Dove and Mourning Dove (Casto 1979). It may, however, be distinguished from *P. longisoma* by its shorter length, the close-set coxae on the ventral midline, and the absence of setae vF on legs III and IV.

ACKNOWLEDGMENT

The type host was collected during studies on acarine taxonomy conducted in the laboratory of Dr. Danny B. Pence, Dept. of Comparative Pathology, Texas Tech University School of Medicine.

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A COMPARISON OF BOLL WEEVIL DAMAGE (%) TO BOLLS
IN DIFFERENT COTTON GENOTYPES^{1/},^{2/}

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ABSTRACT

Studies during 1975 and 1978 at Uvalde, TX indicated that the cotton variety TAMCOT SP-37 experienced a lower % damage to boll locks from boll weevils than several experimental cotton genotypes. A more rapid establishment of bolls earlier in the fruiting period may be responsible for this advantage in TAMCOT SP-37.

INTRODUCTION

Early into their experience with the boll weevil, Anthonomus grandis Boheman, entomologists determined that the pest was less a threat to bolls than squares, and that younger bolls were more vulnerable to injury than older bolls. The utilization of genotypes that established a crop of bolls quickly in a growing season was, for all purposes, the one practical accommodation available in dealing with the insect. For years, the cultivation of more rapidly fruiting cottons was the primary defense against the insect.

Boll weevils do damage cotton bolls, and various theories have been put forth on the factors that impinge on the relationship of the pest and bolls of the plant. Thickness and hardness of the carpel wall have been noted (Hunter and Pierce 1912, Cook 1906, and Fenton and Dunnam 1929), and it has been considered that the presence of abundant squares diverts the pest from bolls (Townsend 1895). In fact, through the years it has become conventional wisdom that if cotton continues to produce squares late in the fruiting period, a buffer will be provided for the plant's bolls.

^{1/} Coleoptera: Curculionidae.

^{2/} Tech. Cont. 15524, approved for publication Sept. 27, 1979. The work herein reported was funded in part by an IBP sponsored project entitled "The Principles, Strategies and Tactics of Pest Population Regulation and Control in Major Crop Ecosystems." This support was made available through a grant from National Science Foundation (NSF GB 34718) and a cooperative agreement with AR/SEA, USDA. The work also was supported by the Rockefeller Foundation.

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A clearer view of the matter was recently provided by Parker et al. (1980). This investigation shows that boll age at the time the weevil begins to increase sharply in numbers is the determinant of whether or not, and to what degree, boll locks will be damaged by the insect. It is a critical relationship — every day a boll ages before weevils become severe increases the chances for escaping damage.

Although we believe that boll age at the time of weevil increase is the overriding consideration in describing lock injury, there is evidence that the % of bolls or locks damaged may be influenced by genotype and perhaps by its fruiting habit. This paper deals with the lowered % of weevil damage to locks that we have measured in the variety TAMCOT SP-37.

METHODS

Field plot experiments were conducted in Uvalde County in 1975 and 1978 in which a number of cotton genotypes were examined for agronomic performance. Several variables were included in the tests. Boll weevils were the only economically important insects at the study locations.

1975. A split split-plot arrangement was used to investigate levels of insecticidal use for the control of overwintered weevils, plant spacing, and cotton genotypes. Main plots were treated with either 1 or 3 early spray applications of an organophosphate insecticide. Main plots were divided into 3 spacing sub-plots: Conventional single-drill planting on beds 0.96 m apart; 2 drills on 0.96 m beds; and single-drills on beds 0.64 m apart. Spacing sub-plots were divided into several sub sub-plots, each containing a different genotype. The genotype sub sub-plots were 2 rows wide where 0.96 m beds were used and 3 rows wide for the 0.64 m beds. Plots were 15.2 m in length; there were 4 replications.

Within each sub sub-plot a 1.9 m length of row was marked. In these plots flowers were tagged each day; bolls developed from these blooms were examined later to determine lock damage. After the early treatments for overwintered weevils, no additional applications of insecticides were made.

1978. This experiment also was arranged in a split split-plot. However, the entire planting was treated in early season with 3 applications for overwintered weevils. Main plots were the 3 spacings used in 1975; cotton genotypes were assigned to sub-plots and treatment levels to sub-plots. The latter were marked 1.9 m row sections that received the following treatments: (1) control, (2) 20% square removal just prior to blooming, (3) 40% square removal, and (4) 3 late-season insecticide applications when square infestation reached 25%. There were 3 replications; plot widths and lengths were the same as in 1975. Blooms were tagged daily, as in 1975, in the 1.9 m sections.

In both years, determinations of % weevil punctured squares were made at intervals through the growing seasons. The data relative to fruiting performance were analyzed using analysis of variance; % weevil injury to locks was subjected to the non-parametric sign test (Ott 1977).

In 1975, comparisons were made between % weevil-damaged locks measured in bolls arising from different days of the flowering period of TAMCOT SP-37 and an average of the experimental genotypes 6M-10 and 1209L. Strains 6M-10 and 1209L possess relatively large bolls with thick carpel walls, while the bolls of TAMCOT SP-37 are smaller and their carpel walls somewhat thinner. Comparisons were not made using % damage to bolls, since a cotton boll can have either 4 or 5 locks. In 1978, similar comparisons were made between TAMCOT SP-37 and an average of the strains 1209L, 1209-619, and 77X3840. The latter two cottons have bolls similar to 1209L.

Comparisons of % damaged locks were accomplished by pooling the data from the sub sub-plots. For example, the bolls of TAMCOT SP-37 produced in all sub sub-plots of a given year were considered as a whole: the % damage to locks arising from the first day of blooming was calculated, the second day.... and

so on. These percentages then were compared to the percentages that were developed from the pooled injured lock data on the other genotypes. We chose this system as it provided a relatively large number of bolls for examination — over 3000 were assayed in 1978 for TAMCOT SP-37 and over 7000 in the pooled genotypes. This large sample permitted accommodation to the variability of weevil injury that would have spelled difficulties in a smaller sample of perhaps 200-300 bolls. For others pursuing this line of investigation we suggest that it is important to tag sufficient blooms so that 1000 or more bolls can be examined.

RESULTS

Weevil-injury to squares in the 1975 test did not reach a high % damage level until nearly the 30th day of the blooming period where 3 overwintered treatments were made; the infestation increased at a somewhat faster rate where only a single application was applied (Table 1). The fruiting measurements in Table 2, which are averages of the 1-treatment and 3-treatment main plots, showed that TAMCOT SP-37 bloomed at a more rapid rate (a consequence of greater square production) and set more of these as bolls.

TABLE 1. Average % Punctured Squares in all Cotton Genotypes in Tests at Uvalde, TX During 1975 and 1978^{a/}.

Sample date and number of treatments for overwintering weevils				
1975	1	3	1978	3
May 28	2	1	May 17	0.4
June 4	4	1	May 23	0.1
June 13	4	3	May 26	0.1
June 18	8	5	May 31	0.3
June 24	30	10	June 6	0.1
July 2	71	51	June 9	1.1
July 8	74	56	June 12	4.0
July 15	81	75	June 15	10.0
			June 22	9.0
			June 27	18.0
			July 4	86.0

^{a/} First blooms appeared in 1975 on June 4; in 1978, May 24.

Percent lock damage from weevils was consistently lower in TAMCOT SP-37 from bolls that arose from the first 2 weeks of flowering ($P=0.99$) than in the bolls of the other two genotypes (Figure 1). However, analysis of variance ($P=0.05$) showed that TAMCOT SP-37 experienced about the same level of lock damage per ha as the other cottons. Although % damage to locks was less in TAMCOT SP-37, it produced more total locks per ha — the % damage, then, was applied against a larger figure, and the absolute level of damage was roughly equivalent to the damage to the other cottons.

In 1978, the weevil infestation was slower to develop, and damage to bolls was less (Figure 2 and Table 1). Blooming (and squaring) was similar in all genotypes, but, as in 1975, TAMCOT SP-37 retained more blooms as bolls earlier (Table 2). Again, a lower % weevil-injury to locks ($P=0.99$) was recorded in TAMCOT SP-37. However, as in 1975, lock injury per ha in TAMCOT SP-37 was not different from the other cottons.

TABLE 2. Accumulated Blooms on Different Days of the Blooming Period; Number of Harvested Bolls that Arose from Different Flowering Periods; and Number of Bolls, 10 Days or Older, that Arose from Different Flowering Periods, Uvalde, 1975 and 1978^{a/}.

<u>Blooms, 1975^{b/}</u>				
<u>Genotype</u>	<u>Day 12 of blooming</u>	<u>Day 18 of blooming</u>	<u>Day 24 of blooming</u>	<u>Day 31 of blooming</u>
6M-10	103 a	301 a	563 a	803 a
1209L	131 a	400 b	753 b	1030 b
TAMCOT SP-37	143 a	467 b	872 c	1277 c
<u>Bolls, 1975^{b/}</u>				
<u>Genotype</u>	<u>June 6-11</u>	<u>June 12-17</u>	<u>June 18-23</u>	<u>June 24-29</u>
6M-10	17 a	67 a	141 a	106 a
1209L	37 a	101 ab	210 a	141 ab
TAMCOT SP-37	47 a	146 b	249 a	165 b
<u>Blooms, 1978^{b/}</u>				
<u>Genotype</u>	<u>Day 7 of blooming</u>	<u>Day 14 of blooming</u>	<u>Day 28 of blooming</u>	
1209L	79 a	227 a	766 a	
1298-619	72 a	222 a	724 a	
77X3840	84 a	244 a	758 a	
TAMCOT SP-37	62 a	225 a	714 a	
<u>Bolls 10 days or older, 1978^{c/}</u>				
<u>Genotype</u>	<u>Day 20 of blooming</u>	<u>Day 25 of blooming</u>	<u>Day 30 of blooming</u>	
1209L	277	343	425	
1209-619	217	281	343	
77X3840	272	348	403	
TAMCOT SP-37	289	380	484	

^{a/} All numbers are in 1000s/ha.

^{b/} Means are compared vertically; those sharing a common letter are not significantly different ($P=0.05$).

^{c/} Data not statistically analyzed.

DISCUSSION

These findings establish that TAMCOT SP-37, experiencing the same level of boll weevil infestation as some other genotypes, suffered a decreased % weevil-injury to locks that arose from the first 2-3 weeks of blooming. The tendency to set bolls earlier and in greater numbers could account for the decrease — the large numbers of bolls simply may "dilute" the damage potential of a given number of weevils. If this is the case, any cotton that acquires a complement of bolls quickly in the fruiting period would enjoy the same advantage.

However, we suggest that there may be an additional element contributing to the lowered % injury. The bolls of TAMCOT SP-37 appear to physiologically age at a somewhat more rapid rate than bolls of many of the other cottons we have examined. That is, an 8-day-old boll or a 10-day-old boll of TAMCOT SP-37

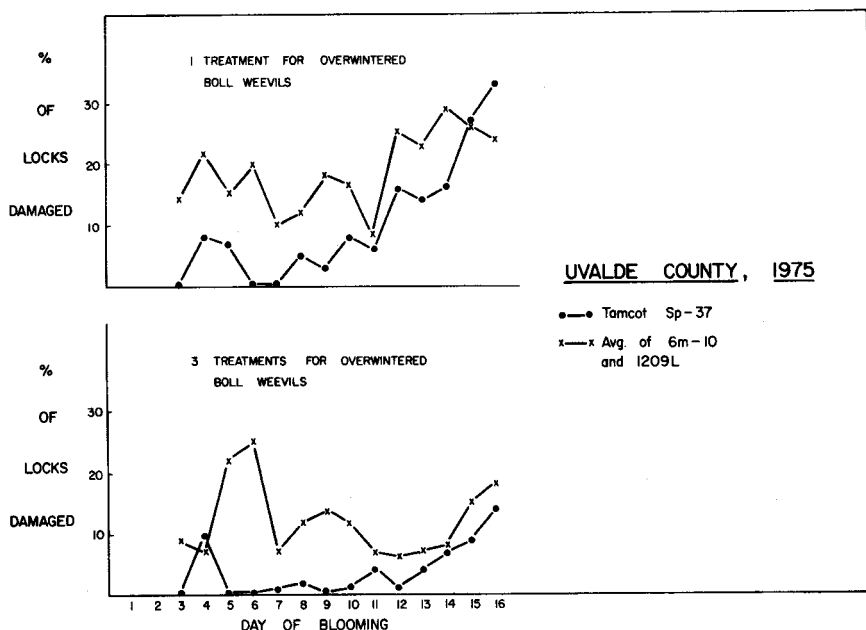


FIG. 1. Percent weevil damage to locks produced at different days of the flowering period, Uvalde, 1975.

may be, in fact, "older" than an 8- or 10-day-old boll of 1209 or 6M-10. If this is the case, considering the findings of Parker et al. (1980), the accelerated maturity might grant a slight advantage in escaping weevil-injury.

In any case, the overall findings of this study reaffirm the value of the rapid accumulation of bolls as a means of producing cotton in the presence of the boll weevil.

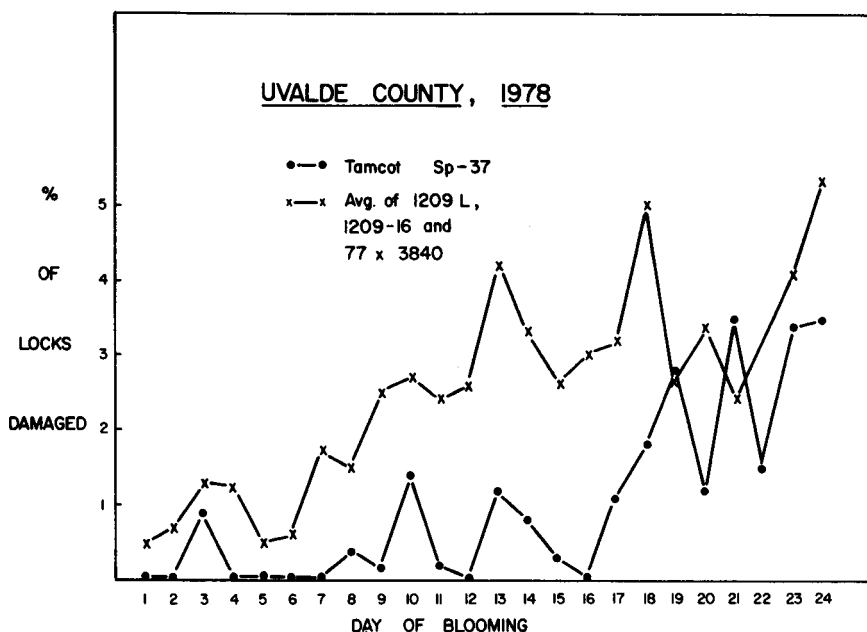


FIG. 2. Percent weevil damage to locks at different days of the flowering period, Uvalde, 1978.

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DEAD-END PARASITISM OF BOMBYLIID LARVAE ^{1/}
IN TABANID ADULTS ^{2/}

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ABSTRACT

During examination of adult tabanids for the parasitic larvae of miltogrammine sarcophagids (*Macronychia* sp.), the larvae of a brachycerous dipteran, believed to represent Bombyliidae, were also discovered in the same host species, *Hybomitra lasiophthalma* (Macquart). Of 871 females taken in CO₂-baited Manitoba Traps from April 1 through May 9 on the Navasota River in south-central Texas, 3.8% were infested with 1-7 larvae per host (avg. 1.4). Most (91.3%) of the 46 infesting larvae were removed from the abdomen where they were found free near the terminal segments. Their size and encapsulation suggest the larvae died soon after penetration of the host. However, the host-parasite relationship is significant because it points out the apparent attractiveness and susceptibility of horse flies as hosts to parasitic insects.

INTRODUCTION

The primary parasitism of Diptera by other dipterous larvae is unusual and has involved only several species of Bombyliidae, Sarcophagidae, and Tachinidae. Several bombyliid species have been isolated from puparia of Glossinidae and Calliphoridae (Glasgow 1963). Rarer instances of adult Diptera being attacked by dipterous parasites, such as the instance reported here, were described by Spratt and Wolf (1972), Smith (1974), Ferrar (1977), Thompson (1978a, 1978b) and Thompson and Love (1979). The latter description is particularly relevant to the present findings because the same tabanid population was simultaneously infested by 2 different kinds of dipterous parasites - a sarcophagid and a bombyliid. The latter host-parasite association has not been reported heretofore.

METHODS

A population of *Hybomitra lasiophthalma* (Macquart) inhabiting a floodplain forest of the Navasota River 12 mi east of Bryan, Texas was sampled daily from March 14 through May 12 using 1 modified Gressitt Trap and 8 modified Manitoba Traps. The physiography and vegetation of this basin were described previously (Thompson 1977). Further details concerning handling and maintenance of the catch in the laboratory were previously described also (Thompson, Hogan, and Peterson 1980). The subjects of study were dying females which were removed from cages twice daily and then frozen until they were examined. From March 29 through May 12, there were 12,713 females of *H. lasiophthalma* collected live from the Navasota study area (Channey Crossing) and returned to the laboratory. A sample of 25 females, when available, was examined internally for each collection date throughout their period of activity. Each specimen was impaled against a wax-filled watch glass, after which the thoracic and abdominal cavities were exposed by means of mid-ventral incisions.

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- ^{1/} Diptera: Bombyliidae.
^{2/} Diptera: Tabanidae.

RESULTS

Populations of *H. lasiophthalma* peaked in mid-April and samples taken from April 1 through May 10 were infested with a brachycerous dipteran believed to represent Bombyliidae. Because of the very small size of the larvae (1.5mm), Mr. George C. Steyskal, SEL, USDA, determined that they represent 1 of 3 families (Bombyliidae, Therevidae, or Asilidae). Endoparasitic forms are unknown from the latter 2 families; therefore, the parasite probably represents Bombyliidae, a family containing many parasitic species.

Of 871 horse fly females collected from April 1 through May 10, 33 were infested, causing a parasitism rate of 3.8%. The first infested fly was taken April 1 and the daily rate fluctuated somewhat through the remaining period of fly activity in spring. The number of bombyliid larvae per host ranged from 1 to 7 with an average of 2.7 parasites per host. Of 46 larvae removed from tabanid females, 91.3% were found in the thoracic cavity.

All parasites were found free within the thoracic or abdominal cavities of the hosts. The condition of these parasites suggests that they died soon after penetration of the host because their size was uniformly small and the integument was heavily pigmented and covered with extraneous material (Fig. 1). When the end of this dark brown capsule was gently removed, the larva within it could be seen (Figs. 2 and 3); and also, the definitive character of the exuvium.

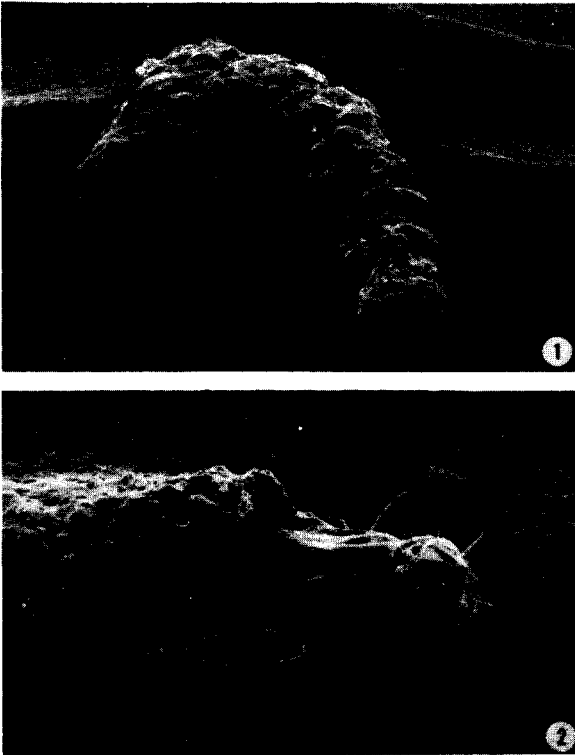


FIG. 1. Scanning electron micrograph of an encapsulated larva, X138.

FIG. 2. Micrograph showing a portion of the capsule broken away to reveal a ventro-lateral view of the head and thorax of the larva, X265.



FIG. 3. Higher Magnification of the Ventral Surface of the Head to Show the Mouthparts of the Larva, X657.

Although the host-parasite association between the tabanid and bombyliid was unsuccessful for the parasite, the association between H. lasiophthalma and Macronychia, on the other hand, was successful for the parasite. This sarcophagid completed development on dead host material within the laboratory. In addition, the 118 larvae observed in dissected specimens during 1978 were either living upon observation, or were apparently in that condition when they were frozen inside the host.

Other similarities between the bombyliid and the sarcophagid associations are of interest; both parasites infested the host to a comparable degree (3.8% and 4.6%, respectively), and both did so throughout most of the season of tabanid activity. Parasite loads ranged from 1-7 larvae per host for both forms; and both forms infested the thorax and the abdomen, although Macronychia predominated in the thorax, the most reliable site of tissue nutrients in horse flies at any given time (especially for this, and other anautogenous species). Instances of simultaneous parasitism by both parasites were rare; of 22 bombyliid infestations and 16 of Macronychia, only 1 instance involved larvae of the 2 forms.

DISCUSSION AND CONCLUSIONS

The susceptibility of horse flies to infestation by entomophagous insects could have much to do with their habit of pupating near the soil surface. Teskey (1969) reared adults of a bombyliid (Villa lateralis Say) from immature specimens of H. lasiophthalma and H. typhus (Whitney), but both of the host specimens - 1 larva and 1 pupa - had probably been infested as larvae. Although pupae of H. lasiophthalma have been taken in the field by Cameron (1926), Davies and LaIT (1970), Philip (1931), and Roberts and Dicke (1964), the specific niche of these specimens in the substratum was not indicated. (Several other authors have reported the presence of pupae under tree bark or within rotten logs.)

The host-parasite association described here is apparently an unsuccessful one for the parasite. On the other hand, the not uncommon frequency of this association - as that involving tabanid and sarcophagid - suggests that the host is particularly attractive and vulnerable to attack by parasitic insects.

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THE RELATIONSHIP OF SOLENOPSIS INVICTA BUREN^{1/} TO SOILS OF EAST TEXAS^{2/}

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ABSTRACT

Red imported fire ant (Solenopsis invicta Buren) colonies over a 19 county area of Texas were found in 9 textural classes of soil. Thirty-six percent of the mounds had soil textures differing from texture of adjacent soil. There were, however, no significant differences in pH, % sand, silt, clay, or organic matter in mound soil versus soil adjacent to mounds.

RESUMEN

Colonias de la "hormiga de fuego roja importada" (Solenopsis invicta Buren) colectadas en 19 condados de Tejas ocuparon suelos de nueve texturas. Treinta y seis por ciento de los nidos presentaron texturas diferentes a las del suelo vecino. No se hallaron diferencias apreciables en pH, % arena, arcilla, barro o materia orgánica entre los suelos de los nidos y las áreas vecinas.

INTRODUCTION

The red imported fire ant, Solenopsis invicta Buren, has become well established in the southeastern U.S. since its accidental introduction at Mobile, Alabama about 39 years ago (Lofgren et al., 1975). Its current range includes every southern state from Florida to Texas. The northern boundary has remained relatively stable for several years and its expansion now appears to be westward. It is generally agreed that the western movement will progress but the rate at which the species spreads and the areas of the west and southwest it eventually occupies is still speculative.

Studies are currently underway at Texas Tech University to define the relationship between the red imported fire ant and its changing environment. A preliminary model (Pimm and Bartell, unpublished data)^{3/} has been designed to explain the current geographical distribution on the basis of biotic and abiotic factors. It is hoped that the model will be useful as a tool for predicting future spread. To this end, extensive data on those factors believed to be most important to fire ant survival are being gathered for incorporation in the model. Among these are the possible influence of soil characteristics on fire ant distribution. The influence of soil on colony founding is ill defined (Lofgren et al., 1975). It is a common observation that S. invicta can infest a wide range of soils but there are no published records of the type of soils infested. Furthermore, it is not known what effect, if any, the ant has on physical characteristics of the soil as a result of mound construction. Therefore, this study was initiated to investigate the relationship of S. invicta to soil in East Texas. The specific objectives were to identify soil types infested by S. invicta and to compare physical properties of soil comprising S. invicta mounds versus undisturb-

^{1/} Hymenoptera: Formicidae

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ed soil. This research was conducted as a first step in answering broader questions about soil as a potential limiting factor to the spread of S. invicta. Before we can determine if S. invicta is excluded from certain areas because of soil or if S. invicta shows a preference to certain soil types we must have knowledge of the soils presently infested.

METHODS AND MATERIALS

One hundred soil samples were taken over a wide area of East Texas, comprising 19 counties, with moderate to heavy infestations of S. invicta Buren. An effort was made to sample as many textural classes of soils as possible. At each site, a mature, active mound was selected. One soil sample was taken from the center of the mound with a standard 1.5 m soil auger. A second sample was taken from soil ca. 2 m away from the mound and apart from the ant colony. All samples extended to ca. 20 cm in depth. Soil samples were bagged and later (at the soil analysis laboratory, Texas Tech University) analyzed for the following: 1) % organic matter, 2) pH, 3) % sand, 4) % silt, 5) % clay, 6) textural class. Ants collected with the mound soil died in the bags and according to soil scientists had no effect on the results of analysis.

An analysis of variance was calculated for these data to identify any significant differences in the physical characteristics of mound soil versus soil adjacent to the mounds. Duncan's New Multiple Range Test was used to separate the means.

RESULTS AND DISCUSSION

Fire ant colonies were found in nine textural classes of soil: sand, sandy-loam, sandy-clay, sandy-clay-loam, silty-clay-loam, loam, loamy-sand, clay-loam, and clay.

Thirty-six percent of the mounds observed had soil textures differing from adjacent soil (Table 1). In most instances the textural components of mound soil showed some overlap with those of adjacent soil. Ants either altered the percent sand, silt, clay or loam enough to affect the textural class or they added or removed soil components as soil was reorganized during mound construction. In only one instance was mound soil lacking all of the components of adjacent soil.

TABLE 1. Observed Differences in Soil Texture in Mounds Versus Soil Adjacent to Mounds.

No. Observations	Textural Class ^{α/}	
	Soil Adjacent to Mound	Mound Soil
1	C	SL
2	CL	C
1	CL	L
1	CL	SiCL
4	SL	SCL
1	SC	SL
1	SCL	CL
2	SiCL	CL
1	L	SL
1	L	SCL
2	LS	SL
1	LS	SCL

^{α/} C = clay, S = sand, Si = silt, L = loam.

The explanation for both of these phenomena appears simple. Solenopsis invicta must gather soil from varying depths for construction of mounds and consequently, depending on the soil profile, may incorporate different textural components which occur at greater depths. The occurrence of sand-loam mound soil in an area of clay soils, for instance, would indicate that the lower soil horizons were excavated for mound construction. Samples of adjacent soils represented ca. the top 20 cm of the soil profile, therefore S. invicta will commonly go beyond this depth to reorganize soil. It is as yet undetermined if S. invicta is selective for certain soil textures.

In spite of the fact that overall textural class of soil may be altered there was no significant difference in the individual physical properties of the soil sampled from mounds versus areas adjacent to mounds (Table 2).

TABLE 2. Comparison of Physical Properties of Mound Soil Versus Soil Adjacent to Mounds.

Soil Property	Mound (M) Versus Soil Adj. to Mound (AM)	Mean Values ^{α/}
% Sand	M	57.54 a
% Sand	AM	60.67 a
% Silt	M	23.23 a
% Silt	AM	23.39 a
% Clay	M	15.93 a
% Clay	AM	19.13 a
% Organic Material	M	1.46 a
% Organic Material	AM	1.57 a
pH	M	6.06 a
pH	AM	6.28 a

^{α/} Number of observations in each data set = 100; means of each data set with common letter not significantly different at 5% level.

These data confirm casual observations that S. invicta is capable of exploiting a wide variety of soil types. It is still premature to say that soils will not be an important limiting factor in its westward spread. However, in view of the fact that many textural classes of soils have been infested by S. invicta in east Texas, it appears that this soil character alone may be of minor importance.

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APPARATUS FOR STICKY TRAP WASHING AND INSECT RECOVERY^{1/}C. L. Green^{2/}, F. A. McCarty^{2/}, L. J. Edson^{3/}, T. L. Payne

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ABSTRACT

A device is described which facilitates cleaning insects from sticky traps. The trap washer is composed of a stainless steel trough, hollow Cone Jet T nozzles, a centrifugal pump and associated plumbing. A standard water heater is an integral part of the system for use in heating Varso[®] as a cleaning agent.

INTRODUCTION

Sticky traps are commonly used for field studies of insects. Several problems arise from using these traps, such as removing the trapped insects and cleaning the traps for recycling. Removing individual insects from the traps is often time consuming, and accurate counts can be affected by debris clinging to the traps. Small, soft-bodied insects can be easily overlooked. In addition, hand washing large numbers of traps for recycling is time consuming. This report describes a trap-washing apparatus that was developed to help remedy these problems.

METHODS AND MATERIALS

A schematic of the assembled equipment is shown in Figure 1. The apparatus included a 75.71l electric water heater (a) fitted with a 200psi pressure valve for safety purposes. Power for the system was provided by a 1/3 hp Dayton[®] motor (b) which drove a Twell[®] centrifugal pump (c) via a 25.4cm V-grooved wheel (d) and a 1.27 x 45.72cm belt. The heater, motor, and pump were secured to a plywood platform. Since hot solvent (Varso[®]) was used in the trap washer, oil-resistant flexible vacuum hose was required; this was

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held in place with Ideal[®] hose clamps. Four 1.91cm shut-off valves were spliced into the line at various places to regulate the flow of the cleaning agent. A 1.91cm tee and two 1.91cm 90° elbow pipe fittings were incorporated into the line to split the solvent stream and direct it to the spray jets.

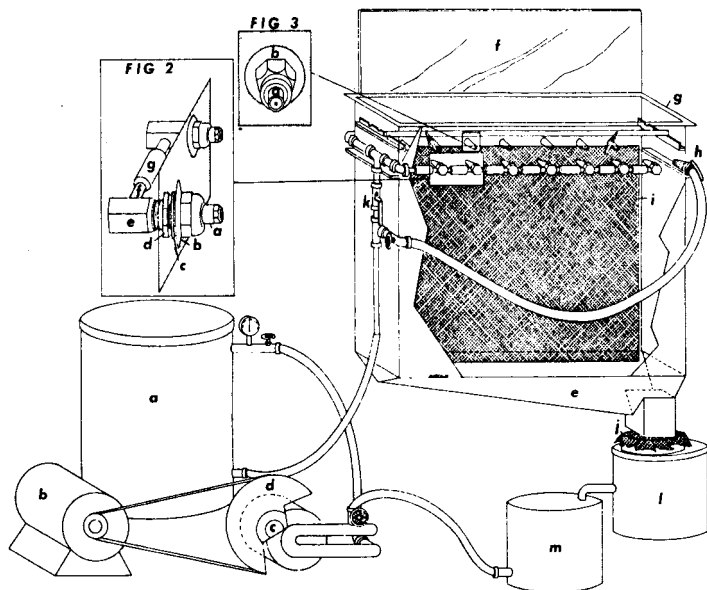


FIG 1 -- Trap washer: (a) electric water heater; (b) 1/3hp Dayton[®] motor; (c) centrifugal pump; (d) wheel; (e) stainless steel trough; (f) lid; (g) weatherstripping; (h) spray nozzle; (i) fiberglass wingtrap vane; (j) organdy cloth filter; (k) shut-off valve; (l) settling bucket; (m) filter and suction inlet tank.

FIG 2 -- Cone Jet spray nozzle assembly: (a) Cone Jet type 1/4T hollow cone orifice spray tip; (b) rubber gasket; (c) metal washer; (d) 12.7mm nut; (e) Delvan 6.35mm brass NPT female pipe connection; (f) 6.35mm x 5.08mm galvanized nipple; (g) 12.7mm oil-resistant hose.

FIG 3 -- Front view of spray nozzle: (a) hollow Cone Jet T nozzle; (b) metal washer.

Figures 2 and 3 illustrate the spray nozzle assembly. Fourteen Cone Jet type 1/4T spray nozzles with hollow cone orifice tips were equally spaced at 11.43cm centers on opposite sides of a trough (e) measuring 91.5 x 76 x 28.6cm (length x height x width) constructed of .063 gauge stainless steel. The spray nozzles were arranged with seven on each side of the trough to facilitate thorough cleaning of the trap on both sides. The sides of the trough gradually sloped at the bottom, leading to a 10.16 x 10.16cm opening. All seams were soldered to prevent leakage. Plexiglass® measuring 0.64 x 31.75 x 99cm (thickness x width x length) served as a lid (f) for the washer. Felt weatherstripping (g) was used between the lid and the trough for a waterproof seal. Metal washers were screwed against all rubber gaskets with 12.7mm brass nuts to prevent leakage around the nozzles. Delvar® 6.35mm brass NPT (National Pipe Thread) female pipe connections were used to hold the spray nozzles in position. The solvent stream was directed to each nozzle through 28 6.35mm x 5.08mm galvanized pipe nipples and 12.7mm oil-resistant hose spliced together with 12.7mm Ideal hose clamps.

An auxiliary spray line for use in manual cleaning was constructed by attaching a Nibco® 15.87mm shut-off valve (k) to a 1.91cm galvanized tee fitting. A 1.91cm to 2.54cm bushing was screwed into the valve to adapt for the 2m of 12.7mm oil-resistant hose which was joined into the line. A Hose Master Supreme® spray nozzle (h) was used for controlled spraying. The effluent from the washer passed through organdy cloth (to collect small insects) to a series of two 11.36l buckets that served as settling (l) and suction (m) tanks.

TRAP WASHER OPERATION

A respirator was used to protect the operator from solvent fumes. The solvent was heated to 70°C, and the centrifugal pump was activated. The pump was placed below the feed reservoir liquid level so that the pump head was flooded and would stay in prime. As the solvent was pumped, the series of shut-off valves regulated pressure and flow. Once the solvent had reached these valves, it could be directed either to the 14 spray nozzles or to the auxiliary spray line. Traps to be cleaned were placed in the stainless steel trough. A wood dowel with alligator clips was used to hold each trap in position. The cleaning tank was equipped with slots for the dowels. The hot solvent sprayed over the traps dissolved the sticky material. Usually two minutes were sufficient to clean a 61 x 61cm fiberglass sticky trap. As the insects and debris were rinsed from the trap, they passed through the opening at the bottom of the trough and were filtered from the solution by the organdy cloth placed over the settling bucket. The insects were separated from the debris and placed in vials for later identification.

Used solvent passed through the filter into the settling bucket. When it reached a certain level in the settling tank, the solution would flow into the filter and suction bucket. The solvent was then recycled to the heater and used again until it became slightly viscous.

ACKNOWLEDGMENT

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COMPARATIVE POPULATIONS OF BENEFICIAL ARTHROPODS AND
HELIOTHIS SPP. LARVAE IN SELECTED FIELDS IN PANOLA AND PONTOTOC
COUNTIES, MISSISSIPPI IN 1977 AND 1978 ^{1/}

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ABSTRACT

Cotton fields were monitored for pest insects and beneficial arthropods weekly in 1977 and semi-weekly in 1978 in Panola (20 fields) and Pontotoc counties (10 fields), Mississippi. Populations of important beneficial arthropods were high in 1977 and moderate in 1978 peaking at averages of 32,000 and 22,000 per acre in Panola County and at 41,000 and 26,000 per acre in Pontotoc County, respectively. Geocoris punctipes (Say) was the most common predator followed by a flower bug, Orius insidiosus (Say); the convergent lady beetle, Hippodamia convergens Guérin-Ménéville; Coleomegilla maculata (De Geer); the common green lacewing, Chrysopa carnea Stephens; a damsel bug, Reduviolus roseipennis Reuter; and a lynx spider, Oxyopes salticus Hentz. Use of insecticides for control of the tarnished plant bug, Lygus lineolaris (Palisot de Beauvois), in early to mid-July reduced the beneficial populations and was conducive to subsequent development of damaging populations of Heliothis spp. larvae later in the season. Treatment for control of thrips, Frankliniella spp., either with conventional insecticides or with in-furrow at-planting applications of a systemic insecticide (aldicarb), had little influence on Heliothis spp. populations when the beneficial populations were high in 1977, but more insecticide applications were needed for control of Heliothis spp. larvae following in-furrow applications of aldicarb when these populations were moderate in 1978.

INTRODUCTION

The Optimum Pest Management Trial (OPM) in Panola County, Mississippi, which is being conducted concurrently with the Boll Weevil Eradication Trial in North Carolina and Virginia, began in 1978. The objective is to develop data that will make it possible to evaluate the biological, economic and environmental impacts of the two programs if they are used across the Cotton Belt.

A prototype operation was conducted in 1977 in Panola and Pontotoc Counties to develop baseline data, to evaluate and refine sampling, recording and data storage techniques, and to develop procedures that would permit smooth expansion of the program when the OPM Trial got underway in 1978. Cotton fields in Pontotoc County were used to develop similar data for a Current Insect Control (CIC) program.

Development of data by the Research Team for Biological Evaluation of the OPM Trials is affording opportunities to make comparisons and to study various relationships that would otherwise be impossible. One such comparison was made between populations of beneficial arthropods and seasonal Heliothis spp. larval populations in selected cotton fields in Panola and Pontotoc Counties, Mississippi, in 1977 and 1978. Results are reported herein.

^{1/} In cooperation with the Mississippi Agricultural and Forestry Experiment Station, Stoneville, Mississippi 38776.

METHODS AND MATERIALS

Cotton fields were monitored in 1977 for populations of pest insects and beneficial arthropods in Panola (20 fields) and Pontotoc Counties (10 fields). In 1978, the numbers of fields monitored were increased to 64 and 32, respectively. However, with few exceptions, the 20 and 10 fields monitored in 1977 were included among those monitored in 1978, therefore, these fields are here compared directly though the monitoring was weekly in 1977 and semi-weekly in 1978. Thus, Heliothis spp. larvae were counted in the terminal buds of cotton plants on 25 feet of row at 5 locations in each field; numbers in the squares and bolls were determined by examining 200 squares and 200 bolls (50 at 4 locations); and numbers in white blooms were counted on 125 feet (25 feet at 5 locations). The row feet required for the inspections were recorded, the number at the 3 sites were totaled and the populations were computed on a per acre basis.

The beneficial arthropod population was sampled with a vacuum suction machine. A total of 40 row feet (4 samples of 10 row feet each) per field were sampled weekly in 1977 and semi-weekly in 1978. The machine was equipped with a 39-cm (10 inch) cone and the plants were vacuumed from one side and from top to bottom while the operator moved along the row. Samples were placed in a freezer at the field operation base and were then brought to the laboratory at Stoneville, MS, where specimens were separated from trash by hand and identified and counted under magnification. However, only total numbers of the most important beneficial arthropods are considered in this report. Geocoris punctipes (Say) was the most common predator followed by a flower bug, Orius insidiosus (Say); the convergent lady beetle, Hippodamia convergens Guérin-Ménéville; Coleomegilla maculata (DeGeer); the common green lacewing, Chrysopa carnea Stephens; a damsel bug, Reduviolus roseipennis Reuter; and a lynx spider, Oxyopes salticus Hentz.

In 1977, yields were estimated by hand picking, once or twice as needed, four 10-foot row sections in each field; in 1978, ten 10-foot row sections in each field were hand picked.

All insecticide applications were made at the discretion of the farmer. Dicrotophos and dimethoate were used for the control of thrips and lygus bugs. Materials used for control of Heliothis spp. larvae were: EPN plus methyl parathion plus methomyl, monocrotophos, monocrotophos plus methomyl plus Bacillus thuringiensis (B.t.), methomyl plus (B.t.), methomyl plus methyl parathion, acephate, fenvalerate, permethrin, and toxaphene plus methyl parathion. No control failures were observed per se.

Regression analyses were done on the pooled data for the two counties and years to correlate density of beneficial arthropod populations with levels of Heliothis spp. larval populations and their damage. The "Statistical Analysis System" package at the Washington, D.C., Agricultural Research Computer Center was used to run the analyses. Peak values were selected from each sampled field for the following variables: Total beneficial arthropod populations; second and third generation Heliothis spp. larval populations, number of bolls, numbers of damaged bolls, and percentage of damaged bolls.

RESULTS

1977. Panola County. Fields 9 and 13 (Fig. 1) were not treated early in the season for control of thrips, Frankliniella spp., or tarnished plant bug (TPB), Lygus lineolaris (Palisot de Beauvois). Populations of beneficial arthropods peaked at about 28,000 per acre in mid-July and decreased drastically in late July. Populations of Heliothis spp. larvae peaked at 1,600 per acre in mid-August. Boll damage peaked at 2,200 on August 20 and September 3. The fields were treated an average of 2.0 times between August 10 and 17 for control of Heliothis spp. larvae. The average yield was 810 pounds of lint per acre.

Field 1 (Fig. 2) was treated once, July 7, for control of TPB. The beneficial arthropod population was high in June and peaked at 28,000 per acre on

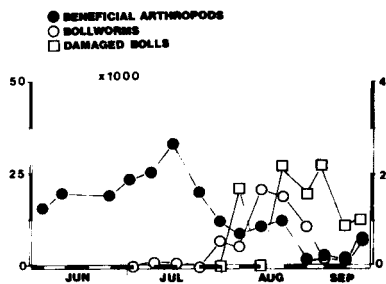


FIG. 1. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls per acre in fields 9 and 13, Panola County, MS, 1977.

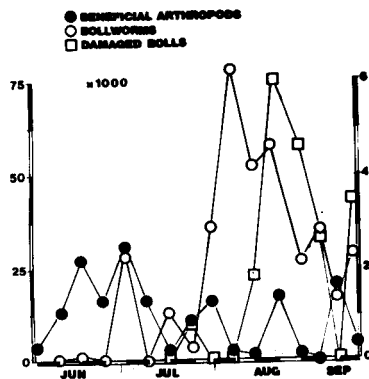


FIG. 2. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls per acre in field 1 in Panola County, MS, 1977.

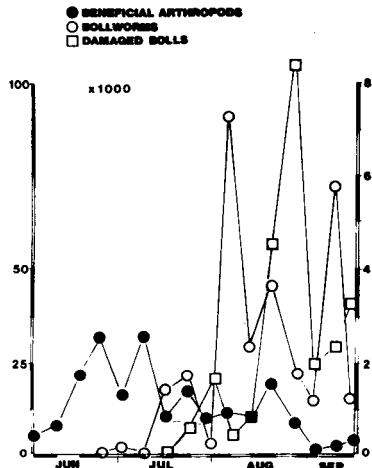


FIG. 3. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls in fields 2 and 4, Panola County, MS, 1977.

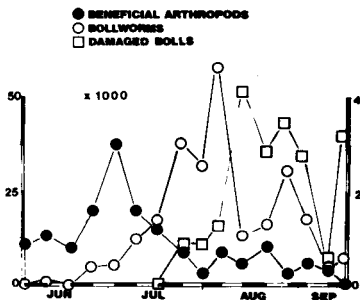


FIG. 4. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and boll damage per acre in fields 12 and 17, Panola County, MS, 1977.

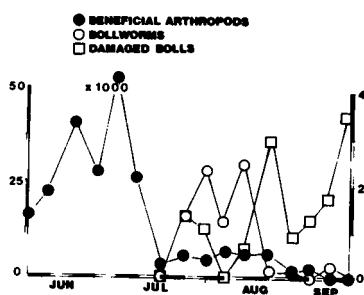


FIG. 5. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls per acre in field 19, Panola County, MS, 1977.

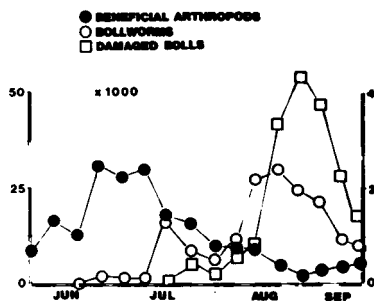


FIG. 6. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls per acre in fields 3, 5, 6, 7, 8, 10, 11, 14, 15, 16, 18 and 20 in Panola County, MS, 1977.

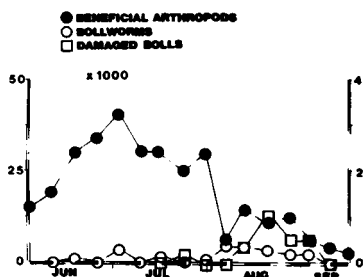


FIG. 7. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls per acre in fields 27, 28 and 30 in Pontotoc County, MS, 1977.

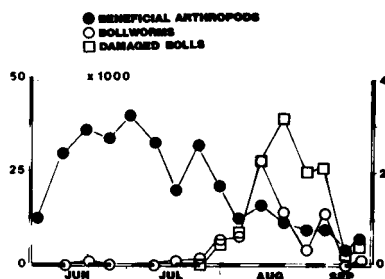


FIG. 8. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls per acre in fields 21, 26 and 29 in Pontotoc County, MS, 1977.

July 2; it decreased drastically on July 9 and 16 after the treatment for control of TPB. There was some recovery in late July. The population decreased further in August after treatment started for control of Heliothis spp. (6 insecticide applications were made between August 5 and 29). Larval populations averaged slightly more than 6,000 on August 6 and declined after treatment, though not much below the 2,000 per acre level. Damaged bolls peaked at 6,000 per acre on August 20. The yield was 645 pounds of lint per acre.

Fields 2 and 4 (Fig. 3) received early-season treatment with foliar insecticides for control of thrips and TPB. The beneficial arthropod population was high in June and peaked at 32,000 per acre on July 9. It decreased in mid-July after treatment on July 7 and 12, respectively, for control of TPB. The Heliothis spp. larval population averaged 7,200 per acre on August 6. An average of 4.0 insecticide applications were made between August 3 and September 10 for control of Heliothis spp. larvae. Average boll damage peaked at about 8,000 per acre on August 27, and yield averaged 892 pounds of lint per acre.

Fields 12 and 17 (Fig. 4) received in-furrow applications of aldicarb at planting. The initial beneficial arthropod population was high. It peaked at an average of 36,000 per acre in early July and declined to about 4,000 per acre at the end of July. Heliothis spp. populations averaged 3,000 per acre on July 30 and 4,600 per acre on Aug. 6. An average of 2.5 insecticide applications were made for control of Heliothis spp. larvae between August 9 and 31. Average boll damage peaked at 4,000 per acre on August 13. Yield averaged 895 pounds of lint per acre.

Field 19 (Fig. 5) received an in-furrow application of aldicarb at planting and a foliar treatment for TPB control on July 23. The beneficial arthropod population was high initially. It peaked at about 50,000 per acre in early July and decreased drastically in mid-July. Average larval population of Heliothis spp. peaked at 2,400 per acre on August 13. Six insecticidal applications were made between August 9 and September 9 for control of Heliothis spp. larvae. Peak boll damage was 3,000 on August 20 and 3,400 per acre on September 17. The yield was 892 pounds of lint per acre.

The remaining 12 fields (Fig. 6) were treated with foliar insecticides early in the season for control of thrips; they were not treated for control of TPB. In these, the beneficial arthropod population peaked at about 31,000 per acre in late June and declined by the end of July to about 9,000. An average of 3.8 insecticide applications were made from July 17 to August 20 for control of Heliothis spp. larvae. The Heliothis spp. larval population averaged slightly more than 2,000 per acre on August 13 and 20. Average peak boll damage was slightly more than 4,000 per acre on August 27. The average yield was 765 pounds of lint per acre.

The results in Panola County in 1977 therefore suggest, as would be expected (though field numbers were small), that applications of insecticide for control of TPB in early or mid-July are conducive to subsequent attacks by Heliothis spp. larvae. Three such fields received an average of 4.7 applications of insecticides for control of Heliothis spp. and the estimated yield averaged 810 pounds of lint per acre. In the 2 fields where no insecticides were applied for control of thrips or TPB, the beneficial arthropod population was high peaking at an average of 28,000 per acre, and an average of 2.5 applications of insecticide were made in late season for control of Heliothis spp. The yield averaged 810 pounds of lint per acre. The 12 fields receiving foliar treatment for thrips control were treated an average of 3.8 times for control of Heliothis spp.; the average yield was 765 pounds of lint per acre. The 2 fields receiving aldicarb in-furrow at planting were treated with insecticides an average of 3.5 times for control of Heliothis spp.; the average yield was 895 pounds of lint per acre. The one field that received aldicarb in-furrow at planting and a foliar insecticide for control of TPB was treated an average of 6 times for control of Heliothis spp.; the yield was 892 pounds of lint per acre.

Pontotoc County. Fields 27, 28, and 30 (Fig. 7) were not treated with

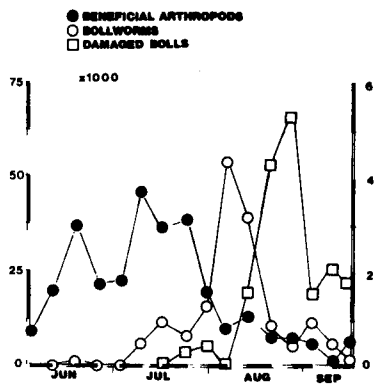


FIG. 9. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls per acre in fields 22, 24, and 25 in Pontotoc County, MS, 1977.

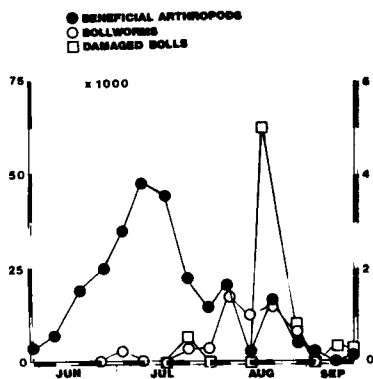


FIG. 10. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls per acre in field 23 in Pontotoc County, MS, 1977.

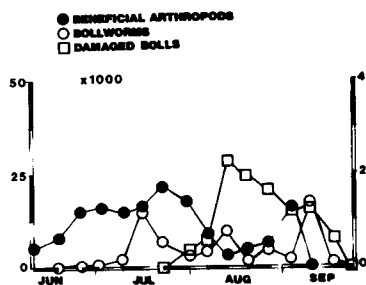


FIG. 11. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls in fields 3, 4 and 11 in Panola County, MS, 1978.

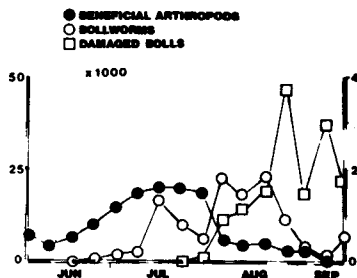


FIG. 12. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls per acre in fields 1, 2, 16 and 20 in Panola County, MS, 1978.

insecticides. The beneficial arthropod population peaked at 40,000 per acre on July 2, and then it began to decline. It remained in the 10,000 to 15,000 range during August. The average Heliothis spp. larval population slightly exceeded 300 per acre on August 6 and 13. Average boll damage peaked at 1,000 per acre on August 20. The estimated average yield was 730 pounds of lint per acre.

Fields 21, 26 and 29 (Fig. 8) received no insecticide applications for control of thrips or TPB but averaged 2.3 insecticide applications between August 2 and 30 for control of Heliothis spp. The initial beneficial arthropod population ranged from 36,000 to 41,000 from mid-June to mid-July. It decreased to 8,500 by early September. The Heliothis spp. larval population peaked at 2,200 on August 13 and boll damage peaked at 3,100 on August 20. The estimated average yield was 597 pounds of lint per acre.

Fields 22, 24, and 25 (Fig. 9) received an application of a foliar insecticide for control of thrips but were not treated for control of TPB. The initial beneficial arthropod population peaked at 41,000 per acre in early July. It declined gradually to 6,000 per acre by August 20. The average Heliothis spp. larval population peaked at 4,300 and 3,100 on August 13 and 20, and average damaged bolls peaked at 5,200 on August 27. An average of 2.3 insecticide applications were made between August 2 and 23 for control of Heliothis spp. The average yield was 623 pounds of lint per acre.

Field 23 (Fig. 10) received an in-furrow at-planting application of aldicarb. The initial population of beneficial arthropods was low but increased rapidly and peaked at 45,000 per acre on July 9. It declined to about 13,000 on July 30, and then decreased steadily for the rest of the season. The Heliothis spp. larval population peaked at 1,400 per acre on August 6 and boll damage peaked at about 5,000 per acre on August 20. The field was treated on August 1 and 6 for control of Heliothis spp. larvae. The estimated yield was 740 pounds of lint per acre.

Three of the 10 fields were not treated for control of any insect pest. The seven treated fields were treated an average of 2.2 times. Three of the treated fields received no treatment for control of thrips or lygus bugs; three were treated with foliar insecticides for control of thrips; and one field received an in-furrow application of aldicarb. Yields in untreated fields averaged 730 pounds of lint per acre compared with 630 pounds in the treated fields. Thus, in Pontotoc County in 1977 when conditions were favorable for a high beneficial arthropod population (early spring with high host plant populations in predator reservoir areas), treatments for thrips control did not result subsequently in an increased Heliothis spp. population.

1978. Panola County. Fields 3, 4, and 11 (Fig. 11) were not treated for control of early-season insects. The initial beneficial arthropod population was low and peaked at about 20,000 per acre in late July. It declined drastically by mid-August though there was some resurgence in early September. The Heliothis spp. larval population was low and averaged about 1,200 and 1,400 per acre on July 15 and Sept. 9, respectively. Average boll damage peaked at about 2,000 per acre in mid-August. The fields were treated with insecticides an average of 2.0 times between August 2 and 15 for control of Heliothis spp. larvae. The average yield was 510 pounds of lint per acre.

Fields 1, 2, 16, and 20 (Fig. 12) were treated with foliar applications of insecticides for thrips control. The initial beneficial arthropod population was low but peaked at about 20,000 on July 29. It declined sharply by mid-August. The Heliothis spp. larval population was low and averaged about 1,800 per acre on August 5 and 19. Average boll damage peaked at 3,600 per acre on August 26. The fields were treated an average of 2.5 times between August 5 and 24 for control of Heliothis spp. larvae. The estimated average yield was 705 pounds of lint per acre.

Fields 5, 6, 7, 8, 9, 10, 14, and 18 (Fig. 13) were treated with foliar insecticides for thrips control but were not treated for control of Heliothis spp. larvae. The initial beneficial arthropod population was higher than in most fields, peaked at about 22,000 per acre on July 29, and declined somewhat in

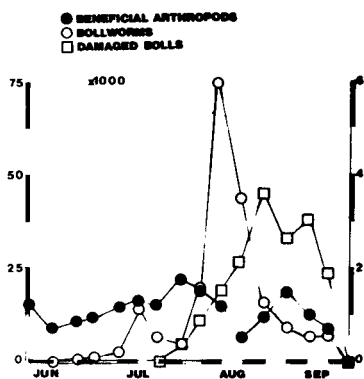


FIG. 13. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls per acre in fields 5, 6, 7, 8, 9, 10, 14 and 18 in Panola County, MS, 1978.

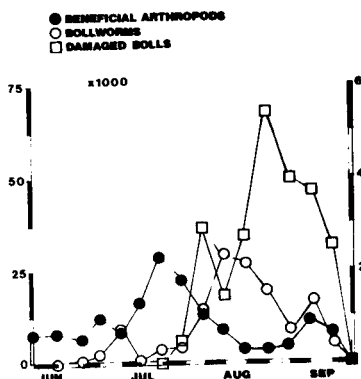


FIG. 14. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls per acre in fields 12, 13, 15 and 17 in Panola County, MS, 1978.

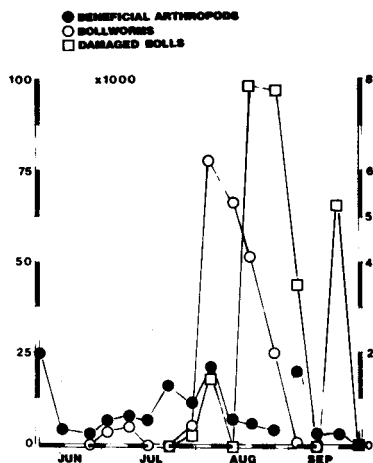


FIG. 15. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls in field 19 in Panola County, MS, 1978.

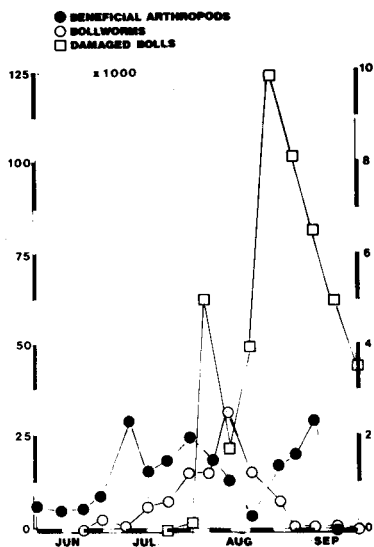


FIG. 16. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls per acre in fields 25, 26, 27, 28 and 30 in Pontotoc County, MS, 1978.

mid-August; there was a subsequent upsurge in late August. The Heliothis spp. larval population peaked at about 6,000 per acre on August 12. Average damaged bolls peaked at about 3,600 per acre on August 26. The average yield was 652 pounds of lint per acre.

Fields 12, 13, 15, and 17 (Fig. 14) received aldicarb in-furrow at planting. The initial beneficial arthropod population was low and peaked at 29,000 per acre in late July; it declined by mid-August. A light Heliothis spp. population began in early July, and average peaks of 2,300 and 2,200 occurred on August 12 and 19. Boll damage was higher than in most fields; the averages were 5,400, 4,000 and 3,700 per acre on August 26 and September 2 and 9. The fields received an average of 4.5 applications of insecticides between August 1 and September 10 for control of Heliothis spp. larvae. The average yield was 666 pounds of lint per acre.

Field 19 (Fig. 15) received aldicarb in-furrow at planting plus a foliar application of insecticide for thrips control. The initial beneficial arthropod population was fairly high, but it decreased and remained below 10,000 per acre from mid-June to mid-July. Then it peaked at about 19,000 on August 5 and decreased sharply after mid-August though there was a slight increase on September 2. The Heliothis spp. larval population became evident by early July, and populations averaged 6,000 and 5,200 per acre on August 5 and 12. Average boll damage was 7,800 per acre on August 26 and September 2. The field received 4 insecticide applications between August 3 and September 9 for control of Heliothis spp. larvae. The estimated yield was 972 pounds of lint per acre.

Thus in 1978 in Panola County, the beneficial arthropod population was much lower than in 1977. When the 4 fields receiving foliar applications of insecticides for thrips control were compared with the 3 fields that were not so treated, the difference in numbers of insecticide applications for control of Heliothis spp. larvae was slight, 2.5 versus 2.0. The beneficial arthropod populations were similar. They were also similar to populations in fields receiving foliar applications for thrips control but no insecticidal treatment for control of Heliothis spp. (Fig. 13).

Fields 12, 13, 15, and 17 (Fig. 14); and field 19 (Fig. 15) receiving aldicarb in-furrow at planting or that treatment plus a foliar insecticide application for thrips control had similar populations of beneficial arthropods. However, the populations were considerably lower than in the groups previously discussed. Fields in Fig. 14 and 15 received 4.2 and 4.0 applications of insecticides, respectively, for control of Heliothis spp. larvae. Yields in the 12 fields receiving treatment for Heliothis spp. control produced 666 pounds of lint per acre; the 8 fields not so treated produced 652 pounds.

Pontotoc County. Fields 21, 22, 23, and 24 (Fig. 16) were not treated for early-season insects but were treated an average of 2.5 times between July 14 and August 15 for control of Heliothis spp. larvae. The beneficial arthropod population was initially low, peaked at 28,000 per acre on July 8, and declined progressively until August 19 when it averaged about 3,000 per acre. There was a subsequent increase to 30,000 per acre on September 30 at a time when fully matured plants exposed to good rains had put out new growth and began fruiting again. The Heliothis spp. population was rather low and peaked at about 2,500 larvae per acre on August 12. Average boll damage peaked at 11,000 per acre on September 2. The average yield was 531 pounds of lint per acre.

Fields 25, 26, 27, 28, 29, and 30 (Fig. 17) received no insecticide treatment. The initial beneficial arthropod population peaked at about 24,000 per acre on July 15. It then decreased to about 17,000 on August 12 and slowly increased to 26,000 per acre on September 2. Heliothis spp. larval populations were low; there was a peak of 1,350 per acre on July 22. Boll damage averaged 5,800 on September 2 and 9, 1,800 on September 16 and 6,000 on September 23. The average yield was 604 pounds of lint per acre.

Beneficial arthropod populations were similar for the 2 groups of fields. Heliothis spp. larval populations and boll damage were slightly higher in the

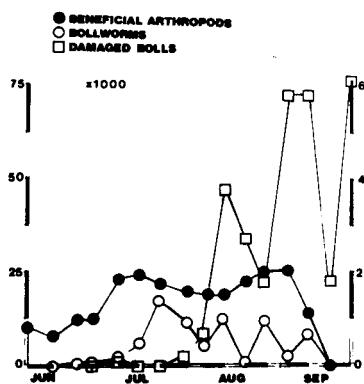


FIG. 17. Populations of beneficial arthropods (left) and *Heliothis* spp. larvae and damaged bolls per acre in fields 4, 7, 25, 26, 27, and 28 in Pontotoc County, MS, 1978.

fields receiving late treatment. Difference in yield was not great but favored the fields that received no insecticidal treatment.

Analyses. A very high probability of correlation (greater than the 99 percent level) was indicated between beneficial arthropod populations and numbers of damaged and percentages of damaged bolls. The r values for the correlation between beneficial arthropod populations and Heliothis spp. second and third generation larval populations, numbers of bolls, numbers of damaged and percentages of damaged bolls were 0.033, 0.026, 0.119, -0.315 and -0.323, respectively.

DISCUSSION

We were fortunate in that beneficial arthropod populations were high in 1977 and moderate in 1978 (about one-half that in 1977). As would be expected, insecticidal treatment for control of TPB in late June or in July adversely affected beneficial arthropod populations and were conducive to development of a subsequent increased Heliothis spp. larval population. However, in 1978 the TPB population was light and insecticides were not applied for its control. Also, the Heliothis spp. larval population was moderate even though beneficial arthropod populations were much lower than in 1977. Thus in that year, the average numbers of insecticide applications necessary for control of Heliothis spp. larvae in Panola and Pontotoc Counties were only 1.9 and 0.9, respectively. In 1977, the averages were 3.9 and 1.6 for the two respective counties.

On the other hand, although foliar insecticide or aldicarb in-furrow at-planting in 1977 resulted in somewhat lower populations of beneficial arthropods compared with populations in fields that did not receive such treatment; the need for control of Heliothis spp. was not greater in the insecticide-treated fields in that year when conditions were extremely favorable for beneficial arthropods. In 1978 when conditions were less favorable for beneficial arthropods, more applications of insecticides had to be made for control of Heliothis spp. in fields that had received aldicarb in-furrow at planting. Whether such treatment should or should not be used should apparently depend on conditions at planting. If good weather prevails and is expected to continue, aldicarb can probably be used without adverse effect. But geographical location too should be considered. Conditions in the hill sections of Mississippi are more favorable for development of such populations than in the Delta where intensive cropping means that little habitat is suitable as reservoirs for beneficial arthropods.

Although the data showed that elimination of insecticide treatment in early or mid-season conserves beneficial arthropods, thrips, lygus bugs or other insect pests may damage the crop if they are not controlled at that time. However, if the farmer and his consultant are aware of the interrelationship between insecticidal treatment and beneficial arthropod populations, they may be able to devise an insect control program that is appropriate for the given situation.

ACKNOWLEDGEMENT

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PITFALL COLLECTED INSECTS FROM VARIOUS LOWER RIO GRANDE
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ABSTRACT

Pitfall collections of arthropods were conducted for approximately 12 months in 11 primary Lower Rio Grande Valley habitats. Insect spp. diversity generally increased as plant spp. diversity increased. Species caught were identified and then listed corresponding to their seasonal habitats. This procedure helps define the Valley ecosystem and records species for future comparisons should the ecosystem undergo change.

INTRODUCTION

The Lower Rio Grande Valley, an extensively farmed area of 1.1 million ha, cultivates approximately 324,000 ha of cotton and grain sorghum annually. Vegetable crops are grown on about 27,000 ha during the winter months following other crops. Citrus (30,000 ha), sugarcane (15,000 ha), unimproved pastures (289,000 ha) and improved pasture (36,000 ha) comprise a further segment of Valley agriculture. Due to this extensive farming; drainage ditches, roadsides and other wasteland (about 26,000 ha) are undoubtedly important to maintenance of insect fauna. The diversity of animal species numbers has been shown to increase as plant species diversity increases within a habitat (MacArthur and MacArthur 1961, Miller 1967 and Price 1975).

Studies of above ground arthropod habitats have been conducted throughout the Valley in recent years to help define the ecosystem (Schuster and Dean 1957, Schuster et al. 1969, Schuster and Boling 1974, Fuchs and Harding 1976, Harding 1976, Harding and Dupnik 1976 and Harding et al. 1976). However, none of these studies specifically identified ground inhabiting arthropods. Therefore, pitfall trap studies of primary Valley habitats were begun in Dec., 1975.

Several recent arthropod studies have utilized pitfall traps as sampling tools. Thomas and Sleeper (1977) studied tenebrionid abundance in a desert habitat while Esau and Peters (1975) and Allen and Thompson (1977) have examined carabid populations in several habitats. It is generally agreed that quantitative estimates of species within or between habitats are unreliable due to various factors such as thatch density, rainfall and temperature (Greenslade

1/ Technical article 14991 from the Texas Agricultural Experiment Station.

2/ Respectively, Research Associate and Associate Professor, Texas Agricultural Experiment Station, Weslaco, Texas.

1964 and Southwood 1968). However, pitfall traps do provide an indication of species located within various habitats and record an important part of the ecosystem not obtained by other sampling procedures.

METHODS AND MATERIALS

The primary habitats selected for study were within a 9.7 km radius of each other in the mid-section of the Lower Rio Grande Valley. Pesticidal usage was not documented for the habitats or adjacent ones because of the enormity of such a task. Habitat descriptions as well as a visual rank of plant diversity are listed in Table 1. Diversity was based on the number of different plants seen in the habitat area surrounding the traps.

Five wide-mouthed, glass, 946 ml jars, 7.5 cm. I.D., were randomly located in each habitat. Each jar was placed into the ground with the mouth flush with ground level. An isopropyl alcohol-kerosene mixture was used in each trap. Traps were inspected and contents removed weekly. Specimens were separated for identification within 24 hrs after collection.

Sampling of habitats 1-7 was begun on Dec. 4, 1975, 8-10 on Feb. 2, 1976 and 11 on Apr. 22, 1976. All sampling was discontinued Dec. 2, 1976, except for habitat 9 which was discontinued Sept. 2, 1976.

Insect identifications were made by Dr. Lloyd Knutson and staff, A.R.S., Systematic Entomology Laboratory, Beltsville, Maryland.

RESULTS AND DISCUSSION

During this study, the largest number of insect species were collected from the drainage ditch bank (#1) habitat which would be anticipated since this habitat had the most diverse plant flora (Table 1). Habitats 7 and 8 did not exhibit this relationship of plant and animal diversity clearly and cannot be explained.

TABLE 1.--Habitat Description and Number of spp. Collected from Pitfall Traps, 1975-76.

Habitat No.	Habitat Description	Primary Habitat Plants	Visual Rank of Diversity ^a	No. Spp. Collected
1	Bank of drainage ditch next to open field planted in grain sorghum during Spring and Summer.	johnsongrass, <u>Sorghum halepense</u> buffel grass, <u>Cenchrus ciliaris</u> cactus, <u>Opuntia vulgaris</u> mesquite, <u>Prosopis chilensis</u>	1	64
2	Roadside next to open field planted in cotton during Spring and Summer.	Kleberg bluestem, <u>Andropogon annulatus</u>	7	30
3	Pasture, hay and grazing land.	Kleberg bluestem African Star grass, <u>Cynodon dactylon</u> hyb.	6	19

TABLE 1.--(Continued).

Habitat No.	Habitat Description	Primary Habitat Plants	Visual Rank of Diversity ^a	No. Spp. Collected
4	Citrus orchard, mowed between rows.	citrus bermuda grass, <u>Cynodon dactylon</u> African Guinea grass, <u>Panicum maximum</u> Afr.	4	51
5	Roadside between citrus orchards.	ornamental date palm, <u>Phoenix carariensis</u> Kleberg bluestem African Guinea grass	3	51
6	Side of drainage ditch, open fields on both sides planted in cotton during Spring and Summer.	Kleberg bluestem buffel grass	8	33
7	Citrus, disked between rows.	citrus	9	41
8	Side of drainage ditch, citrus on one side and open field planted in cotton during Spring and Summer on other side.	buffel grass Kleberg bluestem cattails Paragrass, <u>Panicum purpurascens</u>	2	30
9	Cotton field adjacent to habitat 8.	cotton	11	20
10	Large mowed grass area between citrus orchards.	Kleberg bluestem bermuda grass	5	34
11	Sugarcane field.	sugarcane	10	13

^a1 = greatest plant diversity and 11 = least plant diversity.

Insects seasonal occurrence and habitats in which they were collected are listed in Table 2. Carabids, Pocillus sp. and Scarites subterraneus F., were the most abundant Coleoptera. The presence of certain unexpected species such as winged Diptera in the traps cannot be adequately explained. Recording of all species identified provides a base to assess habitat importance in a pest management system and changes in ecosystem quality sometime in the future.

TABLE 2.--Insect Fauna Collected from Pitfall Traps in Various Lower Rio Grande Valley Habitats, 1975-76.

		Habitat Designation ^{a/}			
		Winter (Dec.-Feb.)	Spring (Mar.-May)	Summer (June-Aug.)	Fall (Sept.-Nov.)
HEMIPTERA					
Belostomatidae					
<u>Belostoma fusciventre</u> (Dufour)	7				
Miridae					
<u>Polymerus basalis</u> (Reut.)	6				
Lygaeidae					
<u>Ligyrocoris litigiosus</u> (Stal)			4		
<u>Pachybrachius basalis</u> (Dallas)				1	
Cydnidae					
<u>Dallasiellus lugubris</u> (Stal)				1,2	
<u>Pangaeus bilineatus</u> (Say)					1
HOMOPTERA					
Membracidae					
<u>Micrutalis calva</u> (Say)	8				
<u>Spissistilus festinus</u> (Say)	1				
Ciccadellidae					
<u>Aceratagallia sordida</u> Oman	4				
<u>Balclutha hebe</u> (Kirkaldy)				5	
<u>Carneocephala sagittifera</u> (Uhler)	4,10			5,7	
<u>Chlorotettix scutellatus</u> Osborn	1				

TABLE 2.--(Continued).

	Habitat Designation ^{a/}			
	Winter (Dec.-Feb.)	Spring (Mar.-May)	Summer (June-Aug.)	Fall (Sept.-Nov.)
<u>Draeculacephala portola</u> Ball	4			
<u>Deltocephalus sonorus</u> Ball			7	
<u>Empoasca</u> sp.	5			
<u>Graminella nigrifrons</u> (Forbes)			5	
<u>Homalodisca insolita</u> (Walker)	4,6			
<u>Negosiana dualis</u> (DeLong)	4			
<u>Paraphlepsius continuus</u> DeLong			7	
<u>Planicephalus flavicosta</u> (Stal)	1,4,5,7			
<u>Polyamia</u> sp.	6			
<u>Stirellus obtutus</u> (Van Duzee)	4,5			
<u>Texananus spatulatus</u> (Van Duzee)	6,7			
<u>Xestocephalus lunatus</u> Peters	6			
<u>X. pulicarius</u> Van Duzee	4,5			
<u>X. tessellatus</u> Van Duzee	4			
Delphacidae				
<u>Delphacodes pseudoseminigra</u> (Muir and Giffard)	10			
Cixiidae				
<u>Oliarus aridus</u> Ball			3	
<u>Oliarus</u> sp.		8		
<u>Pintalia dorsivittata</u> (Van Duzee)		5		
COLEOPTERA				
Carabidae				
<u>Aspidoglossa</u> sp.	7	8	10	
<u>Galerita</u> sp.				11

TABLE 2.--(Continued)

		Habitat Designation ^{a/}			
		Winter (Dec.-Feb.)	Spring (Mar.-May)	Summer (June-Aug.)	Fall (Sept.-Nov.)
<u>Poecilus</u> sp.			1,4,8-10	1,3-5,9	2,5-7
<u>Scarites subterraneus</u> F.			1-3,6,8-11	1,2,11	1,2,5
<u>Selenophorus</u> sp.				2	4
Dyticidae					
<u>Laccophilus proximus</u> Say	5				
Histeridae					
<u>Hololepta cacti</u> LeC.	1				
<u>Phelister subrotundatus</u> (Say)			4		
<u>Xerosaprinus</u> sp.			10		
Hydrophilidae					
<u>Cercyon</u> sp.	1,3				
Scaphidiidae					
<u>Cyparium ater</u> Casey	1				
Cantharidae					
<u>Belotus</u> sp.			1		
<u>Chauliognathus marginatus</u> (F.)				8	2
<u>Silis</u> sp.			7		
Elateridae					
<u>Aeolus scutellatus</u> Schfr.				5	
<u>Conoderus amplicollis</u> (Gyllenhal)			1		
<u>Meristhus scobinula</u> Candeze			10		
<u>Neotrichophorus texanus</u> (LeConte)				5	
Ptilodactylidae					
<u>Ptilodactyla</u> sp. nr. <u>serricollis</u>				8	
Languriidae					
<u>Loberus</u> sp.	1				
Cucujidae					
<u>Ahasverus rectus</u> (LeConte)			10		
<u>Leptophloeus</u> sp.				10	

TABLE 2.--(Continued).

	Habitat Designation ^{a/}			
	Winter (Dec.-Feb.)	Spring (Mar.-May)	Summer (June-Aug.)	Fall (Sept.-Nov.)
Phalacridae				
<u>Acylomus</u> sp.	4			
<u>Stilbus</u> sp.	6			
Nitidulidae				
<u>Carpophilus dimidiatus</u> (F.)	2	1	7	
<u>C. freemani</u> Dobs.	1,5		5	
<u>C. humeralis</u> (F.)	6		5,10	
<u>C. mutilatus</u> Er.	2-4,6,7		7	
<u>Carpophilus</u> sp.	1,3-7	7	7	
<u>Conotelus mexicanus</u> Murr.	4			
<u>Lobiopa insularis</u> (Cast.)	4		1,7	
<u>Stelidota ferruginea</u> Reitt.	1,5	5		
<u>S. geminata</u> (Say)		1,7	7	4
Coccinellidae				
<u>Hippodamia convergens</u> Guerin		9		
<u>Psyllobora renifer</u> Casey	4	8		
<u>Scymnus</u> (Pullus) <u>loewii</u> Mulsant		9		
Anthicidae				
<u>Anthicus</u> sp.	1,7,9	1,10	7,10	2
Mycetophagidae				
<u>Litargus balteatus</u> LeConte		7	7	
<u>Typhaea stercorea</u> (L.)	6			
Rhipiphoridae				
<u>Trigonodera schaefferi</u> Rivnay			1	
Tenebrionidae				
<u>Blapstinus fortis</u> LeConte	1,4,8	11		
<u>Blapstinus</u> sp.	1,2,10	4,10	10	
<u>Opatrinus aciculatus</u> LeConte	1,3	1,5	10	
<u>Platydema micans</u> Zimm.	5	5		
Bostrichidae				
<u>Amphicerus cornutus</u> (Pallas)	1,2,5,6	4		

TABLE 2.--(Continued).

		Habitat Designation ^{a/}			
		Winter (Dec.-Feb.)	Spring (Mar.-May)	Summer (June-Aug.)	Fall (Sept.-Nov.)
Scarabaeidae					
<u>Aromala</u> sp.			1,8		
<u>Aphodius lividus</u> (Oliv.)	10		4,10	9	
<u>Ataenius cognatus</u> (LeConte)			11	8	
<u>Ataenius</u> sp.				9	
<u>Ochodaeus frontalis</u> LeConte				10	
<u>Phyllophaga crinita</u> (Burm.)			1-6,8,9,11		
<u>Pseudocanthion perplexus</u> (LeConte)			1	1	1
Cerambycidae					
<u>Dorcasta cinerea</u> (Horn)	7				
<u>Lepturges angulatus</u> (LeConte)				7	
<u>Lissonotus flavocinctus</u> Dup.			9		
Chrysomelidae					
<u>Chaetocnema pulicaria</u> Melsheimer or very nr.	8,10				
<u>Diachus auratus</u> F.			7		
<u>Kuschelina petaurista</u> (F.)	10				
<u>K. texana</u> Crotch or nr.	4				
<u>Lema texana</u> Crotch				10	
<u>Longitarsus</u> sp. nr. <u>Bicolor</u> Horn	10,7				
<u>Monomacra tibialis</u> (Oliver)	4,5				
<u>Monoxia sordida</u> LeConte				7	
<u>Myochrous denticollis</u> (Say) or nr.	6				
<u>Myochrous</u> sp. nr. <u>floridanus</u> Schaeffer	6				
<u>Myochrous</u> sp.	3,8		11		
<u>Phyllotreta</u> sp.	6				
Bruchidae					
<u>Caryobruchus gleditsiae</u> (L.)	5				
<u>Mimosestes sallaei</u> (Sharp)	1				
<u>Stator subaeneus</u> (Schaeffer)	1				
<u>S. vachelliae</u>			1,4		

TABLE 2.--(Continued).

	Habitat Designation ^{a/}			
	Winter (Dec.-Feb.)	Spring (Mar.-May)	Summer (June-Aug.)	Fall (Sept.-Nov.)
Anthribidae		1,4		
<u>Araecerus fasciculatus</u> (DeG.)			8	
Curculionidae				
<u>Conotrachelus seniculus</u> LeConte		10		
<u>Cophes fallax</u> (LeConte)		8	1	
<u>Sitophilus zeamais</u> Mots.	1,2,4,5	10,11	5	
<u>Sphenophorus compressiro-</u> <u>tris</u> (Say)			2	
<u>Trichobaris texana</u> LeConte	8			
Scolytidae				
<u>Coccotrypes dactyliperda</u> (F.)	3		5	
<u>Coccotrypes</u> sp.	4	5	5	
DIPTERA				
Bibionidae				
<u>Dilophus orbatus</u> (Osten Sacken)		5		
Mycetophilidae				
<u>Allodia</u> sp.	2			
<u>Leia bivittata</u> Say		4		
<u>Leia</u> sp.		1		
<u>Orfelia</u> sp.	3			
Sciaridae				
<u>Bradysia</u> sp.	4,5	5		
<u>Lycoriella</u> sp.	5			
Stratiomyidae				
<u>Hoplitimyia mutabilis</u> (F.)				
Therevidae				
<u>Psilocephala</u> sp.				
Dolichopodidae				
<u>Condyllostylus longitalus</u> (Van Duzee)	4,5,7	1,3,5,8		

TABLE 2.--(Continued).

	Habitat Designation ^{a/}			
	Winter (Dec.-Feb.)	Spring (Mar.-May)	Summer (June-Aug.)	Fall (Sept.-Nov.)
<u>Condyllostylus</u> sp.	7	2,3,5,7,9, 10	2,5,7,8, 10,11	4-8,10,11
<u>Medetera nigripes</u> Loew	1		7	
Phoridae				
<u>Apocephalus</u> sp.		5	5	
<u>Dohnniphora incisuralis</u> (Tw.)	1,2,4-7		5,7	
<u>Dohnniphora</u> sp.	1-5,7,10	1-11	1-11	1-8,10,11
<u>Megaselia</u> sp.	6	7		
Conopidae				
<u>Zodion americanum</u> Wiedemann		4		
Sepsidae				
<u>Palaeosepsis</u> sp.	4			
Sphaeroceridae				
<u>Leptocera</u> sp.	1,2,4-6	1	8	
<u>Sphaerocera varipes</u> Malloch				
Ephydriidae				
<u>Gastrops niger</u> Williston	5			
Drosophilidae				
<u>Drosophila bromeliae</u> Sturt. or nr.			7	
<u>D. falleni</u> Wheeler	1,4,5,7			
<u>D. melanogaster</u> Meigen	4,7			
<u>Drosophila</u> sp.	1,3,4,6,7	4,5,7-10	1-10	1,4,5,7,8,10,11
Chloropidae				
<u>Hippelates dissidens</u> (Tucker)	2			
<u>H. pusio</u> Loew	2,4			
<u>Conioscinella</u> sp.	7			
Heleomyzidae				
<u>Pseudoleria</u> sp.	1,5,10			
Muscidae				
<u>Atherigona orientalis</u> Schiner	1,2,5,8,10	1,9-11	1,2,5,8-11	1,2,8,10,11

TABLE 2.--(Continued).

	Habitat Designation ^{a/}			
	Winter (Dec.-Feb.)	Spring (Mar.-May)	Summer (June-Aug.)	Fall (Sept.-Nov.)
<u>Atherigona</u> sp.	5-7	5-7	5-7	5-7
<u>Coenosia</u> sp.	1,2			
<u>Limnophora narona</u> (Walker)	1-4			
<u>Limnophora</u> sp.	3,4	3-5	3,4	3,4
Calliphoridae				
<u>Cochliomyia macellaria</u> (F.)	1,2,4,5,7, 9,10	1	1,4,6,7	
Sarcophagidae				
<u>Sarcophaga</u> sp.			7	
Tachinidae				
<u>Gonia</u> sp.		9		
<u>Gymnoclytia unicolor</u> Brks.		1		
HYMENOPTERA				
Braconidae				
<u>Apanteles</u> sp.	1,4,5	5,10		
<u>Ichneutidea proteroptoides</u> Vier.		9		
<u>Orgilus</u> sp.	5	5,7	8	
Ichneumonidae				
<u>Enicospilus merdarius</u> (Grav.)	3			
<u>Mesochorus</u> sp.	1			
Chalcididae				
<u>Dirhinus texanus</u> (Ashm.)	10			
Eucoilidae				
<u>Hexacola</u> sp.	4	7		
Proctotrupidae				
<u>Cryptoserphus</u> sp.		4		
Diapriidae				
<u>Pantoclis</u> sp.		4		
<u>Trichopria</u> sp.	1	7		
Scelionidae				
<u>Calotelea</u> sp.	7	4		
<u>Ceratobaeus</u> sp.	6		5	
<u>Scelio</u> sp.		1		

TABLE 2.--(Continued).

	Habitat Designation ^{a/}			
	Winter (Dec.-Feb.)	Spring (Mar.-May)	Summer (June-Aug.)	Fall (Sept.-Nov.)
Bethylidae				
<u>Holepyris</u> sp.		8		
Dryinidae				
<u>Neogonatopus</u> sp.	6			
Mutillidae				
<u>Sphaerophthalma</u> sp.	4			
Pompilidae				
<u>Ageniella obscura</u> Banks		2	2	
<u>Anoplius</u> sp.		10		10
<u>Dipogon melanocephalus</u> (Cam.)	5	8		
<u>Priocnemis cornica</u> (Say)	2			
Sphecidae				
<u>Pluto</u> sp.		9		
Andrenidae				
<u>Perdita lacteipennis</u> Swenk and Cockerell			4	
Halictidae				
<u>Agapostemon texanus</u> Cr.		10		
Megachilidae				
<u>Megachile polcaris</u> Say		5		
Anthophoridae				
<u>Melissodes</u> sp.		6	1	

^{a/}Habitat descriptions listed in Table 1.

Formicidae (Table 3) were collected year-round in each habitat and are therefore shown in a separate table. Campanotus abnominalis transvectus Wheeler, Pachycondyla harpax (F.) and Pogonomyrmex barbatus (F. Smith) occurred in almost all habitats while Solenopsis geminata (F.) was collected from 7 habitats. Schuster and Dean (1957) reported Solenopsis geminata and Pogonomyrmex barbatus as pests of citrus orchards due to their interfering with predators and parasites of scale insects.

TABLE 3.--Formicidae Collected from Pitfall Traps in Various Lower Rio Grande Valley Habitats, 1975-76.

	Habitats Designation ^{a/}
<u>Camponotus abnominalis transvectus</u> Wheeler	1-10
<u>Camponotus</u> sp.	8
<u>Iridomyrmex pruinosus</u> (Roger)	6
<u>Iridomyrmex</u> sp.	10
<u>Labidus coecus</u> (Latreille)	1
<u>Leptogenys elongata</u> (Buckley)	1,2,5,8
<u>Monomorium minimum</u> (Buckley)	2,6
<u>Pachycondyla harpax</u> (F.)	1-8,10,11
<u>Pachycondyla</u> sp.	1,3
<u>Paratrechina</u> sp.	6
<u>Pheidole ridicula</u> Wheeler	5
<u>Pheidole</u> sp.	1,5-7,11
<u>Pogonomyrmex barbatus</u> (F. Smith)	1-10
<u>Smithistruma</u> sp.	5
<u>Solenopsis geminata</u> (F.)	1-7
<u>Solenopsis</u> sp.	3,5-7
<u>Tapinoma sessile</u> (Say)	1

^{a/}Habitat descriptions listed in Table 1.

Large numbers of labidurids, staphylinids and spiders were collected from all habitats. No taxonomist was available for identification of these arthropods and therefore they are not presented.

Habitat preference of several species as well as their place in ecosystem dynamics warrants further intensive investigations based on this study. The importance of some wastelands for species reservoirs has been shown therefore providing a basis for more precise monitoring of the environment.

ACKNOWLEDGEMENT

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AUTOMATIC INDOOR INSECTICIDE DUSTER

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ABSTRACT

An automated method of disinsecting buildings with resmethrin dust has been developed and tested. The device has a dust reservoir, powder measure, vibrator, and CO₂ gas, and premeasures and disperses insecticidal dust, either on command or by programmed timer. The method has been extremely effective for dispersing ULV doses of synthetic pyrethroids for controlling flies in a poultry building.

INTRODUCTION

An automated method has been developed for applying insecticidal dusts to agricultural buildings that attract unwanted insect pests. An electromechanical device meters a predetermined volume of dust into a chamber, and a brief blast (<1 sec) of compressed air or gas propels the dust into the room to be disinsected. The sequence may be actuated either by a pushbutton or an electric timer. Aside from the periodic filling of a dust reservoir, application requires minimum human effort.

MATERIALS AND METHODS

All parts used for this system are standard hardware items: Lyman Ideal® No. 55 adjustable cavity gunpowder measure, 115-V AC 6-rpm gear motor, electric vibratory engraver, solenoid valve, and two micro-switches. The gunpowder measure includes a gravity-feed powder reservoir of 200-cm³ capacity.

As shown in Fig 1, the powder measure is vertically mounted with the reservoir above, and the vibrator touches the base of the measure. The vibrator operates continually during the entire loading and firing cycle to prevent bridging and keep the dust flowing freely. The handle of the measure is activated by the gear motor through a connecting link. Also attached to the gear motor are two cams for activating two micro-switches. One switch energizes the gear motor and vibrator after the pushbutton is released, and the second actuates the solenoid valve in the compressed air or gas line.

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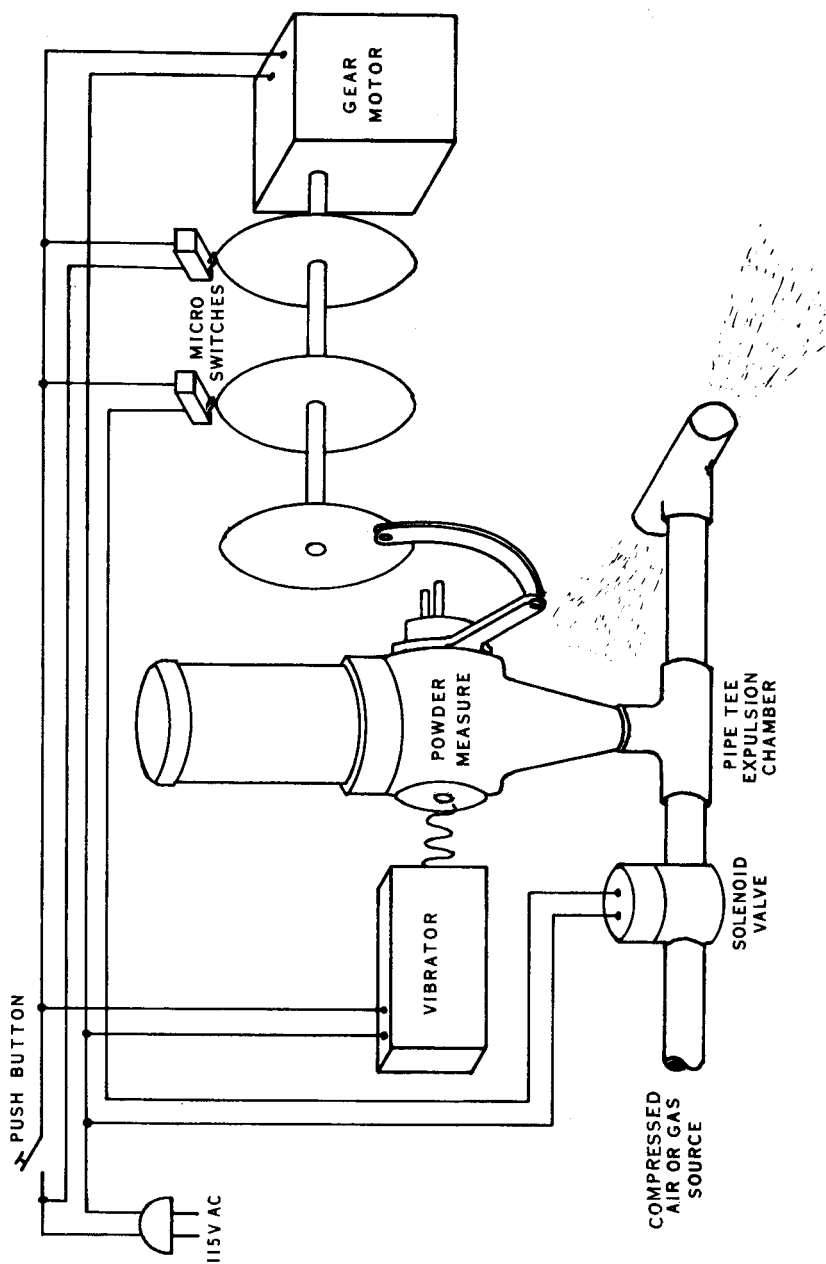


FIG. 1 - Schematic Diagram of Automatic Indoor Insecticide Duster Showing Components in Relative Position.

When the start button is pushed, the measure handle is pulled down and insecticide dust in the cavity is dropped into the "T"-shaped expulsion chamber. As the measure handle is returned to its up position, a seal is formed between the expulsion chamber and the dust cavity. A cam then activates the second microswitch, and the solenoid valve opens and releases high pressure air or gas through the expulsion chamber. The second "T" directs the propelled dust in opposite directions.

Compressed air and CO₂ gas have been satisfactory propellants. CO₂ delivers a uniformly high pressure (ca. 5515 kPa) gas and can be pressure-regulated for the space to be treated (1379 kPa for <150 m³ and 5515 kPa for up to 425 m³). Compressed air from high pressure (20,682 kPa) scuba tanks that were regulated to release 1379 kPa was effective for treating spaces <150 m³. The availability of either air or gas probably will determine which is used.

The three powder measure slides were locked together to meter suitable volumes of dust and the 0-15 scale was chosen as the most practical. Calibrations were made at three volume settings of the 0-15 scale. The insecticide dust was 30% resmethrin on Hi Sil® 233, having a mass of 0.16 g/cm³. For calibration of the measure, it was operated 15 times at each setting, the dust charges were weighed, and the confidence interval was computed.

Practical tests were conducted in a 140 m³ poultry house containing batteries of caged hens. Each day for eight days, house flies, *Musca domestica* L., were caged and placed beneath and above the hen cages. The three powder measure settings were tested (four replicates each). The 30% resmethrin dust was applied as a single daily treatment. Fly knockdown 15 min after and fly mortality four hours after treatment were recorded.

RESULTS AND DISCUSSION

Dust quantities delivered by the powder measure were essentially proportional to the scale setting and were quite uniform from charge to charge as shown in Table 1.

TABLE 1. Results of Powder Measure Dust Delivery for Three Scale Settings.

Powder measure scale setting	Insecticide dust delivered	
	Mean wt. (g)	Conf. interval ($\bar{P} < 0.05$)
5	0.49	+0.11
10	1.12	+0.11
15	1.62	+0.02

The mortality data in Table 2 indicate a scale setting of ten would release enough resmethrin dust to adequately control flies in most situations.

TABLE 2. Resmethrin Efficacy Tests for Controlling the House Fly.

Powder measure scale setting	% knockdown	% mortality	
	\bar{x}	Mean	Conf. interval ($P < 0.05$)
5	65.2	88.3	+0.09
10	67.0	97.7	+0.04
15	96.7	100.0	+0.00

One hundred percent control may be attained by increasing the setting to 11 or 12. Indoor accumulation of insecticidal dusts after repeated treatments may permit lower scale settings to be used as long as the results evaluated cover several days of fly-kill data.

This method of dust application is effective as long as the powder measure reservoir contains flowable dust and 1379 kPa gas pressure is available. The application may be repeated for multiple doses by repeating the pushbutton activation or by adding a sequential timer to the electrical control system.

ACKNOWLEDGEMENT

We thank Mr. Benjamin L. Everett for his mechanical ability in building the duster.

FIRST UNITED STATES RECORD OF ALEUROPTERYX SIMILLIMA, A
PREDATOR OF SCALE INSECTS ON ORNAMENTAL JUNIPER
(NEUROPTERA: CONIOPTERYGIDAE)

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ABSTRACT

The coniopterygid Aleuropteryx simillima Meinander, known previously only from northern Mexico, is recorded from Houston, Texas. Adults and larvae were collected from ornamental plantings of juniper infested with the scale insect Carulaspis minima (Targ.-Tozz.); both stages fed on scales in the laboratory.

The North American Coniopterygidae remained poorly known until Meinander (1972) provided a worldwide revision with modern descriptions and workable keys. One of several species he described was Aleuropteryx simillima, based on males from Baja California and Tamaulipas in northern Mexico. A female from Sonora was only provisionally referred to this new species until additional material from Sonora allowed Meinander (1974) to confirm the relationship. The biology and immature stages have remained unknown. From recent collecting in Texas I can give the first U. S. record for A. simillima and record this coniopterygid as a predator of scale insects on ornamental junipers.

During 27-30 November 1978 I found A. simillima at 2 locations in downtown Houston. Adults, mostly females, and a few 2nd and 3rd instars were present on Pfitzer juniper (Juniperus chinensis cv 'Pfitzeriana') infested with so-called minute cypress scale, Carulaspis minima (Targ.-Tozz.). Adults and larvae, when placed with infested branches from the host plants, readily fed on female scales. A. simillima thus occupies a niche similar to that of A. juniperi Ohm in the eastern U. S. This Old World species, thought to have been accidentally introduced with nursery stock, was recorded from North America by Henry (1976). The latter coniopterygid has become the most effective natural enemy of C. minima and juniper scale, C. juniperi (Bouché), in ornamental plantings in Pennsylvania (Henry 1976, Stimmel 1979).

The third instar of A. simillima is 2.20 mm long and dark gray with extensive white markings. These lighter areas give a mottled effect so that the white and gray transverse bands characteristic of A. juniperi (see Henry 1976, Fig.4,p.198) are indistinct.

Entomologists are encouraged to determine the distribution and abundance of A. simillima. This species, like its eastern counterpart A. juniperi, may be an important predator of scale insect pests on ornamental junipers.

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CONTROL OF THE CARMINE SPIDER MITE ON
GREENHOUSE GROWN FOLIAGE PLANTS^{1/2/}

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ABSTRACT

Oxamyl provided adequate mite control on Brassia actinophylla, Codiaeum variegatum, and Epipremnum aureum and appeared to be acceptable for use on these plants. Tetranychus cinnabarinus (Boisduval) populations were not effectively controlled either by butocarboxime or oxamyl on Dieffenbachia sp. 'Exotica' or by butocarboxime on Brassia actinophylla.

INTRODUCTION

The Lower Rio Grande Valley of Texas is a major commercial production area for tropical foliage plants. In 1977 ca. 1,198,819.7 m² of greenhouses and shade houses plus an additional 270.54 ha of field grown trees and shrubs accounted for gross sales of \$20,039,000 (Anonymous 1978). Insect and disease losses, as well as fluctuations in the commercial market, reduce grower profits. Spider mites (Acari: Tetranychidae) are among the most common of foliage plant pests. Mite feeding damage ranges from yellow or gray stippled patterns on the leaves in light infestations to leaf death in heavy infestations (Short 1977). The carmine spider mite, Tetranychus cinnabarinus (Boisduval), is commonly found infesting foliage plants in the Lower Rio Grande Valley and frequently causes severe plant damage. Carbamate and organophosphate compounds currently labelled for control of carmine mite on ornamentals give inconsistent control.

This paper reports results of tests with several alternative miticides for controlling the carmine spider mite under greenhouse conditions.

METHODS AND MATERIALS

Six tests were designed to test the effectiveness of butocarboxime (3-methylthio-0-[(methylamino)carbonyl]oxime-2-butanone, Drawin 50EC(R)) and oxamyl (Vydate L(R)) for control of the carmine spider mite. The chemicals and plants tested are listed in Tables 1-3. Malathion 55 or diazinon 2EC was used as a standard in all tests, and an untreated replicate was counted as a check. The butocarboxime and oxamyl applications also included 51.47 cc/100 liters spreader sticker to assure uniform deposition of pesticides on the plant surfaces. All chemical rates and dates of pesticide application are given in Tables 1-3. Numbers of applications were based on the proposed labels for each product. The chemicals were applied to the point of run-off on upper and lower leaf surfaces. A 7.56 liter compressed air sprayer equipped with a 65° brass nozzle, and operated at a pressure of ca. 2.25 kg/cm² was used to apply the pesticides.

^{1/} Approved for publication as TA 15647 by director, Texas Agricultural Experiment Station. This paper reports the results of research only. Mention of pesticide does not constitute recommendation for use by TAES or the USDA nor does it imply registration under FIFRA as amended. Also, mention of a proprietary product does not constitute an endorsement by TAES or the USDA.

^{2/} This research was conducted in cooperation with the Texas Agricultural Experiment Station.

TABLE 1. Evaluation of Miticides for Control of *Tetranychus cinnabarinus* on *Dieffenbachia* sp. 'Exotica'.

Average no. female mites/plant and %control at indicated day										
Chemical	Rate A.I./ 100 liters	Avg. No. Female mites/plant Pretreatment	post initial treatment							
			7		14		21		28	
			No. mites	%control	No. mites	%control	No. mites	%control	No. mites	%control
					Test 1 ^{a/}					
Butocarboxime	0.03kg	17 ab ^{b/}	4 c	82 ^{c/}	25 b	73	38 bc	58	38 a	60
Butocarboxime	0.06kg	9 b	10 abc	21	48 a	2	44 ab	8	38 a	25
Butocarboxime	0.12kg	14 ab	10 abc	49	26 b	65	32 bc	54	28 a	62
Butocarboxime	0.24kg	18 a	7 bc	71	14 c	85	26 c	72	28 a	72
Malathion 55	0.18kg	13 ab	12 ab	10	26 b	61	35 bc	48	34 a	52
Untreated	-----	10 ab	14 a		55 a		54 a		57 a	
					Test 2 ^{d/}					
Oxamyl	0.03kg	13 c	21 a	-18	52 a	-111	103 a	-520	177 a	-654
Oxamyl	0.06kg	14 bc	22 a	-13	41 a	-54	77 ab	-329	108 b	-327
Oxamyl	0.24kg	25 ab	23 a	32	31 a	32	42 cd	-34	22 d	51
Malathion	0.18kg	16 abc	27 a	-27	52 a	-76	67 bc	-233	72 bc	-153
Untreated	-----	26 a	35 a		48 a		33 d		46 cd	

^{a/} Application dates: 8-7, 8-14, and 8-28-79. Pretreatment count taken 8-7-79.^{b/} Values followed by the same letter within a column are not significantly different (P=0.05) according to Duncan's new multiple range test.^{c/} Percent mortality adjusted for natural decrease by Henderson and Tilton's formula.^{d/} Application dates: 3-28, 4-4, and 4-18-79. Pretreatment count taken 3-28-79.

TABLE 2. Evaluation of Miticides for Control of *Tetranychus cinnabarinus* on *Brassica actinophylla*.

Chemical	Rate A.I./ 100 liters	Avg. No. Female mites/plant Pretreatment	Average no. female mites/plant and %control at indicated day							
			7				post initial treatment			
			No. mites	%control	No. mites	%control	No. mites	%control	No. mites	%control
			Test 3 ^{a/}							
			14				21			
			28							
Butocarboxime	0.03kg	10 ^{b/} _a	8 b	59 ^{c/}	9 b	64	36 a	1	37 a	11
Butocarboxime	0.06kg	16 a	3 b	91	4 b	90	19 b	65	21 b	67
Butocarboxime	0.12kg	9 a	2 b	87	2 b	92	19 b	38	25 b	28
Butocarboxime	0.24kg	10 a	3 b	87	2 b	90	20 b	43	22 b	46
Malathion 55	0.18kg	6 a	3 b	76	5 b	61	16 b	20	23 b	-1
Untreated	-----	12 a	22 a		27 a		40 a		46 a	
			Test 4 ^{d/}							
Oxamy1	0.03kg	18 a	9 b	54	9 b	76	4 c	91	1 b	99
Oxamy1	0.06kg	25 a	10 b	60	4 b	93	7 bc	88	1 b	99
Oxamy1	0.12kg	22 a	4 b	84	3 b	94	1 c	98	0 b	100
Oxamy1	0.24kg	17 a	2 b	89	6 b	82	1 c	97	0 b	100
Malathion 55	0.18kg	13 a	8 b	43	6 b	78	23 b	24	0 b	100
Untreated	-----	19 a	20 a		41 a		44 a		60 a	

a/ Application dates: 7-9, 7-16, and 7-30-79. Pretreatment count taken 7-9-79.

b/ Values followed by the same letter within a column are not significantly different (P=0.05) according to Duncan's new multiple range test.

c/ Percent mortality adjusted for natural decrease by Henderson and Tilton's formula.

d/ Application dates: 6-4, 6-11, and 6-25-79. Pretreatment count taken 6-4-79.

TABLE 3. Evaluation of Miticides for Control of Tetranychus cinnabarinus on Codiaeum variegatum (Test 5) and Epipremnum aureum (Test 6).

Chemical	Rate A.I./ 100 liters	Avg. No. Female mites/plant Pretreatment	Average no. female mites/plant and %control at indicated day							
			post initial treatment							
			7	14	21	28				
			No. mites %control	No. mites %control	No. mites %control	No. mites %control	No. mites %control	No. mites %control	No. mites %control	No. mites %control
			Test 5 ^{a/}							
Oxamy1	0.03kg	5 ^{b/}	0 b	100 ^{c/}	0 b	100	2 b	88	0 b	100
Oxamy1	0.06kg	9 b	0 b	100	0 b	100	2 b	95	0 b	100
Oxamy1	0.12kg	19 ab	0 b	100	0 b	100	1 b	99	0 b	100
Oxamy1	0.24kg	18 ab	0 b	100	0 b	100	0 b	100	0 b	100
Diazinon 2 EC 0.06kg	25 a		0 b	100	0 b	100	2 b	99	0 b	100
Untreated	-----	13 ab	32 a		36 a		43 a		39 a	
			Test 6 ^{d/}							
Oxamy1	0.03kg	5 a	6 ab	-2	5 ab	39	7 b	21	0 b	100
Oxamy1	0.06kg	4 a	1 bc	84	1 b	93	1 c	93	0 b	100
Oxamy1	0.24kg	4 a	0 c	100	0 b	100	0 c	100	0 b	100
Malathion 55 0.18kg	5 a		7 a	-40	8 a	-2	5 bc	41	3 b	69
Untreated	-----	7 a	7 a		11 a		12 a		16 a	

a/ Application dates: 8-6 and 8-27-79. Pretreatment count taken 8-6-79.

b/ Values followed by the same letter within a column are not significantly different (P=0.05) according to Duncan's new multiple range test.

c/ Percent mortality adjusted for natural decrease by Henderson and Tilton's formula.

d/ Application dates: 4-24, 4-30, and 5-14-79. Pretreatment count taken 4-23-79.

Each treatment was replicated 4 times in a randomized block experimental design with 2 plants/rep. Effectiveness of the pesticide was determined by counting the number of female mites per plant before initial treatment and every 7 days afterwards until termination of the test (28 days). The mite counting method was based on research of Jeppson (1951) and Ehler (1974), which indicates that density of females per plant is a sufficient index of population trends. Percent control due to treatment was derived according to Henderson and Tilton's (1955) modification of Abbott's formula. Phytotoxicity ratings on a scale from 1 (0-10% plant damage) to 10 (91-100% plant damage) were taken weekly in each test.

Plants utilized in these tests were grown in 7.62 cm plastic containers filled with premixed potting soil. A commercial controlled-release fertilizer was applied every 4 months, and plants were irrigated twice a week. Plants were maintained on 0.92m raised greenhouse benches at temperatures ranging from 15° to 36°C and relative humidities of 40 to 100%.

RESULTS AND DISCUSSION

Populations of T. cinnabarinus on Dieffenbachia 'Exotica' were significantly reduced following initial application of butocarboxime at 0.03 and 0.24 kg A.I./100 liters (Table 1). However, 2 additional applications of all rates of butocarboxime did not provide any definite pattern of mite control. Mite population increases were noted in all treatments. Butocarboxime at 0.24 kg A.I./100 liters did exhibit the highest overall % control. Tetranychus cinnabarinus populations also were not effectively controlled on Dieffenbachia with oxamyl (Table 1). Populations increased throughout the test period despite additional treatments. These results indicate that adequate control of established mite populations on Dieffenbachia with the test miticides would be difficult at best.

Butocarboxime at all rates except 0.03 kg A.I./100 liters significantly suppressed T. cinnabarinus populations on Brassaia actinophylla Endl throughout the span of the test period (Table 2). The 3 remaining rates of butocarboxime and malathion were equally effective but did not provide adequate control of the pest. Populations increased despite 3 pesticide applications. In contrast, all rates of oxamyl significantly reduced mite populations on B. actinophylla after initial treatment (Table 2). Also, each additional pesticide application further reduced the populations, and mites were eliminated by 0.12 and 0.24 kg A.I./100 liters.

All rates of oxamyl, as well as malathion, eliminated T. cinnabarinus on Codiaeum variegatum (L.) after initial pesticide application (Table 3). No applications were needed 14 days afterwards indicating good residual action. A minor mite reinfestation occurred 21 days after initial treatment on all plants but those exposed to 0.24 kg A.I./100 liters. Mites were again eliminated after treatment.

Tetranychus cinnabarinus populations were reduced on Epipremnum aureum (Linden and Andre) after initial treatment of oxamyl at 0.06 and 0.24 kg A.I./100 liters (Table 3). Subsequent applications eventually eliminated the pest on all plants.

No plant phytotoxicity was noted during any test. Except Dieffenbachia, all plants treated with the 3 highest rates of oxamyl were marketable.

In conclusion, neither butocarboxime or oxamyl effectively controlled T. cinnabarinus on Dieffenbachia. Control of the pest on this plant is increasing in difficulty, either because of pesticide resistance or host plant neutralization of the chemical. Butocarboxime did not provide adequate mite

control on B. actinophylla and did not appear to be an effective miticide in these experiments. Oxamyl provided good control on the remainder of the plants tested. The results of these studies are in agreement with results obtained by Hamlen and Wettstein (1978). Some mite reinfestation can be expected with oxamyl, but they can be eliminated with additional treatments. Oxamyl could be used in a greenhouse mite control program if applied properly at recommended concentrations on species tested except Dieffenbachia.

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BIOLOGY OF PHYLLOPHAGA CRINITA^{1/} (BURMEISTER)
IN LOWER RIO GRANDE VALLEY SUGARCANE^{2/}

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ABSTRACT

The biology and movement of larval stages of *Phyllophaga crinita* (Burmeister) reared on sugarcane were determined in boxes of soil during 1975-76. Most larvae completed the 1st and 2nd stages within 48 days after adult females were introduced into the soil. During the 1st larval instar, the insects moved downward, movement upwards toward the cane occurred during later stages of the 2nd instar. Larvae remained around the cane stools during the 3rd larval instar, finally stopped feeding and began to build earthen cells during late October 1975. Most were pupae (63.8%) and adults (33.6%) on the last sample date, Apr. 31, 1976.

INTRODUCTION

In the Lower Rio Grande Valley of Texas, sugarcane provides a favorable habitat for *Phyllophaga crinita* (Burmeister) larvae. Damaging populations have occurred in several sugarcane fields since 1970. Economic losses have resulted from stunting of plant growth and lodging. This damage has necessitated replanting of what would normally be a 4-5 yr. ratoon crop.

P. crinita larvae have been recorded as pests on a number of field crops and ornamentals throughout Texas (Frankie et al. 1974, Huffman et al. 1976, Reinhard 1940, Teetes 1973, Teetes et al. 1976). Reinhard (1940), in laboratory studies, found the average duration of the egg, first, second, and third larval instar and pupal stage to be approximately 13, 34, 35, 213, and 20 days, respectively. Several insecticides have provided effective control of larvae on grain sorghum, wheat, and sugarcane in recent years (Fuchs et al. 1974, Huffman et al. 1976, Teetes 1973 and 1975).

Application time and placement of insecticide are critical for effective control of the larvae. Frankie et al. (1973) reported that the pests were most effectively controlled in turf while in the 1st and 2nd larval instars. This study was conducted to document the movement in soil and development of *P. crinita* larvae on sugarcane under South Texas conditions.

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METHODS AND MATERIALS

Phyllophaga crinita was reared in the field in 90 boxes (61 cm³) constructed of Masonite siding. Sugarcane seed pieces were planted in Hidalgo sandy loam soil in the middle of each box during January, 1975. Adult females were collected on Apr. 15, 1975 from a black light trap and then placed individually in holes ca. 10 cm deep that were made with a pencil in the soil at each quarter section of a box. Four females were placed in each box. Cane plants were ca. 15 cm high when females were introduced.

Weekly observations of 5 randomly selected boxes per date were made during the period Apr. 22 through June 23, 1975. Less than 2.5 cm of soil from each box quarter was examined at one time so that each box was carefully sampled. After June 23, the sampling interval was increased and similar observations were made on Aug. 25, Oct. 25, Jan. 23, 1975 and Apr. 30, 1976. Since the boxes were destroyed during sampling, none was sampled twice. The developmental stages and locations of all larvae were recorded for each box sampled; eggs and head capsules were measured with an ocular micrometer.

RESULTS AND DISCUSSION

Larval Development--Data on the development of egg and larval stages are shown in Fig. 1. Most eggs were deposited and hatched within 2 weeks after adult females were introduced. Since 1st stage larvae were found on the first sample date, it was apparent that less than 7 days were required for eggs to hatch. Most 1st stage larvae had molted within 34 days of oviposition; however, a few remained in the 1st larval instar through October. Second stage larvae were collected from May 12, 1975 through Apr. 30, 1976; the largest percentage (96.6%) was found on May 19. Some 3rd stage larvae (2.3%) were found on May 26, but most did not develop to the 3rd instar until June 2, 1975.

Most individuals were either pupae or adults (63.8% and 33.6%, respectively) on Apr. 30, 1976 (Fig. 1). Two percent of the individuals collected were still in the 2nd instar while 0.6% were in the 3rd instar. This supports Teetes et al. (1976) statement that some *P. crinita* may require a developmental period longer than 1 yr.

Measurements of eggs, and of larval head capsules for each instar, are listed in Table 1. Eggs were oval shaped. Head capsule sizes were distinctly different for each instar.

TABLE 1.--Egg and Larval Head Capsule Measurements for *P. crinita*, Weslaco, Texas, 1975-76.

Developmental Stage	No.	Mean \pm SE (mm).
Egg	28	2.73 \pm 0.04 x 2.04 \pm 0.07
1st instar larvae	93	1.81 \pm 0.02
2nd instar larvae	40	3.25 \pm 0.02
3rd instar larvae	50	5.13 \pm 0.01

Distribution--The mean depths in the soil found for larvae on each sample date are shown in Fig. 2. Individuals collected on Apr. 22, 1975 were located at a mean depth of 26.5 \pm 0.9 cm. Two weeks later the 1st

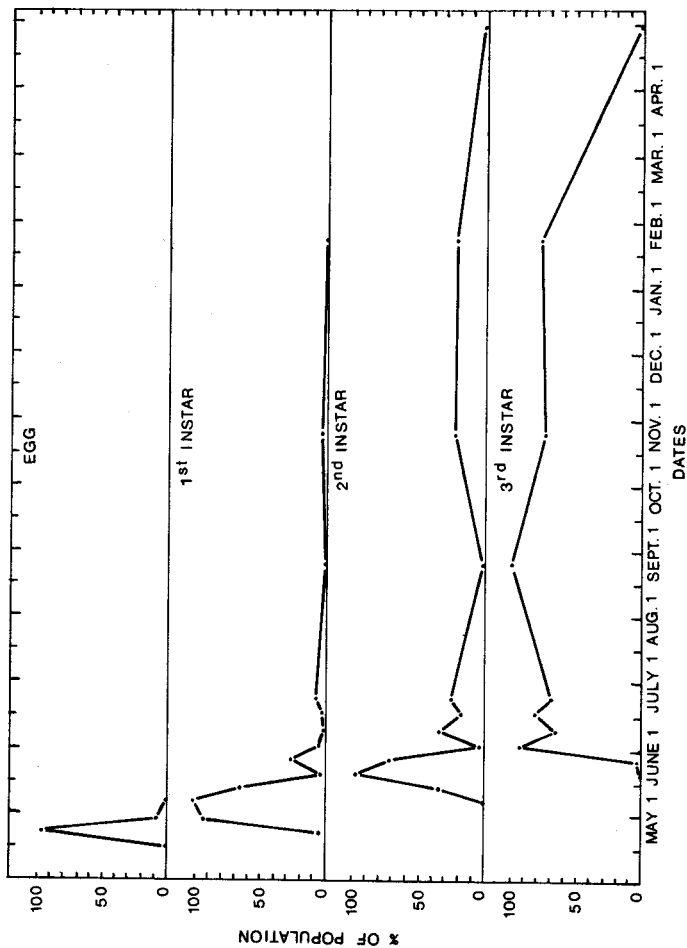


Fig. 1. Percent of the total number of larvae in each developmental stage on each sample date.

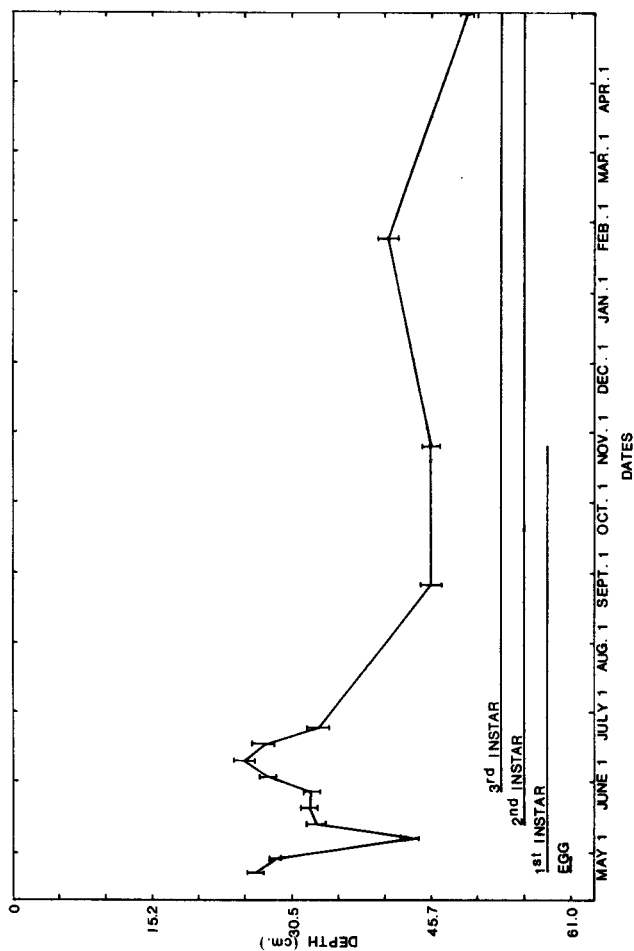


Fig. 2. Mean depth \pm SE of larvae on each sample date.

stage larvae had moved downward to a depth of 43.4 ± 1.0 cm. The larvae began to move upward a week later and continued to remain between depths of 33.2 cm and 25.4 cm until Aug. 25 when most were found at a mean depth of 45.7 ± 1.1 cm. Throughout the remainder of the sampling period, the mean depth was below 40.8 cm. Results in Table 2 reflect this same upward and downward grub movement and also show the percent of the larvae at various depths in the soil.

TABLE 2.--Percent of the Total Number of Larvae Located at Various Depths on Each Sample Date, Weslaco, Texas, 1975-76.

Date		Depth (cm.)				
		0-7.6	7.6-15.2	15.2-30.5	30.5-45.7	45.7-61.0
Apr.	22	0.0	13.8	65.4	20.1	0.6
	28	0.0	3.1	70.5	24.0	2.3
May	6	0.0	5.7	19.6	16.1	58.7
	12	0.8	15.5	41.0	14.7	27.9
	19	6.7	20.6	24.2	23.1	25.3
	26	31.0	19.0	15.8	14.6	19.6
June	2	10.0	25.1	26.8	19.2	18.9
	9	10.4	11.8	29.4	20.3	27.7
	16	13.4	26.5	24.9	16.2	18.9
	23	13.2	11.1	23.9	21.1	30.7
Aug.	25	3.9	2.6	7.1	22.7	63.6
Oct.	24	3.1	2.5	9.3	23.5	61.7
Jan.	23	2.5	8.2	12.6	34.0	42.8
Apr.	30	0.7	0.0	4.7	26.2	68.5

TABLE 3.--Percent of the Total Number of Larvae Located at Various Distances From Row Center on Each Sample Date, Weslaco, Texas, 1975-76.

Date		Distance (cm.)			
		0-7.6	7.6-15.2	15.2-22.9	22.9-30.5
Apr.	22	21.4	27.0	31.4	20.1
	28	34.1	25.6	35.7	4.7
May	6	33.9	23.5	23.9	18.7
	12	36.3	29.5	25.1	9.2
	19	33.3	26.7	18.6	21.4
	26	31.6	26.9	19.6	21.9
June	2	43.1	27.7	19.5	9.7
	9	45.3	19.0	17.9	17.9
	16	39.5	26.1	14.6	19.8
	23	42.9	22.5	16.4	18.2
Aug.	25	33.8	28.6	20.1	17.5
Oct.	4	44.4	25.3	11.1	19.1
Jan.	23	44.0	27.0	17.0	11.9
Apr.	30	41.6	32.2	12.8	13.4

A comparison of Tables 2 and 3 indicates that most individuals on Apr. 22 were evenly distributed away from the cane row center between depths of 15.2 cm and 45.7 cm. On May 6, greater than 50% of the larvae collected were below the depth of 45.7 cm but had moved slightly toward the row center. Larvae began to move upwards more towards the cane row by May 19 and some were found feeding around the cane stools. Approximately 32% of the larvae collected on May 26 were found within 7.6 cm of the soil surface with greater than 58% within 15.2 cm of row center. At this time, most were still in the 2nd larval instar with a few in the 3rd larval instar.

Gerard and Hipp (1975) found that greater than 60% of sugarcane roots develop above 30.5 cm with usually more than 45% of these located in the bed area rather than between bed and furrow and in-furrow. Our data indicated that the larvae were moving into this more concentrated root area around the cane stool until late in the growing season. Most larvae on Aug. 25 were still located near the row center; however, 86.3% had moved below 30.5 cm (Tables 2 and 3). Greater than 75% were located below this depth on the remaining sample dates. Third stage larvae had begun to form earthen cells on Oct. 24. They were also clearing the abdomen of fecal material, an indication that feeding had been discontinued. On Jan. 23, 1976 most of the 3rd stage larvae were found in earthen cells with their abdomens clear of fecal material.

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EFFECT OF TEMPERATURE ON THE DEVELOPMENT AND SURVIVAL OF THE
IMMATURE STAGES OF BRACON PLATYNOTAE^{1/}, A NATIVE PARASITE OF
PECTINOPHORA GOSSYPIELLA (LEPIDOPTERA: GELECHIIDAE)^{2/}, ^{3/}

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ABSTRACT

In the laboratory, Bracon platynotae (Cushman), a parasite of the pink bollworm, Pectinophora gossypiella (Saunders), proved to have a total development time (\pm S.D.) on the pink bollworm from 18.0 ± 0.7 days at 20° C to 6.8 ± 0.5 days at 35° C. Development was slightly faster on the beet armyworm, Spodoptera exigua (Hübner): 17.5 ± 0.7 days at 20° C to 6.8 ± 0.5 days at 35° C. The percentage of parasites completing development was 27-29% lower on beet armyworms than on pink bollworms at the temperature extremes of 20° and 37.8° C. However, survival was generally good on both hosts within the temperature range of 25 - 35° C. Some successful development also occurred at 37.8° C, indicating that the immature stages of B. platynotae are adapted to the high temperatures that occur in the southwest desert areas.

INTRODUCTION

The genus Bracon contains several species of wasps that are externally parasitic on the pink bollworm, Pectinophora gossypiella (Saunders), and have been tested in the United States as biological control agents of this important insect pest. Bryan et al. (1971; 1973) discussed the use of B. kirkpatricki (Wilkinson), an African species, in Arizona, and McGough and Noble (1955; 1957) summarized colonization attempts with B. gelechiae (Ashmead), an Indian species, for control of pink bollworms in Texas. Also, Rude (1937) recorded B. platynotae (Cushman), a native species, as probably the most abundant parasite of pink bollworms in the Laguna District of northern Mexico, observed parasitization of the hosts in both squares and bolls, and suspected that B. platynotae was a factor in control of long-cycle pink bollworms while they were in their overwintering cells. Additional hosts include Lineodes interrupta (Zeller), Pachyzancia periusalis (Walker), Platynotae stultana (Walsingham) (Muesebeck et al. 1951; Krombein, 1958; Krombein and Burks, 1967 and Isophrictis sp. (Rude, 1937). The known distribution of B. platynotae includes California, Arizona, Texas, Louisiana, Georgia, and northern Mexico.

METHODS

A colony of B. platynotae was started from insects reared from parasitized pink bollworms collected in a cotton field near Tucson, AZ. The colony was maintained in the laboratory for 2 years on pink bollworms and beet armyworms by using the rearing technique described for B. kirkpatricki (Bryan et al. 1969). The beet armyworm, Spodoptera exigua (Hübner), has not been recorded previously as a host of B. platynotae, but is readily accepted in the laboratory.

1/ Hymenoptera: Braconidae.

2/ Lepidoptera: Gelechiidae.

3/ In cooperation with the Arizona Agricultural Experiment Station, Tucson, AZ 85721.

For the more precise rearing required in our study of the developmental stages, we used the smaller cage described by Bryan et al. (1971). Thus the opening of a 17-ml clear plastic cup (43 mm diam) was covered with nylon organdy, and 1 female wasp was introduced through a hole in the bottom. Then a 35 x 6 mm screw-top vial filled with a 20% sugar solution and plugged with a piece of cellulose sponge was inserted through the hole to provide food for the wasp. A 4th or 5th-stage host larva was then placed in the top half of a 50 x 8 mm clear plastic petri dish and covered with a single layer of tissue. The cage with the wasp was inverted over the larva, so the parasite could sting it through the organdy and tissue. After a 2-hr exposure period the cage was removed and the number of eggs deposited on the larva was recorded. Finally the bottom half of the petri dish was fitted into place so the parasitized larva was held in place between the tissue and the petri dish top. In this way, the development of the parasites could be observed through the clear plastic top.

RESULTS AND DISCUSSION

General Observations--Male and female parasites usually emerged from cocoons on the same day if parasitized hosts were held at a room temperature of 26.7° C. Mating commenced within the 1st few hours and recurred frequently for several days. Unmated females produced only male progeny. However, these virgin females were observed to mate when they were 17-19 days old and thereafter produced female progeny. Mating lasted only a few seconds, but the female had a preoviposition period of 3-4 days, which was followed by an oviposition period of as much as 2 months. As with *B. kirkpatricki* (Bryan et al., 1971) the actual deposition of the eggs was preceded by the stinging and paralyzing of the host. Individual females laid several eggs on 1 host larva, and it was not unusual for as many as 8 parasites to develop on 1 pink bollworm larva.

For ca. 2/3 of the larval stage, the developing *B. platynotae* fed on the host; then it left the host, crawled a short distance, and attempted to wedge itself in available small crevices where spinning of the cocoon began. However, if the larva was not satisfied with the 1st choice of site, or if it was disturbed, it wandered about the rearing container and finally pupated naked. In such cases, completion of development was not nearly as successful as when a proper cocoon was spun.

Developmental Times--The average total developmental times (\pm S. D.) for the parasite on the pink bollworm (Table 1) ranged from 18.0 \pm 0.7 days at 20° C

TABLE 1. Length of Stages of *B. platynotae* on 2 Hosts.

Rearing temp. $\pm 1.1^{\circ}$ C	No. parasites observed	Duration in days (\pm SD)			
		Egg stage	Larval stages	Pupal stage	Total development period
Pink Bollworms					
20	114	2.6 \pm 0.4	6.6 \pm 0.8	8.7 \pm 0.6	18.0 \pm 0.7
25	111	2.0 \pm .0	4.0 \pm .2	5.3 \pm .4	11.3 \pm .5
30	166	<1.0	3.0 \pm .2	3.8 \pm .4	7.8 \pm .4
32.2	179	<1.0	3.0 \pm .0	3.0 \pm .2	7.0 \pm .2
35	137	<1.0	2.9 \pm .3	2.9 \pm .4	6.8 \pm .5
37.8	49	<1.0	3.0 \pm .4	3.2 \pm .4	7.2 \pm .5
Beet Armyworms					
20	90	2.4 \pm 0.5	6.5 \pm 0.9	8.6 \pm 0.6	17.5 \pm 0.7
25	159	2.0 \pm .0	3.8 \pm .5	5.3 \pm .4	11.0 \pm .3
30	138	1.0 \pm .1	3.0 \pm .2	3.7 \pm .5	7.7 \pm .5
32.2	121	<1.0	3.0 \pm .2	3.1 \pm .3	7.1 \pm .3
35	104	<1.0	2.8 \pm .5	3.0 \pm .4	6.8 \pm .5
37.8	6	<1.0	3.3	3.8	8.2

to 6.8 ± 0.5 days at 35°C , but the average times differed only ca. 1 day for temperatures between 30 and 37.8°C . The egg plus the larval periods took slightly more than $1/2$ the total developmental period. The time required for development of the various stages was similar for both sexes.

The developmental periods of the parasite on the beet armyworm (Table 1) and the pink bollworm at 20 , 25 , and 30°C were not significantly different. On the beet armyworm, the time for total development ranged from 17.5 ± 0.7 days at 20°C to 6.8 ± 0.5 days at 35°C . Again the sex of the parasite had little influence on developmental time.

Survival--Survival of the parasite on the pink bollworm was high at temperatures of 20 – 32.2°C , dropped slightly at 35°C , and dropped precipitously at 37.8°C (Table 2). Maximum survival (95.2%) occurred at 32.2°C .

TABLE 2. Survival of B. platynotae When Reared on 2 Hosts^{a/}

Rearing temp. $\pm 1.1^{\circ}\text{C}$	No.				% Survival egg to adult
	Eggs	Larvae	Cocoons	Adults	
<u>On pink bollworm</u>					
20	123	121	116	114	92.7
25	118	118	114	111	94.7
30	177	177	171	166	93.8
32.2	188	186	181	179	95.2
35	159	158	138	137	86.2
37.8	144	133	58	49	34.0
<u>On beet armyworm</u>					
20	138	134	90	90	65.2
25	171	170	161	159	93.0
30	158	154	141	138	87.0
32.2	141	138	124	121	85.8
35	138	134	110	105	76.1
37.8	135	123	12	6	4.4

a/ Ca. 40 host larvae parasitized at each temperature.

Survival on the beet armyworm was lower and more variable; however, 93% survived at 25°C . The very low survival (4.4%) of the parasite on the beet armyworm at 37.8°C resulted because of the rapid breakdown of host tissue at that temperature (the tissue usually became liquid, turned black, and dried up before the parasite larvae finished feeding). Similarly, at 20°C , survival was low (65.2%), again because the host tissue broke down. At temperatures ranging from 20 to 37.8°C , most mortality on both hosts occurred early in the larval stage. At 37.8°C , ca. 8% of the eggs failed to hatch, and at a constant 41.1°C , this figure rose to 100% .

The high survival rates at 35°C and the limited survival at 37.8°C indicate the adaptiveness of B. platynotae to the high temperatures that occur in the southwestern desert region of the United States. Successful development of the immature stages compares favorably with the results obtained by Bryan et al. (1971) for B. kirkpatricki.

Sex Ratio--The ratio of male to female parasites was consistently greater for parasites produced on the beet armyworm than for those produced on the pink bollworm (Table 3). The ratios for the beet armyworm ranged from $3.1:1.0$

TABLE 3: Sex Ratio of B. platynotae Reared on 2 Hosts

Rearing temp. $\pm 1.1^{\circ}\text{C}$	Parasites on pink bollworms			Parasites on beet armyworms		
	No. ♂	No. ♀	Ratio	No. ♂	No. ♀	Ratio
20	59	55	1.1:1.0	68	22	3.1:1.0
25	60	51	1.2:1.0	95	64	1.5:1.0
30	91	75	1.2:1.0	88	50	1.8:1.0
32.2	100	79	1.3:1.0	86	35	2.5:1.0
35	77	60	1.3:1.0	65	40	1.6:1.0
37.8	25	24	1.0:1.0	1	5	

at 20°C to 1.5:1.0 at 25°C ; at all higher temperatures, the ratios were intermediate in value. The trend was the same as that for survival (Table 2) except at 35°C , indicating a differential survival of the sexes on this host. The ratios for the pink bollworm ranged from only 1.0:1.0 to 1.3:1.0 and showed no apparent trend except that the more equal ratios occurred at either extreme in temperature.

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TITERS OF 20-HYDROXYECDYSONE IN BOLL WEEVIL PUPAE

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ABSTRACT

Pupae of the boll weevil, Anthonomus grandis Boheman, were found to have a maximum of 20-hydroxyecdysone 48 hrs after the prepupal stage. No α -ecdysone was detected. A simplified high performance liquid chromatography (HPLC) method for analysis of ecdysones was developed.

INTRODUCTION

Rapid progress been made in microdetermination of ecdysteroids titers in insect development since bioassays as used by Shaaya and Karlson (1965) in the blowfly, Calliphora erythrocephala Meigen (= C. vicina Robinson-Desvoidy). Morgan and Poole (1976) summarized the analytical methods available as being thin-layer chromatography, optical spectroscopy, radioimmunoassay, gas-liquid chromatography, mass fragmentography, and HPLC. The HPLC technology can be used for both purification and analysis and provides a rapid microdetermination of both ecdysone and 20-hydroxyecdysone in extracts of whole insects (Holman and Meola 1978, O'Neill et al. 1977). This investigation was undertaken to determine the presence of ecdysteroids in pupae of the boll weevil, Anthonomus grandis Boheman by HPLC.

MATERIALS AND METHODS

Ecdysteroid assays were made on pupae at closely synchronized ages. At 5 days post egg implantation in diet trays, the larvae, prepupae, and pupae were washed from the larval rearing diet. The age of each sample was determined with respect to the time of prepupal formation from last instar larvae. Accurately timed samples were taken at 0 hr, and then at 4 hr intervals through 24 hrs and at 8 hr intervals thereafter through pharate adult stages at 64 and 72 hrs. In a modification of the procedure of Holman and Meola (1978) the 4 to 7 g (400-700 weevils) samples were ground for 1 min in 50 ml acetonitrile with a Willems Polytron homogenizer and stored at 4°C.

The samples were filtered in a Buchner funnel and the retained material was rinsed three times with acetonitrile. The filtrate was evaporated at 60°C to dryness under reduced pressure and the dry material was taken up in 20 ml of acetonitrile. Two 10 ml aliquots were removed and one was spiked with 25 ng ecdysone and 20-hydroxyecdysone. The aliquots were partitioned 3 times against twice the volume of n-hexane in a separatory funnel. The acetonitrile layer was removed and evaporated to dryness under reduced pressure. The dry material was taken up in 1.0 or 0.5 ml of methanol. The samples were filtered through a Swinney adaptor 4.5 micron filter before injecting 10 μ l aliquots on a Waters Associates reverse phase μ bondapak C₁₈ HPLC column (3.9 mm i.d. x 30 cm) in a Waters Associates HPLC. The eluting solvent was 18% v/v acetonitrile in H₂O at a flow rate of 2 ml/min. UV detection was at 254 nm. Quantitation was by measurement of peak height with a standard curve and ecdysone as an internal standard. The minimum detectable quantity of 20-hydroxyecdysone was 5 ng. The retention time for 20-hydroxyecdysone was 5 min and for ecdysone was 12

min. Samples were compared to standards and spiked with standard before homogenization, filtration, and evaporation. To confirm the HPLC peak identity, samples were collected for determination on a Hewlett-Packard 5930A mass spectrometer. The samples were evaporated to dryness at reduced pressure, dissolved in methanol, and placed on the solid probe. The spectra were compared to a standard run on the same instrument with the same conditions.

RESULTS AND DISCUSSION

No detectable levels of ecdysone or 20-hydroxyecdysone were found in the first 20 hrs after pupariation. However, at 24 hr an appreciable titer which was obtained in three experiments of 20-hydroxyecdysone was identified, as can be seen in Fig. 1. The level of 20-hydroxyecdysone rose to a maximum consistently at 48 hr after pupariation in 6 separate experiments. In addition, hemolymph was extracted from samples at this time interval and 20-hydroxyecdysone was determined to be present also within the hemolymph. The titer of 20-hydroxyecdysone rapidly diminished after 48 hr and only trace amounts could be detected up to 64 hr or immediately before the pharate adults at 72 hr. No 20-hydroxyecdysone was observed in the pharate adults at the 72 or 80 hr intervals. The pattern in Fig. 1 was consistently obtained in all experiments, however, each group of boll weevils had a different quantity of 20-hydroxyecdysone within the pattern. The quantity of 20-hydroxyecdysone at 48 hrs. was averaged over 6 experiments to give 600 ± 40 ng/g.

Although ecdysone is known to be a precursor for 20-hydroxyecdysone no trace was found in any sample. Every sample assayed was tested for recovery with a spiked portion of that sample. The recoveries averaged 95% and no sample was used unless the recovery was above 90%. The rapid extraction process used for these samples was verified to have 95% average recovery with samples spiked with 20-hydroxyecdysone before the weevils were ground in the homogenizer.

The titer of 20-hydroxyecdysone is expressed graphically in Figure 1. The developmental events indicate that the titer of 20-hydroxyecdysone consistently peaks at a given time (48 hrs) as pupal metamorphosis to adult occurs. Our data for 20-hydroxyecdysone peaks is unlike that found in Stomoxys calcitrans (L.) where 2 peaks of 20-hydroxyecdysone appeared in the pharate pupae (O'Neill et al. 1977). Hodgetts et al. (1977) found in Drosophila melanogaster Meigen that the titer of the hormone was greatest at 38 hrs after puparium formation and occurred just prior to secretion of adult cuticle. Although we did no adult cuticle determination, we observed that darkening of the entire pupae occurred immediately after the 48 hr period and perhaps this may be correlated with the 20-hydroxyecdysone activity and increased activity of dopa decarboxylase.

ACKNOWLEDGEMENT

We are indebted to Drs. A. C. Thompson and Paul A. Hedin of the Chemistry Research Unit, this laboratory, for determinations with the mass spectrometer, Mrs. Donna Kellum of the Sterility Section of this laboratory, for technical assistance, and G. M. Holman of USDA, College Station, TX contributed the 20-hydroxyecdysone standards.

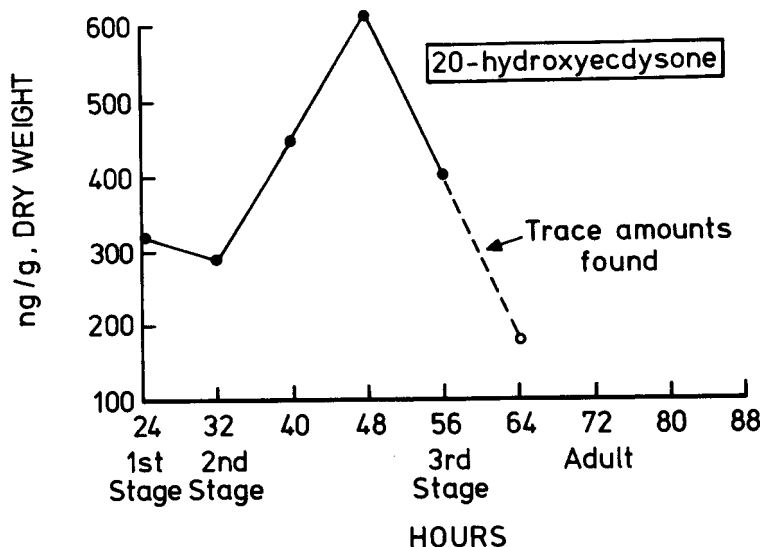


FIG. 1. Analysis of 20-hydroxyecdysone titers from 24 to 80 hr after pupariation of Anthonomus grandis.

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CONSTITUTION OF
SOUTHWESTERN ENTOMOLOGICAL SOCIETY

Article I. Name

This corporation, chartered under the laws of Texas in the name and style of the "Southwestern Entomological Society," herein and after called the "Society," is formed as an educational institution, not contemplating financial gain or profit.

Article II. Purpose

The object and purpose of the Society is to foster entomological accomplishment among its members and to promote the science of Entomology through the encouragement of: (1) association and free discussion among all entomologists; (2) the preparation, reading, and publication of papers; (3) the dissemination of entomological information to the general public; and (4) publication of the Southwestern Entomologist.

Article III. Membership

Section 1. - Membership: Membership shall be open to all persons interested in entomology.

Section 2. - Procedure to Obtain Membership: Any person desiring to become a member of the Society shall do so by application to the Secretary-Treasurer. A person shall become such member upon the approval of a majority of the Executive Committee and the payment of such dues as may be established by the Executive Committee.

Section 3. - Member in Good Standing: One who is current in payment of dues.

Article IV. Membership Rights

Section 1. - Voting: Each member in good standing shall be entitled to one vote at any regular or special meeting. Voting by proxy shall not be allowed.

Section 2. - Privileges: All members in good standing shall have equal privileges as to presentation of papers and discussion at meetings.

Section 3. - Journal: Each member in good standing shall be entitled to receive as often as published a copy of the Southwestern Entomologist and any other Society publications.

Section 4. - Termination of Membership: Upon the cessation of membership of any member of the Society at any time and for any reason or cause, all rights, title, and interest in and to any and all of the Society's assets shall automatically terminate.

Article V. Membership Certificates

Section 1. - Certificates: Each member in good standing shall be entitled to receive such evidence of membership as may be decided upon by the Executive Committee.

Section 2. - Transfer: Membership in the Society shall not be transferable or assignable.

Article VI. Dues

Section 1. - Annual Dues: The annual dues for membership in the Society shall be such amount as may be established by the Executive Committee from time to time.

These dues shall be kept in a fund for use by the Secretary-Treasurer for expenses incurred in publishing and distributing the journal as well as normal expenses of the Society.

Section 2. - Time of Payment: The Executive Committee shall set such times during each year as it deems advisable for the payment of annual dues by members. The name of a member more than one year in arrears in payment of dues shall be dropped from the roll, provided two notices of indebtedness shall have been mailed to him, and such members shall have no further rights, title, or interest in the Society as provided for by Article IV, Section 3, of this Constitution.

Article VII. Meetings of Society

Section 1. - Annual Meetings: The Society shall hold annual meetings at such times and places as may be designated by the Executive Committee and specified in the notice thereof, for the purpose of conducting such business as may be properly brought before the meeting.

Section 2. - Registration Fee: A registration fee, in an amount to be determined by the Executive Committee, shall be paid at each annual meeting by all members and non-members who attend.

Section 3. - Special Meetings: Special meetings of the Society shall be held at any time at such place as may be specified in the waiver or notice thereof, whenever called by the President or any two or more members of the Executive Committee.

Section 4. - Notice: Notice of all meetings of the Society, annual and special stating the time, place, and agenda shall be mailed to each member by the President, Secretary-Treasurer, or Officer calling the meeting not less than 7 days prior to the meeting.

Article VIII. Officers

Section 1. - Officers: The officers of the Society shall consist of a President, President-Elect and Secretary-Treasurer, all of whom, except the President, shall be elected by and from the membership by a plurality vote of those present at the first regular annual meeting or at the direction of the Executive Committee. The first President and a member of the Executive Committee to represent the Past-President of the Society shall be elected by and from the membership at the organizational meeting for a term extending to the next annual meeting. Thenceforth, the President-Elect shall automatically accede to the office of President at each annual meeting, or should the President be unable or unwilling to act for any reason. Nominees for such elective offices of the Society shall be selected by a Nominating Committee of three members appointed by the President. Nominations may also be presented from the floor. The President and President-Elect shall hold office from the date of election at the annual meeting until the election of their successors at the next annual meeting and shall not be eligible for re-election to the same office for a successive term. The Secretary-Treasurer shall hold office from the date of election at the annual meeting until election of their successors at the third following annual meeting and shall be eligible for re-election. No member may occupy more than one office at any one time.

Section 2. - Powers of President: The President shall be the chief executive officer of the Society and shall preside at all meetings of the Society and Executive Committee, have and exercise general and active management of the Society, execute and enforce all orders and resolutions and regulations duly adopted by the Executive Committee, execute all contracts in the name of and on behalf of the Society, and perform such other duties as assigned by the Executive Committee.

Section 3. - Powers of President-Elect: In the absence of the President, or in case of his failure to act, the President-Elect shall have all of the powers of the President and shall perform such other duties as shall from time to time be imposed upon him by the Executive Committee.

Section 4. - Powers of Secretary-Treasurer: The Secretary-Treasurer shall attend and keep the minutes of all meetings of the Executive Committee, shall have charge of the records and seal of the Society, and shall, in general, perform all of the duties incident to the office of Secretary-Treasurer of the Society. The Secretary-Treasurer shall keep full and accurate accounts of receipts and disbursements on the books of the Society and shall deposit all monies and other valuable properties and effects in the name of and to the credit of the Society in such depository or depositories as may be designated by the Executive Committee. The Secretary-Treasurer shall disburse funds as may be ordered by the Committee, taking proper vouchers for such disbursements; and shall render to the Executive Committee, whenever it may require, an account of all his transactions as Secretary-Treasurer and of the annual financial condition of the Society.

Section 5. - Vacancies in Office: Any vacancy in the office of President-Elect or Secretary-Treasurer, however occasioned, may be filled, pending the election of his successor by the Society, by a majority vote of the remaining Committee. Should the office of President-Elect be filled by vote of the Executive Committee the person so elected as such by the Society according to the procedures set forth for election of officers of the Society in Article VIII, Section 1, of this Constitution relating to nominations and election. In such case, a President and a President-Elect shall be elected as specified in Article VIII, Section 1, of this constitution.

Article IX. Executive Committee

Section 1. - Members and Qualifications: All properties, property rights, objects, and purposes of the Society shall be managed, promoted, and regulated generally by an Executive Committee to consist of the immediate past President, the President, President-Elect and Secretary-Treasurer, of the Society. Any three officers shall constitute a quorum for the transaction of business. The Editor, as appointed in Article X, Section 2, shall be an ex officio member of the Executive Committee.

Section 2. - Annual Meetings: The Executive Committee shall meet immediately after the adjournment of the annual meeting of the members for the transaction of such business as may properly come before the Committee. No notice of such annual meeting shall be required and, should a majority of the newly-elected officers fail to be present, those present may adjourn without further notice to a specified future time.

Section 3. - Other Meetings: The Executive Committee shall not be required by this Constitution to hold regular meetings but may, by resolution, establish such order of meetings as it deems desirable. Special meetings of the Committee shall be held at any time at such places as may be specified in the notice or waiver thereof, whenever called by the President or any two or more officers.

Section 4. - Notice: Notice of all meetings of the Executive Committee other than the annual meeting, stating the time, place, and agenda for which the meeting has been called, shall be given to each officer by the President or officers calling the meeting not less than 3 days prior to the meeting.

Section 5. - Vacancies in Executive Committee: Any vacancy in the office of any officer, however occasioned, may be filled, pending the election of his successor by the Society, by a majority vote of the remaining Executive Committee.

Article X. Publications

Section 1. - Journal: The official publication of the Society shall be the Southwestern Entomologist and any other publication deemed appropriate by the Executive Committee.

Section 2. - Editor: The journal shall be edited and published by the Editor. The Editor shall be appointed by the Executive Committee for a 3-year term and may succeed himself. The Editor shall also serve as an ex officio member of the Executive Committee. The Editor may appoint an Associate Editor for a 1-year term.

Section 3. - Editorial Board: There shall be an Editorial Board to assist the Editor upon his request in (1) making recommendations to the Executive Committee concerning publication policies, and (2) the review of submitted papers and determining their suitability for publication in the journal. The Editor, Associate Editor, and Editorial Board may enlist the services of other persons where special needs exist. The Editorial Board shall consist of five Society members appointed for 5 years each, so that one member retires each year. Each year the Editor shall nominate a slate of two members, from which the Executive Committee shall appoint one.

Article XI. Miscellaneous Provisions

Section 1: All checks and drafts shall be signed in such manner as the Board of Directors may from time to time determine.

Section 2: At all duly constituted meetings of the Society of the Executive Committee of the Society, 25% of the membership in good standing or three officers, respectively, present shall constitute a quorum for the transaction of any business presented at such meetings.

Section 3: All notices required to be given by this Constitution relative to any regular or special meeting of the Society of the Executive Committee may be waived by the Committee or members entitled to such notice, either before or on the date of the meeting and shall be deemed equivalent thereto. Attendance at any meeting of the Society of the Executive Committee shall be deemed a waiver of notice thereof.

Section 4: If for any reason the Society shall disband, any and all assets shall be transferable to the Southwestern Branch of the Entomological Society of America.

Article XII. Amendments

Section 1: This Constitution may be altered or amended or By-Laws adopted by a majority vote of the quorum present at any annual or special meeting thereof, provided that notice of such proposed amendment or By-Laws shall have been set forth in the notice of the meeting.

VISUAL RESPONSES OF SOME SUGARBEET INSECTS TO STICKY TRAPS OF
VARIOUS YELLOW AND ORANGE HUES POSITIONED AT TWO HEIGHTS

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ABSTRACT

The visual responses of several sugarbeet insects to cylindrical sticky traps painted various yellow and orange hues and positioned at 2 heights were evaluated. Palestriped flea beetle, Systema blanda Melsheimer, Lygus spp., convergent lady beetle, Hippodamia convergens Guérin-Méneville, and different leafhopper spp. were caught most frequently on fluorescent yellow, yellow, or light orange traps although in some cases catches did not differ significantly from similar hues. The common green lacewing, Chrysopa carnea Stephens, did not exhibit a significant color preference. Significantly more adults of alfalfa caterpillar, Colias eurytheme Boisduval, a legume pest, were captured by fluorescent orange and fluorescent yellow sticky traps. Significantly more southern cabbageworm adults, Pieris proctodice Boisduval and LeConte, a crucifer pest, were captured by fluorescent orange sticky traps. Traps positioned at 0.6 m above the soil collected significantly more insects than those at 1.2 m except Lygus and C. carnea, which were captured most frequently by the traps positioned 1.2 m above the soil.

INTRODUCTION

Effective management of sugarbeet insects requires development of population monitoring techniques. The visual responses of some diurnal sugarbeet insects to colored sticky and water pan traps were studied by Capinera and Walmsley (1978). The palestriped flea beetle, Systema blanda Melsheimer, was found to be most attracted to yellow, while the aster leafhopper, Macrostelus fascifrons (Stål), and the leafhoppers Aceratagallia uhleri (Van Duzee) and Baicalutha neglecta (Delong and Davidson) were attracted to orange and yellow. Harper and Story (1962) reported that the sugarbeet root maggot, Tetanops myopaeformis (Röder), was attracted to yellow. Similarly, Scott (1976) reported attraction of beet leafhopper, Circulifer tenellus (Baker), to yellow-green, and Moericke (1969) indicated that the bean aphid, Aphis fabae Scopoli, was attracted to yellow.

Yellow or a similar hue may be a useful color for traps designed to exploit the visual host selection responses of sugarbeet insect pests. However, trap design and position, as well as color, can influence the effectiveness of an insect monitoring system. Thus, an additional study was undertaken to evaluate sugarbeet insect responses to sticky traps of various yellow and orange hues positioned at 2 heights.

MATERIALS AND METHODS

Cylindrical sticky traps were positioned on vertical poles 0.6 and 1.2 m above the soil. Trap design is described by Capinera and Walmsey (1978). Each pole supported 2 traps painted 1 of 6 hues. Both traps on a single pole were painted with the same hue. The hue names and their respective sources were yellow (Sherwin WilliamsTM Lemon Yellow F65 Y44), light orange (30 parts yellow and 1 part Sherwin WilliamsTM Tartar Red Dark F65 B1), orange (6 parts yellow and 1 part red), dark orange (2 parts yellow and 1 part red), fluorescent orange (Ace Hardware Glo-Spray, Oak Brook, IL., Sun Glow Orange 301), and fluorescent yellow (Illinois Bronze, Lake Zurich, IL., Solar Yellow 842). The fluorescent colors were applied over a base coat of white; a base coat was not applied in conjunction with the non-fluorescent colors.

Three replicates of each hue sticky trap were randomly positioned at 10 m intervals along the southern margin of a 2.5-ha sugarbeet field located at Fort Collins, CO. A large irrigation ditch separated the sticky traps from an 8.0-ha alfalfa field. Trap catches were tabulated at 4- to 7-day intervals from 12 June to 18 August, 1978, providing 10 collections, except for palestriped flea beetle, where 5 collections were obtained. No attempt was made to differentiate between leafhopper spp. because of the similarity of response exhibited in the earlier study. Data for each species or species group were converted to percent of total caught for each sampling interval. Because of the small percentile values and zeros, percent data were transformed to $\sqrt{x+0.5}$ for analysis of variance (Steel and Torrie 1960). Tests with significant F values were subjected to Tukey's HSD analysis at the 5% level. Tabular data presented herein were determined by averaging percent trapped on each sampling interval over the several intervals.

RESULTS AND DISCUSSION

The insects of economic importance collected from the sticky traps were palestriped flea beetle, Systema blanda Melsheimer; leafhopper spp.; Lygus spp.; the common green lacewing, Chrysopa carnea Stephens; convergent lady beetle, Hippodamia convergens Guérin-Méneville; alfalfa caterpillar, Colias eurytheme Boisduval; and southern cabbageworm, Pieris proctodice Boisduval and LeConte.

The sticky traps at the lower position caught significantly more of each insect species (Table 1) except for Lygus spp. and C. carnea, where significantly more insects were captured on the upper traps. For some species, especially alfalfa caterpillar, insects were almost exclusively captured by the lower traps. Thus, the position of the trap has a significant influence on trap catch, and trap height should be standardized if comparisons of catches are desired, although neither of the heights tested may be optimal. Other factors, such as distance between traps, height of crop, background color, and prevailing wind also may influence trap catch, but these factors were not evaluated in this study.

The color responses of the insects captured also are shown in Table 1. Palestriped flea beetles were captured more frequently on fluorescent yellow traps, but flea beetle response did not differ statistically among traps of similar colors except for a significantly lower catch on dark orange traps. Capinera and Walmsey (1978) also reported a strong orientation to yellow. Leafhoppers were caught most frequently on light orange, but leafhopper response did not differ statistically among the colors tested, except for

TABLE 1. Insects Recovered from Orange and Yellow Sticky Traps Positioned at 2 Heights (0.6 or 1.2 m Above the Soil).

Insect	Trap position ^{a/}	Total no. caught	Mean % distribution among trap hues ^{b/} (\pm SD)					
			Fluorescent yellow	Yellow	Light orange	Orange	Dark orange	Fluorescent orange
<u>Systema blanda</u>	upper *** lower ***	1205	8.8 \pm 3.4 ^a 17.1 \pm 6.0	6.0 \pm 2.1 ^a 17.7 \pm 3.6	5.6 \pm 3.6 ^a 13.0 \pm 3.8	4.8 \pm 3.1 ^a 9.0 \pm 3.2	1.6 \pm 0.9 ^b 3.2 \pm 2.7	3.6 \pm 1.1 ^{ab} 9.4 \pm 1.6
<u>Leafhoppers</u>	upper *** lower ***	18851	4.1 \pm 0.9 ^a 15.1 \pm 2.1	5.1 \pm 1.2 ^a 14.5 \pm 1.9	5.0 \pm 1.6 ^a 14.9 \pm 0.9	3.5 \pm 0.8 ^{ab} 11.9 \pm 1.6	3.0 \pm 0.9 ^b 7.1 \pm 1.7	4.0 \pm 0.9 ^{ab} 12.1 \pm 1.4
<u>Lygus</u>	upper * lower	1083	10.8 \pm 6.4 ^a 8.0 \pm 8.2	9.8 \pm 3.4 ^{ab} 8.4 \pm 5.9	12.2 \pm 6.4 ^a 9.8 \pm 5.4	9.0 \pm 5.0 ^{ab} 7.1 \pm 2.5	7.2 \pm 3.1 ^b 4.6 \pm 2.3	8.0 \pm 3.4 ^b 5.6 \pm 3.1
<u>Colias eurytheme</u>	upper *** lower ***	519	2.4 \pm 2.5 ^a 23.8 \pm 16.8	1.5 \pm 2.2 ^b 4.7 \pm 3.9	0.7 \pm 1.3 ^b 3.5 \pm 3.3	0.9 \pm 1.5 ^b 5.2 \pm 4.1	1.1 \pm 2.1 ^b 2.7 \pm 3.9	5.8 \pm 4.5 ^a 47.8 \pm 28.1
<u>Pieris proctodice</u>	upper *** lower ***	79	0 1.3 \pm 4.2	0 1.9 \pm 4.2	3.1 \pm 6.8 ^b 0.6 \pm 1.9	0 2.2 \pm 7.2	0 3.7 \pm 7.6	20.3 \pm 19.3 ^a 66.9 \pm 29.6
<u>Hippodamia convergens</u>	upper * lower ***	1231	8.7 \pm 4.2 ^{ab} 13.1 \pm 4.6	8.9 \pm 2.8 ^a 15.9 \pm 4.0	8.5 \pm 5.1 ^{abc} 12.7 \pm 3.0	4.2 \pm 2.8 ^{cd} 7.3 \pm 2.0	3.5 \pm 1.7 ^b 3.3 \pm 2.0	5.6 \pm 1.4 ^{bcd} 8.2 \pm 3.2
<u>Chrysopa carnea</u>	upper *** lower	474	4.2 \pm 5.0 ^a 6.4 \pm 6.1	16.6 \pm 12.9 ^a 4.9 \pm 4.5	8.1 \pm 6.5 ^a 6.7 \pm 6.2	7.5 \pm 7.5 ^a 6.0 \pm 7.5	13.2 \pm 8.7 ^a 9.7 \pm 5.7	9.4 \pm 5.5 ^a 7.5 \pm 6.6

^{a/}Position followed by asterisk indicates significantly higher catch (***) indicates $P < 0.001$; * indicates $P < 0.05$.^{b/}Numbers within row followed by the same letter indicate that insect catches associated with particular trap hues (upper and lower positions combined) are not significantly different at the 5% level according to Tukey's HSD.

dark orange, which was least preferred. Similarly, Lygus spp. catches did not differ statistically among fluorescent yellow, yellow, light orange, and orange. Alfalfa caterpillar was most attracted to the fluorescent colors. Southern cabbageworm exhibited an overwhelming preference for fluorescent orange while fluorescent yellow was one of the least attractive hues. Hippodamia convergens exhibited an attraction to yellow hues and light orange, an orientation not found in the earlier study. Chrysopa carnea did not exhibit color preferences in this study.

Yellow seems to be attractive to a number of sugarbeet insect pests. It appears possible to monitor sugarbeet insect abundance in sugarbeet fields by establishing yellow sticky traps on field margins. Similarly, alfalfa caterpillar abundance could be monitored with fluorescent orange or possibly yellow traps, while southern cabbageworm numbers could be monitored with fluorescent orange traps. Alfalfa caterpillar and southern cabbageworm are not pests of sugarbeet, but attack legumes and crucifers, respectively. Except in the case of Lygus spp., traps situated near the soil will catch more of these pest insects.

Sticky traps, to be useful for population monitoring, must not only attract insects but also reflect the relative abundance of captured species. The sticky traps used in this study demonstrated oscillations in insect numbers. For example, palestriped flea beetle catches declined dramatically in August after a period of abundance in July, while catches of Lygus spp., alfalfa caterpillar, and southern cabbageworm catches peaked in early June and early July. These changes in abundance paralleled shifts in numbers as determined by sweep net and direct observation (Capinera unpublished). However, additional work is required to establish a reliable correlation between trap catches and scouting reports. If this correlation can be established sticky traps should provide a convenient technique for monitoring sugarbeet insect densities.

ACKNOWLEDGMENT

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HORN FLY^{1/} PRODUCTION AS AFFECTED BY SEASONAL CHANGES IN RANGELAND FORAGE CONDITIONS

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ABSTRACT

Cattle manure collected throughout the year and bioassayed with laboratory-reared horn flies, *Haematobia irritans* (L.), showed production peaks similar to those present in the field at the time of sample collections. Numbers of horn flies produced, pupal weights and adult emergence rates were highest during the period of peak forage growth. Correlation (0.688 and 0.703) was highly significant for pupal production and pupal weights when compared to changes in the nitrogen content of the breeding medium.

INTRODUCTION

Investigations in Texas have shown that horn fly, *Haematobia irritans* (L.), populations decline to low levels in midsummer following high populations in spring and preceding higher populations again in late summer-early fall (Kunz and Cunningham 1977). Although the authors showed correlations between environmental factors and horn fly populations, these factors alone could not account for the midsummer declines. To elucidate this question, a study was conducted to determine if seasonal changes in the breeding medium, cattle manure, influenced horn fly production.

METHODS AND MATERIALS

This study was conducted in Burleson County, TX on a cooperator-owned ranch. Hereford cattle of different ages were maintained on a mixed Coastal Bermudagrass native grass range, and manure collections (5 pats/collection) were made weekly from mid-March through October 1977. Samples of manure were labeled, frozen, and stored for use in bioassays.

For bioassay, each sample (160 total) was subdivided 3X into 50-g subsamples and placed in 180-cc paper cups. Fly production was determined by infesting each sample of manure with 50 eggs obtained from a laboratory colony. One replicate of each sample was infested on each of 3 days with eggs obtained from the same flies for each of the 3 days to account for possible egg hatch differences in day-to-day fly populations.

The cups containing the manure sample and horn fly eggs were placed in an incubator at 29°C for 6 days to allow larval and pupal development. After pupation, each sample was scored for pupal yield, pupal weight, adult yield, and sex ratio.

A composite sample containing a subsample from each of the five weekly samples was assayed with the standard Kjeldahl method to determine the percentage of total nitrogen (dry weight basis) in the sample. The moisture content was also determined for each weekly sample.

^{1/} Diptera: Muscidae.

RESULTS AND DISCUSSION

Production of pupae (Fig. 1) and emergence of adults (Fig. 2) from pupae

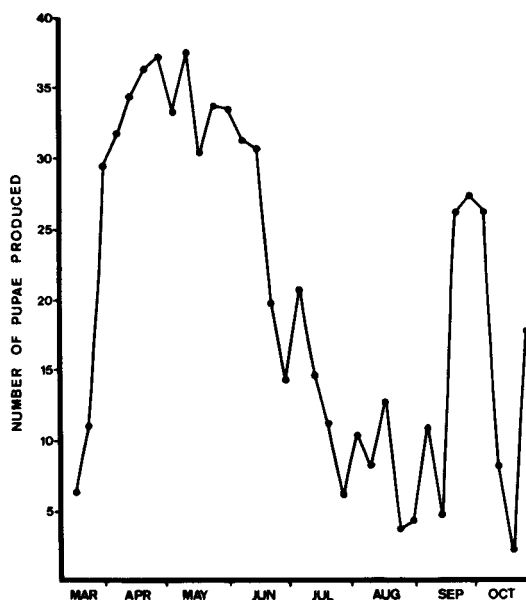
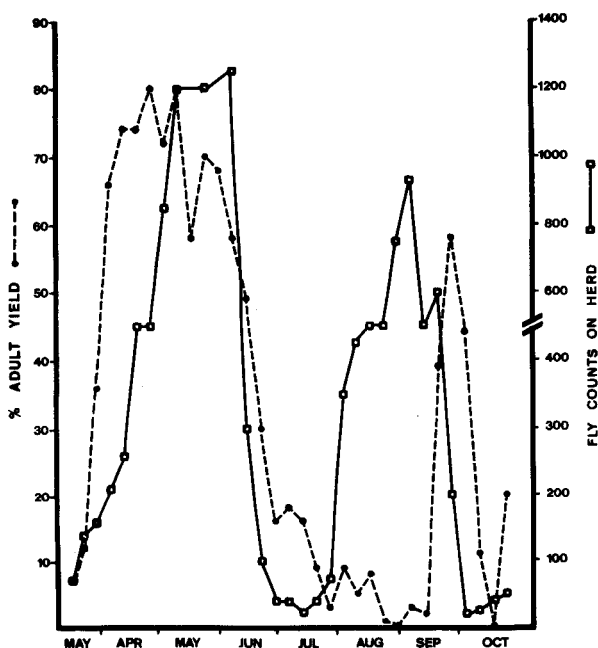


FIG. 1. Horn fly pupae produced from manure collected at different times of the year in Burleson County, TX 1977.

FIG. 2. Adult yield from bio-assay samples and respective horn fly populations on herd during sampling period in Burleson County, TX 1977.



produced from different manure samples was highest during the 8-wk April-May period and lowest during the July-August period. Another peak in production was observed during the September-October period. Although the laboratory studies showed highs and lows similar to those observed among fly populations on animals in the field, the peaks were not entirely synchronous (Fig. 2). However, the adults produced in the laboratory were not subjected to the adverse environmental factors that probably influenced mortality of the field populations that emerged during the spring. Asynchrony of late summer peaks is unexplained because predation and parasitism, which might be expected to delay adult buildup in the field, were not factors in the laboratory production.

The mean weights of pupae also showed seasonal fluctuations that appeared to be correlated with the highs and lows of production peaks (Fig. 3); heaviest pupae were produced during periods of the greatest yield of adults. The sex ratios of adults produced in this study were normal. Seasonal moisture content of the manure was fairly consistent ($\pm 3\%$) and averaged ca. 80% (Fig. 4).

In central Texas, pasture forage generally has two growing seasons, spring and late summer-early fall, that correspond with seasonal rainfall. Although the phenology of the pasture forage was not described in this study, the greatest number of horn flies were produced from manure collected during periods of greatest forage growth. The total percentage of nitrogen for the weekly collected manure samples also showed seasonal changes (Fig. 4). When pupal production and pupal weights for the seasonlong collections were compared with changes in percentage of nitrogen in the breeding medium, correlation coefficients of 0.688 and 0.703 were calculated for the respective measures; both values were highly significant at $P = .01$. Because a direct correlation has been established between heavier weights of female flies (as pupae) and increased egg deposition (Schmidt and Blume 1973), and because the current data show that the "nutrient" level of spring and fall-produced manure supports larger horn fly populations, it follows that larger populations are present on cattle during these high production periods. Greenham (1972) has also shown that egg-to-adult survival and the ultimate size of the Australian bushfly, *Musca vetustissima* Walker, strongly

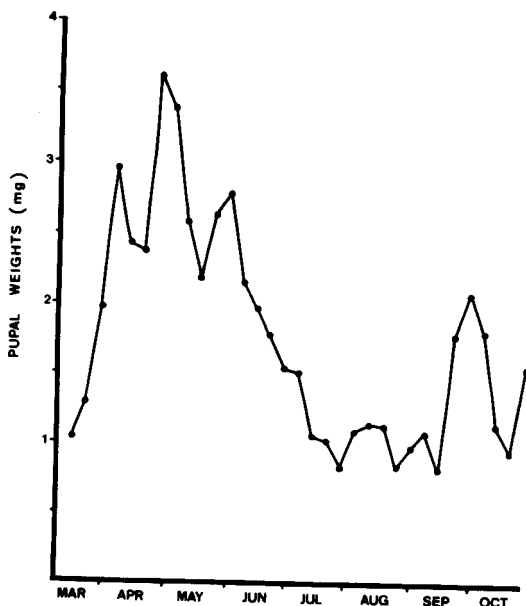


FIG. 3. Average weights of horn fly pupae that developed from seasonal collections of manure.

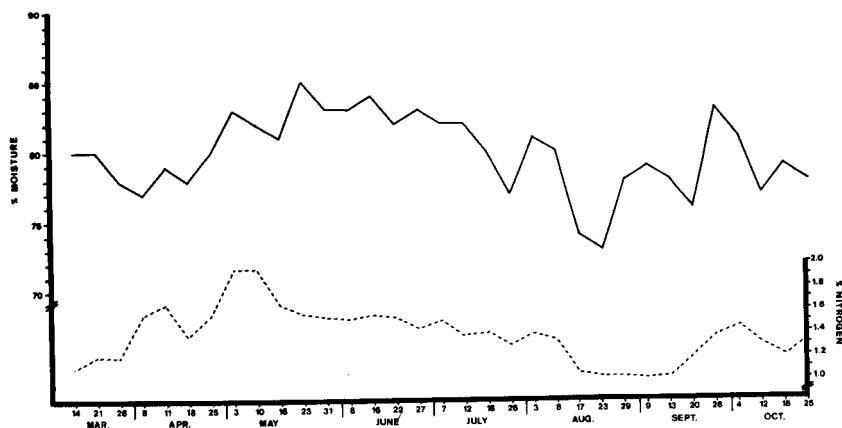


FIG. 4. Percentage moisture and nitrogen in manure samples.

depend on the texture, water content, and nitrogen content of the cattle manure. Morgan and Graham (1966) showed that different forage diets influenced the production of horn flies, but gave no indication what constituents or properties were responsible for these differences.

Thus our data indicate that seasonal changes in forage also cause changes in cattle manure that ultimately influence the production of horn flies. Additional study is needed to determine the influence of different well-defined rangeland vegetation types on horn fly production throughout the areas where they are found. If the nitrogen content of cattle manure is in itself not responsible for the increased production, then the condition of the manure as a result of seasonal changes in forage quality may have an effect on certain microflora or fauna that influence the production of horn flies. That the productivity of manure is apparently influenced by the season and is correlated with production peaks in the field, independent of environmental factors other than those exerted on the manure at time of collection, suggests that horn fly production may depend more on the "nutritional" aspect of the manure rather than on the direct effect of external weather factors. Further study to obtain a better understanding of the composition of cattle manure and its relationship to fly development is fundamental to the study of horn fly dynamics and could also provide important leads in the development of a defined artificial laboratory rearing medium.

ACKNOWLEDGMENT

I wish to thank VTERL personnel at College Station, TX, including Mr. B. Hogan, who were responsible for the samples collected.

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BOLL WEEVIL STERILITY: EFFECTS OF DIFFERENT COMBINATIONS
OF DIFLUBENZURON, ANTIBIOTICS, FUMIGATION, AND IRRADIATION^{1/2/3/}

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ABSTRACT

Studies were made of sterility, mortality, microbial contamination, and mating behavior of Anthonomus grandis Boheman after different kinds of treatments. Diflubenzuron and antibiotics were administered in the adult diet, and the weevils were further treated by irradiation (10 Krad in a nitrogen atmosphere) or by fumigation with bisazir (P,P, bis (1-aziridinyl)-N-methylphosphinothioic amide). When adults were fed diflubenzuron for 5 days after emergence, followed by irradiation or fumigation on the 6th day, a high level of sterility (99%) was induced. Diflubenzuron treatment apparently reduced sperm transfer and increased mortality, and antibiotics reduced pheromone production.

INTRODUCTION

Insects that are to be used for the sterile-insect technique of suppressing or eradicating a species should be 100% sterile. Such sterility can be manifested as failure to oviposit, failure of eggs to hatch, or lack of development in those eggs that do hatch from treated females or from untreated females mated with sterile males. Sterile boll weevils, Anthonomus grandis Boheman, should have a level of competitiveness that is expressed by the production of sufficient male pheromone to attract females and the capability to mate and transfer sperm to native female weevils. Previous studies have demonstrated that these attributes are difficult to attain in boll weevils because of problems with reduced

1/ Coleoptera: Curculionidae.

2/ This paper reports results of research only. Mention of a proprietary product does not constitute a recommendation or an endorsement by the USDA.

3/ In cooperation with the Mississippi Agricultural and Forestry Experiment Station, Mississippi State University, MS. 39762.

viability induced by the sterilizing treatment and microbial contamination that develop during mass rearing (Bartlett et al. 1968, Gueldner et al. 1977). Because of such problems, adult boll weevils released in the eradication test conducted in Mississippi in 1971 were fed a chemosterilant, busulfan, and the treated adults then had to be sexed by hand for removal of treated females which produced some fertile eggs (Haynes et al. 1973).

Recently attempts were made to sterilize boll weevils by treating pupae with fractionated doses of irradiation over a period of 4 1/4 days with 25 exposures of 270 rads each, or by fumigating adults with bisazir (P,P-bis(1-aziridiny)-N-methylphosphinothioic amide) and then immersing them in an acetone solution of 1.35% penfluron (2,6-difluoro-N-[[[4-(trifluoromethyl)phenyl]amino]-carbonyl]benzamide) (Haynes et al. 1978). However, complete sterility was not attained in these 2 procedures because the females were found to be partially fertile. Thus, the objective of this study was to develop a set of combined treatments that would sterilize both males and females, and also reduce the debilitating effects of microbial contamination.

METHODS AND MATERIALS

Boll weevils (ebony strain) used in the test were reared in the Robert T. Gast Rearing Facility at Mississippi State, MS. Newly emerged adults (groups of 5000) were placed in 30x40-cm nylon marquisette bags laid on 32x42x1-cm slabs of diet. Insects were held for (1) 3 days on treated diet and then an additional 2 days on untreated diet; (2) 5 days on treated diet; and (3) 5 days on untreated diet. The treated diet contained 500 ppm diflubenzuron added as a 25% WP and other ingredients as described by Lindig and Malone (1973) and Lindig et al. (1979). Slabs of diet were covered with ca. 8 tablespoons of grits (Grit-O-Cobs 0.60^R) that had been mixed dry with antibiotics for 1 h or contained antibiotics added during diet preparation. The slabs were used for 2 days; weevils fed on one side for a day and then the slab was turned over and covered with the antibiotic mixture for feeding on day 2. Antibiotics mixed with grits for treatments included streptomycin sulfate (0.01%), neomycin sulfate (0.01%), and folpet (0.03%); those mixed into the diet were streptomycin sulfate (0.03%), neomycin sulfate (0.03%), chloramphenicol (0.01%), and kanamycin (0.01%). Diets without diflubenzuron also contained or were coated with the antibiotics.

After 5 days on the different diets, the adults were treated further on the 6th day: (1) with 10 Krad gamma irradiation from a ¹³⁷Cs source in a nitrogen atmosphere; (2) with 10 Krad gamma radiation from a ¹³⁷Cs source in a nitrogen atmosphere plus dipping in a solution of 0.01% diflubenzuron in acetone for 5 sec; (3) by fumigation for 90 min with bisazir; and (4) by fumigation for 90 min with bisazir plus dipping in a solution of 1.35% penfluron in acetone for 5 sec. Untreated controls were included with each treatment.

Mating success was determined by setting up 25 pairs of the same age on day 3 and 7 after exposure to the treatment (treated male x untreated female; untreated male x treated female; and untreated male x untreated female). Copulatory behavior was observed during the 1st h of the 4-h ad lib. mating (0900-1300) h. Then, females were dissected for determination of sperm transfer. The effects of diflubenzuron, antibiotics, and irradiation and fumigation on sterility from the paired matings were evaluated on the basis of percentage of egg hatch and percentage of eggs that developed to adults. The experiment was replicated 6 times and mortality was recorded daily. The extent of bacterial contamination in weevils was determined at day 0 (when adults were placed on the diets), day 6 (when weevils were removed from the diets and

treated), and at days 3 and 7 after treatment by irradiation or fumigation (Sikorowski 1975). Pheromone was analyzed in frass collected daily for 10 days from males held separately and fed fresh field collected squares (5 adults/square) (McKibben et al. 1976).

RESULTS AND DISCUSSION

Adult males that ingested diflubenzuron (500 ppm) transferred less sperm to virgin females than males that were treated similarly but not fed diflubenzuron in all treatments except fumigation only (Table 1). The coadministration of diflubenzuron also appeared to reduce the frequency of observed matings among weevils treated by irradiation. For example, after 5, 3, and 0 days of feeding on diflubenzuron, the average percentages of sperm transfer were 31.2, 31.6, and 49.3; and percentages of observed matings were 40.9, 50.2, and 51.4, respectively. Irradiation, fumigation, and combinations of these treatments with the IGRs, significantly reduced matings and sperm transfer ($P > 0.05$) when compared to the untreated controls (Table 1). Inclusion of the antibiotics, either as a surface treatment of the diet, or as a combined application on the surface and within the diet, did not significantly affect matings or sperm transfer. In these tests, the percentages of observed matings and sperm transfer by 6-day-old adults were higher than was previously reported in similar sterilization treatments of newly emerged adults (Wright et al. 1979).

TABLE 1. Effects of Different Treatments on Mating and Sperm Transfer by Boll Weevils.^{a/}_{b/}

Sterilization procedure at day 6	Antibiotics location ^{c/}	% Observed matings			% With sperm		
		0/5	3/5	5/5	0/5	3/5	5/5
Fumigation only	A ♂	63 a	39 a	52 a	53 a	39 b	45 ab
	B ♀	35 b	50 ab	50 a	47 ab	43 b	57 a
Fumigation plus 1.35% penfluron dip	A ♂	61 a	60 a	53 a	45 a	19 c	25 c
	B ♀	30 c	43 a	46 a	36 ab	24 c	28 c
Irradiation only	A ♂	64 a	61 a	41 b	47 ab	32 c	31 c
	B ♀	54 a	39 ab	28 c	75 a	21 c	31 c
Irradiation plus 0.1% diflubenzuron dip	A ♂	42 b	50 ab	33 c	38 bc	33 c	15 c
	B ♀	54 b	44 b	29 c	56 a	31 c	20 c
Control	A ♂	77 a			88 a		
	B ♂	67 a			77 a		

a/ Adults were held for 5 days with no diflubenzuron in diet (0/5); with diflubenzuron in the diet for the first 3 days and then none for 2 days (3/5); and with diflubenzuron in the diet for 5 days (5/5) after adult emergence.

b/ Means followed by the same letter not significantly different at the 0.05% level according to Duncan's multiple range test.

c/ A - Antibiotics placed on surface of diet only for 5 days. B - Antibiotics placed on surface and within diet for 5 days.

Pheromone production averaged ca. 100 µg/weevil/day when weevils were treated by irradiation or fumigation on day 6 after feeding for 5 days on diets that received only surface treatments with the antibiotics. However, the pheromone level was less than 20 µg/weevil/day with similar weevils and treatments when the antibiotics were applied to both the surface and within the diet.

Mortality was significantly lower among adults fed 5 days on diets surface-treated with antibiotics than among those fed diets treated on the surface and within the diet ($P > 0.05$) (Table 2). Irradiation or fumigation of the 6-day-old weevils that were not fed diflubenzuron caused less mortality than similar treatments of adults that fed for 3 or 5 days on diets containing diflubenzuron. Thus, the diflubenzuron at 500 ppm in the adult diet appeared to have an additive effect on mortality when combined with irradiation or fumigation for sterilization. Dipping of the adults in 0.1% diflubenzuron or 1.35% penfluron after irradiation or fumigation had little additional effect on adult mortality.

TABLE 2. Effects of Different Treatments on Boll Weevil Mortality.^{a/}^{b/}

Sterilization procedure at day 6	Antibiotics location ^{c/}	% Mortality at indicated days posttreatment					
		0/5		3/5		5/5	
		3	7	3	7	3	7
Fumigation only	A ♂	5 a	5 a	15 a	25 ab	7 a	37 ab
	♀	10 a	10 a	5 a	40 ab	0 a	40 ab
	B ♂	30 b	95 c	50 b	90 c	27 ab	63 c
	♀	35 b	95 c	15 a	95 c	13 a	77 c
Fumigation plus 1.35% penfluron dip	A ♂	10 a	20 a	15 a	45 ab	10 a	46 ab
	♀	0 a	15 a	15 a	35 ab	0 a	17 a
	B ♂	35 b	70 c	15 a	80 c	13 a	77 c
	♀	40 b	75 c	15 a	85 c	10 a	80 c
Irradiation only	A ♂	0 a	10 a	0 a	45 b	10 a	70 c
	♀	0 a	15 a	5 a	70 c	10 a	70 c
	B ♂	25 ab	70 c	5 a	80 c	10 a	80 c
	♀	25 ab	85 c	35 b	90 c	15 a	93 c
Irradiation plus 0.1% diflubenzuron dip	A ♂	0 a	15 a	10 a	25 ab	6 a	40 b
	♀	0 a	50 b	0 a	75 c	6 a	57 bc
	B ♂	0 a	10a	10 a	95 c	6 a	90 c
	♀	30 ab	60 bc	0 a	85 c	2 a	93 c
Control	A ♂	3 a	17 a				
	♀	10 a	15 a				
	B ♂	23 ab	34 ab				

^{a/} Adults were held for 5 days with no diflubenzuron in diet (0/5); with diflubenzuron in the diet for the first 3 days and then none for 2 days (3/5); and with diflubenzuron in the diet for 5 days (5/5) after adult emergence.

^{b/} Means followed by the same letter not significantly different at the 0.05% level according to Duncan's multiple range test.

^{c/} A - Antibiotics placed on surface of diet only for 5 days. B - Antibiotics placed on surface and within diet for 5 days.

The beneficial effects of diflubenzuron in the diet of adult weevils are well illustrated by the data on sterility shown in Table 3. Although irradiation or fumigation alone gave a high level of sterility in adults, some hatch in eggs and larval development occurred in the treated x untreated matings. The incorporation of diflubenzuron in the adult diet prevented hatch and development of adults from eggs deposited after exposure of adults to 10 Krad of gamma irradiation or to 90 min fumigation with bisazir.

TABLE 3. Effects of Different Treatments on Fertility of Boll Weevils.^{a/}

Sterilization procedure at day 6	Antibiotics location ^{b/}	0/5				3/5				5/5			
		%		%		%		%		%		%	
		hatch		develop-		hatch		develop-		hatch		develop-	
		3	7	3	7	3	7	3	7	3	7	3	7
Fumigation	A ♂	7	0	0	2	5	0	0	3	0	0	0	0
	♀	4	0	0	0	0	0	0	0	0	0	0	0
	B ♂	10	0	5	0	25	0	0	0	0	29	0	10
	♀	0	0	0	0	10	0	0	0	0	0	0	0
Fumigation plus 1.35% penfluron dip	A ♂	16	0	0	0	23	0	3	0	7	0	0	0
	♀	10	0	0	0	0	0	0	0	0	0	0	0
	B ♂	0	0	0	0	0	0	0	0	0	0	0	0
	♀	0	0	0	0	0	0	0	0	0	0	0	0
Irradiation only	A ♂	23	0	0	0	15	20	16	20	0	10	0	0
	♀	11	45	0	0	27	0	0	0	0	0	0	0
	B ♂	0	0	7	0	3	0	0	0	0	0	0	0
	♀	0	0	10	0	0	0	0	0	0	0	0	0
Irradiation plus 0.1% diflubenzuron dip	A ♂	8	0	0	0	23	4	0	0	9	0	0	0
	♀	32	0	0	0	29	0	0	0	0	0	0	0
	B ♂	90	22	28	0	0	4	0	0	0	0	0	0
	♀	0	0	0	0	5	0	0	0	0	0	0	0
Control	A ♂	99	97	47	47								
	♀	95	98	43	36								
	B ♂	82	78	17	26								

^{a/} Adults were held for 5 days with no diflubenzuron in diet (0/5); with diflubenzuron in the diet for the first 3 days and then none for 2 days (3/5); and with diflubenzuron in the diet for 5 days (5/5) after adult emergence.

^{b/} A - Antibiotics placed on surface of the diet only for 5 days. B - Antibiotics placed on surface and within diet for 5 days.

Observations of bacterial contamination revealed that our test weevils had only 0-500 colonies/weevil (Class I Sikorowski 1975) and that there were no differences between those exposed to antibiotics applied only to the surface of the diet or as combined applications to the surface of and within the diet. For mass rearing and sterilization techniques used in the Boll Weevil Eradication Trial in North Carolina in 1979, it was necessary to use antibiotics to minimize microbial contamination. However, different antibiotics must be found because those used in our tests apparently increased mortality and reduced pheromone production.

Feeding of the adults for 5 days on a standard diet apparently allowed sufficient time for the initiation of pheromone production. Our results agree with those of Guelndner and Wiygul (1978) who showed that untreated weevils began significant pheromone production at that age. There were no significant differences in observed matings and sperm transfer for weevils sterilized by 10 Krad of gamma irradiation or by 90 min fumigation with bisazir. Therefore, the logistics involved in handling and sterilizing millions of boll weevils appear to favor the use of gamma irradiation because of the simplicity of handling, the efficiency in monitoring doses, the possibility of using non-specialized facilities, and the greater safety to personnel performing these operations.

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DISTRIBUTION OF LOBOMETOPON OVALE (CASEY) IN TEXAS;
A "LOST" SPECIES REDISCOVERED ^{1/}

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ABSTRACT

Lobometopon ovale (Casey, 1884), until now known only from the type specimens labelled "Texas", is shown to be fairly abundant in southern Texas where it appears to be associated with wild buckwheat, Eriogonum multiflorum Benth.

Epitragus ovalis Casey (1884, p. 184) was described from 5 specimens labelled "Texas." In 1907, Casey (p. 385) described the genus Lobometopon to which he assigned his species ovalis (=ovale). Apparently Casey did not see additional specimens of this species since he merely repeated "Texas." His collection in the U. S. National Museum now contains only two specimens of the original series of 5 on which the original description was based.

The next mention of Lobometopon ovale in the literature (other than checklists and catalogs) is in Freude's 1968 paper (p. 59). Freude merely repeated Casey's 1884 description verbatim and stated that he knew the species only from Texas with no mention of specific localities.

Several years ago, among specimens received for identification from J. E. Gillaspay, J. E. Wappes and R. H. Turnbow, I discovered specimens which, upon comparison with Casey's types (U. S. National Museum #46326) proved to be L. ovale (habitus as in Fig. 1; illustration prepared by John Nagy). These collectors, at my suggestion, have managed to collect over 160 specimens which provide some interesting distributional and ecological data on this species.

Casey's description was prepared in his usual meticulously detailed style and I can add nothing at this time other than to point out that L. ovale is fully winged and that the size range, based on 160 specimens is: Length: 7.8 - 10.6mm; width: 3.4 - 5.3mm.

According to available records, L. ovale occupies a rather restricted range in southern Texas. Except for one specimen from Lee County, the records are from a cluster of 11 contiguous counties along the Gulf Coast (Aransas, Bee, Brooks, Duval, Hidalgo, Kenedy, Kleberg, Nueces, Refugio, San Patricio and Willacy). See map, Fig. 2.

Specimens have been collected from 10 July to 5 December, with by far the largest numbers taken in October. J. E. Wappes (in litt.) says that peak populations of L. ovale seem to follow the heavy late fall rains which normally start in south Texas from mid-September to mid-October. All 3 collectors (Gillaspay, Turnbow, Wappes) report that L. ovale appears to be closely associated with wild buckwheat (Eriogonum multiflorum Benth.). Wappes (in litt.) indicated that this beetle is quite commonly taken in company with the cerambycid, Tanyochraethes tildeni Chemsak and Linsley, which was described in 1965 from Welder Wildlife Refuge, San Patricio County, TX.

^{1/} Coleoptera: Tenebrionidae: Epitragini.

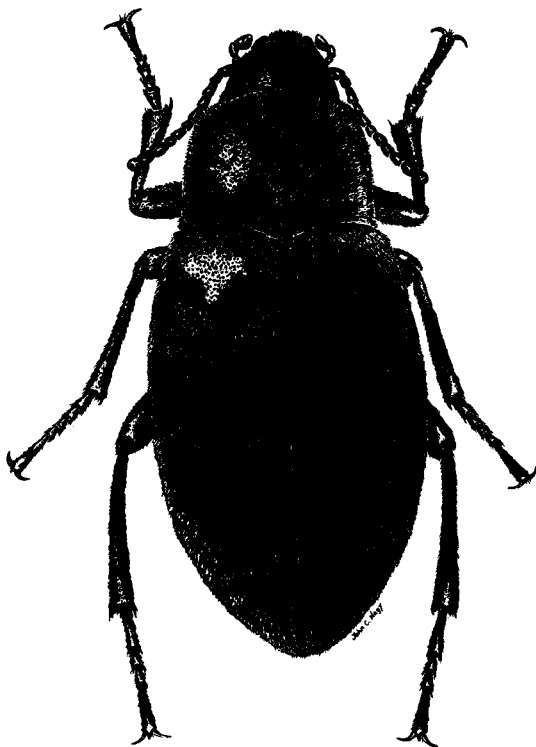


FIG. 1. Lobometopon ovale (Casey)
(actual length: 8.5mm).



FIG. 2. Distribution of Lobometopon ovale (Casey).

The old adage that few things are really rare if one knows where, when and how to look for them is well illustrated by this example. Until now, all the specimens of L. ovale I have seen, including the types (U. S. National Museum), were 4 in the Canadian National Collection and 2 in the Illinois Natural History Survey Collection.

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COTTON: SAMPLE SIZES REQUIRED TO ESTIMATE NUMBER OF PLANTS AND FRUITING FORMS BY THREE SYSTEMS OF SAMPLING^{1/}

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and J. A. Harding^{5/}

ABSTRACT

Three systems, the grid, the X, and the area, were used to sample whole cotton plants for squares, undamaged green bolls, insect-damaged and undamaged open bolls, and rotted bolls, from the seventh through the nineteenth week after the cotton was planted in a 0.4-ha plot near Brownsville, TX, in 1972. The number of plants sampled could not be related statistically to the total number of plants in the plot. The formula $n = (CV/30\%)^2$ was used to determine the number of plants needed to be sampled for each of the indices. In general, as the number of fruiting forms increased, fewer plants had to be sampled to satisfy the criterion.

When 0.4-0.7 squares/plant were present, 31, 47-120, and 67-189 plants had to be sampled by the grid, X, and area systems, respectively, but when 0.5 or more bolls were present during the mid-growing season, only 10-20 plants had to be sampled by any of the 3 systems. Thus, the type of fruiting form being sampled and plant maturity greatly influenced the sample size needed.

INTRODUCTION

Much of the economics of cotton production depends on the level of insect damage in the field. To forestall the buildup of infestations that could inflict damage of economic importance, growers should be aware not only of the density of pest insects, but of the density of the cotton fruit and the extent to which it has matured during the growing season, as was clearly presented by Gonzalez (1971) and Reynolds (1975) for pest management programs. Then based on the relationship between these conditions as determined by sampling, decisions on when to take measures to control the pest population can be made. For example, when bolls are 12 or more days old, there is no need to apply insecticides for the control of the boll weevil. However, larvae of the *Heliothis* spp. can destroy bolls of any age; therefore, sampling for fruit as well as for pests, can serve not only to prevent economic damage but to obviate needless control efforts.

Thus, having established the necessity to sample, the next step is to select a sampling system and, concomitantly, to determine the amount of sampling necessary to achieve the desired level of statistical probability.

Dupnik et al. (1973) while evaluating systems of sampling for fruit on cotton, found that frequently more sampling was done than was necessary to arrive at a reliable estimate. However, these authors did not determine

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the numbers of plants that had to be sampled before accuracy at some probability could be attained. We therefore compared 3 commonly-used sampling systems, the grid, X, and area, and determined the numbers of plants needed to be sampled for each system.

The X system has been used in the cotton pest management program in Texas since 1972. We acknowledge the fact, well documented by Wadley (1967), that in general the grid and X systems are more effective than the area system because they permit greater coverage of the total experimental area. We wished to evaluate these systems for their specific advantages as methods for sampling fruit of the cotton plant.

MATERIALS AND METHODS

Planting. The cotton cultivar 'Stoneville 7A' was planted in a 123-ha field in Cameron County during March 1972 in rows 1 m apart. In late April a 0.4-ha test plot, 62-rows wide X 64-m long, was selected in the middle of the field.

Sampling. For the grid system, 10-11 plants selected at random in 5-6 rows, spaced 10 rows apart, constituted the 1st replicate; the 2nd replicate consisted of plants in another 5-6 rows similarly spaced, etc., across the test plot to insure that all the rows except the 2 outside rows would be entered at least once during the growing season. This sampling system was replicated 2-3 times/sampling date except on May 2 and 18 when we sampled once, and on August 21-22 when we sampled 9 times.

The X system required the sampling of plants that were located on imaginary diagonal lines (X-pattern) connecting the opposite corners of the plot. About 30 plants, equidistant from one another, were sampled on each diagonal line, or 60 plants/replicate, 2-3 replicates/sampling date except on August 21-22 when we sampled 4 times.

The area system required the sampling of all plants in 4 m of row located near the center of each of the 4 quarters of the plot; 1-3 replicates/sampling date were made.

All sampling was done in the same plot. Data for squares, flowers, green bolls, damaged and undamaged open bolls, and rotted bolls were obtained from soil-to-terminal examinations of whole plants *in situ*. No breakdown of square or boll size was made. At harvest (August 23-25), each plant in each row was counted.

Analysis. The means and standard deviations of the samples and the coefficients of variation (CV) were calculated from the data recorded for each plant; then the numbers of plants to be sampled were calculated by $n = (CV/30\%)^2$ (Harcourt 1961, Mukerji and Harcourt 1970) for the 3 sampling systems.

The senior author deemed the use of the 30% value to be practical because it provided adequate information with a minimum amount of sampling effort. The theory and application for relative precision in estimating sample size is discussed by Hansen et al. (1953). Then, according to Cochran (1953), the grid and X systems utilized the random stratification, and the area system, the complete stratification method of sampling plants. In addition, we used a standard linear regression analysis to compare means per plant of squares, flowers, and green bolls recorded for the grid and X systems on May 18 and 26, June 22, July 3 and 28-29. The total number of plants counted at harvest was compared to the mean \pm s and range of numbers of plants for the replicates recorded by the area sampling system.

RESULTS AND DISCUSSION

Calculated Plant Populations Compared to Actual Counts. Based on the number of plants that were sampled by the area system, 1-3 replicates, encompassing 0.4-1.2% of the 0.4 ha (Table 1), we estimated within 1 standard

deviation of the actual count only once in 6 times. However, on 3 of the first 4 sampling dates, the actual total number of plants was within the range of the number determined by sampling.

TABLE 1. Comparison of the number of Cotton Plants Estimated by the Area System of Sampling with the Actual Number.^{a/}

Date	Week after plantings		No. repli- cates	% of 0.4 ha examined	X no. ^{b/} replicate \pm s ^{c/}	plants/ha per	Range for replicates (no./ha) ^{b/}	
							Low	High
May	2	7	2 _{d/}	9.8	115,432 \pm 12,148*		98,765	155,556
	18	9	1 _{d/}	0.4	121,605 \pm 16,970*		103,704	125,926
	26	10	3	1.2	94,025 \pm 13,430NS		74,074	111,111
June	22	14	3	1.2	72,222 \pm 13,857*		54,321	106,173
July	3	16	3	1.2	46,099 \pm 12,316*		37,037	56,790
	28-29	19	3	1.2	52,667 \pm 16,252*		29,630	74,074

a/ Planted 2nd week of March, 1972, projected from mean number plants (654.5 \pm 158.7 per row) in an experimental plot of 62 rows, 64 m long; range 255-992 plants/row.

b/ Projected from sampling records of 62-row plots.

c/ Outside 1 s (*) or inside 1 s (ns) of actual plant counts: 100,200.

d/ Statistics computed from 4 quadrants of a single replicate.

Estimates of Fruit Populations. Tables 2, 3, and 4 show results of sampling by each system for fruiting forms of cotton as mean number per plant \pm s per replicate per sampling date during the growing season. All sampling systems showed similar ranges of mean \pm s for squares per hectare. The number of plants to be sampled was related to the population of plants in the test area. When the square populations were less than about 50,000/ha, 31, 47-120, and 67-189 plants had to be sampled to achieve an accuracy of 30% of CV by the grid, X, and area systems, respectively, or about 50% and 25% fewer plants with the grid and X systems, respectively, than with the area system. Although differences in sampling techniques prohibited direct comparisons among all 3 of the systems, we could compare the grid and X systems because they were sampled in essentially the same way, i.e., by random stratification. When the means for the grid and X systems comprised the Y and X variables, respectively, of a linear regression analysis, the correlation coefficients (r) ranged from 0.96 to 0.99 for squares, flowers, and green bolls (significant at the 5% level of probability with 3 degrees of freedom). Later when populations of squares were about 100,000, 222,000, and 227,000/ha, no more than 20 plants had to be sampled by the grid, X, and area systems, respectively.

When there were no more than 0.06 white flowers per plant (or 1 in 16 plants), maximums of 556 and 578 plants had to be sampled by the grid and X systems, respectively. When the flower population was 0.1 or more per plant, 26-135, 18-65, and 22-135 plants had to be sampled by the grid, X, and area systems, respectively. The need to sample more plants for flowers than for squares and bolls was attributable to the short life (24 h) of the flower as compared to the other forms.

When green boll populations averaged 1-5/plant, 4-20 plants were required for sampling at 30% of CV regardless of the system used, but when green boll populations were 0.01-0.1/plant, the maximum numbers of plants to be sampled were 131, 283, and 867 for the grid, X, and area systems, respectively. Thus when the populations of green bolls per plant were 0.1 or less, the grid system was superior to the others.

Regardless of the sampling systems used, the CV's for all fruiting forms generally decreased as the season progressed (Table 5). The late season rise in CV's for squares was probably due to a reduction in squaring and to boll set. The variation among the CV's for the systems and indices was not great, although the grid and X systems showed more variation in CV's for flowers than did the area system, and the CV's for green bolls were lower for the X system than for the other two systems.

Sampling by the grid and X systems was done for rotted, undamaged and damaged open bolls in the 0.4 ha on August 21-22. When undamaged open bolls/plant numbered 0.2 or less, 112-667 and 419-667 plants had to be sampled by the grid and X systems, respectively. When 0.46-4.0 undamaged open bolls/plant were present, 4-36 plants had to be sampled by the grid and X systems. When there was less than 1 damaged open boll or rotted boll/plant, 16-156 plants had to be sampled by the grid and X systems, respectively.

Table 6 shows the number of plants that should be sampled with the grid or X systems when the indicated number of fruiting forms are present. The sampling time required for any of the recommended numbers should not exceed 2 h/ha.

Wadley (1967) showed that the sampling effort was reduced when smaller samples were taken at numerous spacially separated sites, rather than when sampling was concentrated in a limited area in a field. The grid and X systems require a wide spacial distribution of sampling.

In addition, these results could be used to designate minimum sample sizes for fruit populations by sequential sampling (Kuno 1972) and for the double sampling methods as described by Cochran (1953).

TABLE 2. Numbers of Fruiting Forms Estimated by Grid System of Sampling.

Sampling date	No. plants sampled (per replicate)	Range (per replicate)		No. plants to be sampled (n = (CV/30%) ²)	Estimated no./ha ^{a/} (X1000)
		Per plant			
		Mean	s		
<u>Squares</u>					
May 2	70	0.40	0.8	31	40.1
18	60	1.00	2.0	15	100.2
26	50- 60	4.00-4.0	2.0-3.0	4- 5	389.0-401.2
June 22	59- 60	5.00-3.0	3.0-5.0	3- 4	529.1-752.0
July 3	59- 60	6.00-9.0	4.0-8.0	5- 9	356.3-912.3
28-29	59- 60	2.00-3.0	3.0-4.0	11- 18	200.5-325.7
<u>Flowers</u>					
May 26	50- 50	0.00-0.06	0.1-0.2	0-556	2.0- 6.0
June 22	59- 60	0.30-0.4	0.6-0.6	26- 35	34.1- 40.0
July 3	59- 60	0.20-0.2	0.4-0.6	81-135	15.1- 17.0
28-29	59- 60	0.30-0.4	0.6-0.7	39- 56	27.2- 37.3
<u>Green Bolls</u>					
May 26	50- 50	0.08-0.1	0.3-0.4	93-131	3.0- 14.1
June 22	59- 60	1.00-2.0	1.0-2.0	9- 15	102.2-168.4
July 3	59- 60	2.00-3.0	2.0-2.0	8- 9	241.0-252.0
28-29	59- 60	3.00-3.0	2.0-3.0	5- 8	272.3-343.0
<u>Open Undamaged Bolls</u>					
July 28-29	50- 60	0.02-0.2	0.1-0.5	112-667	2.0- 15.1
Aug. 21-22	99-116	0.60-3.0	0.7-4.0	5- 25	55.1-326.0
<u>Open Damaged Bolls</u>					
Aug. 21-22	99-116	0.07-0.4	0.3-1.1	42-156	7.0- 42.0
<u>Rotted Bolls</u>					
Aug. 21-22	99-116	0.08-0.7	0.3-2.0	23-144	8.0- 73.1

a/ Mean times 100,200 plants/ha.

TABLE 3. Numbers of Fruiting Forms Estimated by X System of Sampling.

Sampling date	No. plants sampled (per replicate)	Range (per replicate)		No. plants to be sampled (n = (CV/30%) ²)	Estimated no./ha ^a (X1000)
		Per plant			
		Mean	s		
<u>Squares</u>					
May 2	62- 62	0.10-0.4	0.4-0.8	47-120	10.1- 40.0
18	31- 31	2.00-3.0	1.7-1.8	4- 5	248.4-310.6
26	52- 53	4.00-4.0	2.0-2.0	4- 4	377.5-394.3
June 22	61- 62	7.00-7.0	5.0-5.0	5- 7	504.4-693.0
July 3	61- 62	7.00-8.0	5.0-5.0	4- 6	653.0-760.0
28-29	60- 61	2.00-4.0	2.0-5.0	10- 14	222.5-409.0
<u>Flowers</u>					
May 26	52- 53	0.02-0.04	0.1-0.3	109-578	2.0- 9.1
June 22	61- 62	0.30-0.5	0.6-0.9	37- 39	30.0- 50.1
July 3	61- 62	0.20-0.2	0.4-0.5	57- 65	20.0- 20.0
28-29	60- 61	0.40-0.5	0.6-0.6	18- 25	39.0- 47.2
<u>Green Bolls</u>					
May 26	52- 53	0.04-0.09	0.2-0.3	109-283	4.0- 9.0
June 22	61- 62	1.00-2.0	2.0-2.0	14- 18	136.3-195.3
July 3	61- 62	2.00-2.0	2.0-2.0	8- 12	221.5-239.0
28-29	60- 61	3.00-4.0	2.0-2.0	4- 6	318.0-367.0
Aug. 21-2	347-410	0.20-0.7	0.4-1.2	8-144	8.0-167.4
<u>Open Undamaged Bolls</u>					
July 28-29	60- 61	0.03-0.08	0.3-0.5	419-667	3.0- 8.0
Aug. 21-22	347-410	0.46-3.5	0.7-2.8	4- 36	46.2-352.1
<u>Open Damaged Bolls</u>					
Aug. 21-22	347-410	0.07-0.62	0.3-1.2	42-156	7.0- 62.2
<u>Rotted Bolls</u>					
Aug. 21-22	347-410	0.07-0.92	0.3-1.4	16-156	7.0-286.4

^a/ Mean times 100,200 plants/ha.

TABLE 4. Numbers of Fruiting Forms Estimated by Area System of Sampling.

Sampling date	No. plants sampled (per replicate)	Range (per replicate)		No. plants to be sampled (n = (CV/30%) ²)	Estimated no./ha ^a (X1000)
		Per plant			
		Mean	s		
<u>Squares</u>					
May 2 _b /18 _b	40- 63	0.05-0.25	0.2-0.7	67-189	4.9- 27.2
	42- 51	0.70-1.4	0.9-1.8	8- 27	69.1-140.2
26	140-157	2.00-3.0	2.0-3.0	7- 8	227.4-322.7
June 22	104-136	5.00-6.0	4.0-5.0	7- 7	496.1-603.2
July 3	57- 85	7.00-8.0	5.0-6.0	6- 7	719.5-799.5
28-29	78-100	3.00-4.0	4.0-5.0	16- 20	288.6-410.9
<u>Flowers</u>					
May 26	140-157	0.10-0.2	0.3-0.4	67-135	7.7- 16.1
June 22	104-136	0.30-0.55	0.6-0.8	22- 36	29.9- 55.1
July 3	57- 85	0.24-0.37	0.5-0.6	31- 44	24.0- 37.0
28-29	78-100	0.31-0.5	0.6-0.8	22- 56	31.1- 50.1
<u>Green Bolls</u>					
May 26	140-157	0.01-0.1	0.1-0.4	90-867	1.23-13.1
June 22	104-136	1.00-2.0	1.4-1.7	11- 20	101.2-157.3
July 3	57- 85	2.00-3.0	1.8-2.5	8- 10	217.5-256.5
28-29	78-100	4.00-5.0	3.3-5.4	10- 13	359.0-507.0

^a/ Mean times 100,200 plants/ha.^b/ Statistics computed from 4 quadrants of single replicate.

TABLE 5. Time of Fruiting of the Cotton Plant and the CV's for Squares (SQ), Flowers (FLO), and Green Bolls Sampled by 3 Systems in a 0.4-ha Plot Near Brownsville, TX 1972.

Sampling date	Weeks after planting		CV for sampling system								
			Grid			X			Area		
			SQ	FLO	Bolls	SQ	FLO	Bolls	SQ	FLO	Bolls
May	2	7	166	-	-	261	-	-	327	-	-
	16	9	116	-	-	62	-	-	129	-	-
	26	10	63	606	323	59	434	380	82	298	365
June	22	14	60	164	98	76	186	119	76	166	111
July	3	16	86	316	89	68	233	94	76	179	92
	28-29	19	111	215	73	107	139	65	117	118	101

TABLE 6. Number of Cotton Plants to be Sampled by the Grid or X Systems.

Name	Fruiting Form	
	No./ha	No. of plants to be sampled ^{a/}
Square	20,000	25-35
	100,000	10
Flower	15,000	35-50
Green bolls	55,000	15
Undamaged open bolls	75,000	15
Damaged open bolls	15,000	40
Rotten bolls	100,000	35

a/ Using the criterion $n = (CV/30\%)^2$.

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LABORATORY STUDIES OF THE DEVELOPMENT OF THE PARASITE
ANAGYRUS PSEUDOCOCCI (GIRAULT)^{1/} ON INSECTARY-REARED
PLANOCOCCUS CITRI (RISSO)^{2/,3/}

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ABSTRACT

Anagyrus pseudococci (Girault) was found to prefer 3rd instar Planococcus citri (Risso) for production and survival of progeny. The average developmental time of the parasite was 15.5 days at 25.6°C and 60% RH, with virgin and mated females averaging 8.2 and 6.9 days longevity, respectively. Virgin females were found to produce only male progeny.

INTRODUCTION

The citrus mealybug, Planococcus citri (Risso), is a pest in orchards and greenhouses in most tropical, subtropical, and temperate regions of the world (McKenzie 1967). It is especially severe on grapefruit in the Rio Grande Valley of Texas (Dean et al. 1971). High mealybug populations cause extensive fruit drop and produce honeydew that leads to excessive growth of sooty mold, Capnodium citri (Berkeley & Desmazieres). The use of broad spectrum insecticides to control citrus mealybug often eliminates natural control agents (Meyerdirk et al. 1979) and induces rapid increases in mealybug populations that damage fruit severely. In 1970, grapefruit producers were advised to seek and preserve beneficial insects to promote control of this pest (Dean et al. 1971).

Studies in 1970 indicated that the only 2 native entomophagous species affecting P. citri in south Texas were Pauridia peregrina Timberlake (Encyrtidae) and Symphorobius barberi (Banks) (Hemerobiidae) (Dean et al. 1971). Since then, 2 other parasites have been observed: Leptomastix dactylopii Howard (Encyrtidae) and Anagyrus sp. near sawadai Ishii (Encyrtidae) (Meyerdirk et al. 1978). The latter species closely resembles Anagyrus pseudococci (Girault), a solitary internal parasite that attacks P. citri in many regions of the world. Anagyrus pseudococci is established in California (Bartlett and Lloyd 1958), Argentina (Compere 1939), Brazil (Bartlett and Lloyd 1958), Israel (Berlinger 1972), Sicily (Compere 1939), and the U.S.S.R. (Niyazov 1969). A laboratory culture of A. pseudococci (determined by E. E. Grissell) from California is currently maintained at USDA facilities in Weslaco, TX, for inoculative releases against P. citri.

Earlier studies in Israel of the biology of A. pseudococci indicated that offspring survival and female progeny ratios increased with increased host age and size as well as with temperature (Avidov et al. 1967). This information is vital for efficient rearing of the parasite. However, Israeli researchers, Rosen and Rossler (1966), reported that the parasites

^{1/} Hymenoptera: Encyrtidae.

^{2/} Homoptera: Pseudococcidae.

^{3/} Approved as TA No. 15525 by Director, Texas Agricultural Experiment Station.

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used in their morphological studies were different in structure and coloration from original descriptions of A. pseudococci. These differences were also noted in Israel by Avidov et al. (1967). Thus, the strain of A. pseudococci in Israel may be a distinct form or species of Anagyrus.

In view of the apparent discrepancies in the identification of these parasites, a study was designed to obtain accurate information on the biology of a California strain of A. pseudococci that was being considered for use in a mass rearing program. Herein we report information on survival, ovipositional behavior, and fecundity of these parasites.

METHODS AND MATERIALS

Four battery jars, each containing 4 sprouted potatoes infested with P. citri (at least 250/potato) were used to determine the host stage most heavily attacked and the parasite developmental time. Each jar was covered with fine mesh cloth, and contained either 1st-instar (6 days old), 2nd-instar (12 days old), 3rd-instar (18 days old), or adult (24 days old) mealybugs. Three hundred mated adult A. pseudococci females were placed in each jar, allowed to oviposit for 24 h, and then anesthetized with CO₂ and removed. After 7 days, 500 P. citri from each jar were selected at random and placed individually in gelatin capsules (size 000). Parasite emergence was recorded daily.

For determinations of the numbers of progeny produced by females, groups of 10 virgin or 10 mated A. pseudococci, all 48 h old, were confined to 0.473-L jars with at least 250 3rd-instar P. citri on a sprouted potato. Mated females were obtained by confining a newly emerged male with a newly emerged female in a gelatin capsule for 24 h. Disturbance of the female parasite was minimized by leaving them in the jars until they died; their progeny were counted and sexed 30 days later.

To determine when oviposition occurred, we placed groups of 10 virgin or 10 mated female parasites, all 48 h old, in 0.473-L jars as previously described. Each parasite was transferred daily to a fresh jar until it died. Progeny were counted 30 days later.

All experiments were conducted at 25.6°C and 60% RH under continuous artificial light. These conditions simulated those used in the insectary for parasite production.

RESULTS AND DISCUSSION

The rate of parasitization of P. citri by A. pseudococci was highest (67.8%) and the sex ratio closest to 50% (41.6%) when 3rd-instar host material was used (Table 1). Only 1 parasite developed per host. The different sex ratios of parasites reared from 2nd-instar and adult hosts might be the result of possible host size preference for laying of male or female eggs. We conclude that 3rd-instar P. citri are the most efficient insectary hosts. This conflicts with the report by Avidov et al. (1967) that the Israel strain of A. pseudococci produced more offspring in adult females with ovisacs.

TABLE 1. Parasitism and Sex Ratios of A. pseudococci in Relation to Age of P. citri.

Host stage	<u>P. citri</u> No. in Capsules	% Parasitized by		Sex ratio of parasites	
		<u>A. pseudococci</u>		% male	% female
1st instar (6 day old)	500	0.2		100.0	0.0
2nd instar (12 day old)	500	19.4		92.8	7.2
3rd instar (18 day old)	500	67.8		58.4	41.6
Adult (24 day old)	500	6.6		6.1	93.9

a/ 300 adult A. pseudococci/host stage.

Developmental times for A. pseudococci are presented in Table 2. The range was 12-27 days for males (avg 15.4 days) and 14-18 days for females (avg 15.6 days). Pooled mean developmental time for males and females was 15.5 days. In similar studies at 24° and 28°C, Avidov et al. (1967) found that A. pseudococci females of the Israel strain developed in 17.4 and 13.2 days, respectively; males developed slightly faster. They also found that age of the host did not affect duration of development. Our studies showed that developmental time of A. pseudococci on 3rd-instar hosts (avg 14.9 days), was not significantly different ($F = 0.6$) from 2nd instars (avg 17.1 days) and adults (avg 16.3 days).

Undisturbed virgin females produced 68.9 ± 12.7 adult offspring, whereas those transferred daily produced only 15.5 ± 4.2 . Our observations that all of the progeny of virgin females were males confirm reports by Avidov et al. (1967) that this species is arrhenotokous. Undisturbed mated females each produced 19.7 ± 2.6 female and 19.9 ± 3.9 male offspring while disturbed females each produced 9.3 ± 2.6 females and 19.9 ± 6.6 males. T-tests indicated that numbers of males and females produced were significantly different ($T = 2.32$) only in the experiments using disturbed mated females. The disturbance of the female could be a factor in the difference shown above.

Virgin and mated female parasites were found to oviposit eggs from 48 h after emergence until death, with adult life spans averaging 8.2 ± 1.3 and 6.9 ± 1.2 days after emergence, respectively (Table 3). Two peak oviposition periods were observed in virgin females. The 1st occurred at 3-4 days following adult emergence and a 2nd much smaller peak at 8-9 days. During the first 3 days of oviposition mated females produced more males than females. Sex ratios did not differ after day 3. No differences were noted between oviposition periods of virgin and mated females.

The above results indicate that insectary cultures of the California strain of A. pseudococci can be optimally maintained by confining newly emerged male and female parasites with 3rd instar P. citri host material. Adequate parasite oviposition can be expected any time after a postemergence period of 48 h, with the new parasite generation emerging approximately 2 wk after initial egg laying and continuing for an average of 7 days.

TABLE 2. Developmental Time of Anagyrus pseudococci Attacking Various Stages of Planococcus citri.

No. of adult parasites emerging daily on indicated days after oviposition													
Host stage	Parasite Sex	12	13	14	15	16	17	18	19	20	21 - 27	Total	
1st instar	Females	0	0	0	0	0	0	0	0	0	0	0	
	Males	0	0	0	0	0	0	0	0	0	1	1	
2nd instar	Females	0	0	0	0	1	5	1	0	0	0	7	
	Males	0	0	1	12	28	20	18	3	4	4	90	
3rd instar	Females	0	0	5	85	41	9	1	0	0	0	141	
	Males	1	25	102	50	12	2	1	2	0	3	198	
Adult	Females	0	0	0	3	16	10	2	0	0	0	31	
	Males	0	0	0	1	1	0	0	0	0	0	2	
Total	Females	0	0	5	88	58	24	4	0	0	0	179	
	Males	1	25	103	63	41	22	19	5	4	8	291	

TABLE 3. Ovipositional Period of Anagyrus pseudococci Attacking Planococcus citri.

Female progeny	Total progeny produced/day at indicated days after parasite postemergence														Avg/day	Total	
	1	2	3	4	5	6	7	8	9	10	11	12	13	14			
Virgin																	
males	7	6	30	19	11	8	11	23	21	11	7	0	0	1	11.07	155	
Mated																	
males	23	12	25	22	8	14	15	53	14	3	5	5	-	-	16.58	199	
females	0	0	1	10	11	6	11	19	20	7	1	6	-	-	7.75	93	
total	23	12	26	32	19	20	26	72	34	10	7	11			24.33	292	

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THE EFFECTS OF HORN FLY LARVAE ^{1/} AND DUNG BEETLES ^{2/} ON AMMONIA LOSS FROM BOVINE DUNG

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ABSTRACT

The presence of larvae of the horn fly, Haematobia irritans (L.), in cattle feces significantly ($P=0.05$) increased the loss of ammonia. The addition of adult brown dung beetles, Onthophagus gazella F., to the feces significantly ($P=0.05$) reduced the loss of ammonia from feces and made it more effective as a fertilizer.

INTRODUCTION

The influence of house flies, Musca domestica (L.), on loss of nitrogen, mainly present as ammonia, from bovine feces was determined by MacQueen and Beirne (1975). They reported that much of the lost nitrogen was incorporated into insect tissue; however, when feces were heavily infested, more nitrogen was lost than was incorporated. They also reported that up to 8.5% of the nitrogen in the feces could be lost because of house fly infestations. These losses reduce the maximum effective use of dung as a fertilizer and may contribute to non-point pollution.

The influence of pasture-breeding insects such as the horn fly, Haematobia irritans (L.), and dung-burying scarabs on ammonia loss from cattle feces has not been investigated. Since these insects are common in pasture ecosystems, their impact on recycling of plant nutrients should be determined. This information may aid in manipulating the insect fauna of cattle dung dropped on pasture so as to minimize nitrogen loss and lower the production of horn flies and face flies, Musca autumnalis De Geer.

METHODS AND MATERIALS

Cattle dung pats (200 g) were held in 250-mm-diameter vacuum desiccators that were partly filled with sandy loam soil (75 mm deep). The pats were collected fresh from a steer fed only on alfalfa cubes that contained about 16% protein. The feces contained about 2.8% protein (wet weight, 80% moisture). Adult insects and/or insect eggs obtained from laboratory colonies were placed on top of the dung. Two species of insects were tested; the horn fly and the brown dung beetle Onthophagus gazella F.

The amount of ammonia released from the various pats of dung was determined by trapping as described by Fenn and Kissel (1973). Thus air was drawn via a vacuum from the desiccator through a 250-mL stoppered graduated cylinder containing 200 mL of 2% boric acid solution. A fritted gas dispenser dispensed the air in the boric acid solution. Two airflow rates were tested 1 at 700 mL/min and the other at 7 L/min. The airflow was measured with flow meters on the intake of the desiccator. The air was bubbled through distilled water before entering the desiccators when the 7 L/min airflow was used due to excess drying of the dung. A 50-mL aliquot of the boric acid solution was titrated with 0.01N HCl (in the presence of 8 drops of 0.04% bromocresol green indicator) to determine the ammonia content. We studied the loss of nitrogen from pats that

^{1/}
^{2/} Diptera: Muscidae
Coleoptera: Scarabaeidae

consisted of dung only, of dung + 4 pair of O. gazella, of dung + 500 horn fly eggs, and of dung + 4 pair of O. gazella + 500 horn fly eggs. All dung beetles were 5-7 days old. Losses were measured for 14 days. All tests were replicated 6 times. Two-way analysis of variance and the Q test (Snedecor 1957) were used to determine whether means were significantly different.

RESULTS AND DISCUSSION

In the test conducted with an airflow rate of 700 mL/min, the presence of horn fly larvae significantly ($P=0.05$) increased ammonia loss; addition of O. gazella + horn flies significantly ($P=0.05$) reduced this loss to almost the same level as that from dung alone, and the addition of O. gazella alone also significantly ($P=0.05$) reduced the loss from dung (Table 1).

TABLE 1. Effects of Horn Flies and O. gazella on Ammonia Loss From Bovine Dung (Airflow 700 mL/min, 200 g Dung, 4 Pair O. gazella., 500 Horn Fly Eggs).

Reps	mg ammonia measured ^{1/} from			
	Dung	Dung + <u>O. gazella</u>	Dung + horn flies	Dung + horn flies + <u>O. gazella</u>
1	14.6	16.6	36.1	21.0
2	23.1	14.2	31.9	20.3
3	17.2	7.6	32.0	29.2
4	20.3	16.2	37.1	26.6
5	17.0	7.7	20.3	15.3
6	11.9	5.1	19.1	5.7
Mean ^{2/}	17.4a	11.2b	29.4c	19.7a

^{1/} Measured for 14 days.

^{2/} Means followed by same letter not significantly ($P=0.05$) different.

When the test was conducted with an airflow rate of 7 L/min (Table 2), the amount of ammonia trapped was doubled; however, the loss pattern was the same except the amount of ammonia trapped from the dung containing only dung beetles was not significantly different from dung alone. These results show a consistent pattern of ammonia loss due to insect activity. Even though the quantities lost were not great, at least 140 mg ammonia per kg of dung can be conserved by dung

TABLE 2. Effect of Horn Flies and O. gazella on Ammonia Loss From Bovine Dung (Airflow 7 L/min, 200 g Dung, 4 Pair O. gazella, 500 Horn Fly Eggs).

Reps	mg ammonia measured ^{1/} from			
	Dung	Dung + <u>O. gazella</u>	Dung + horn flies	Dung + horn flies + <u>O. gazella</u>
1	47.4	40.8	86.7	43.0
2	37.2	33.8	57.6	38.1
3	38.9	28.6	42.5	33.6
4	44.0	43.9	60.0	54.5
5	31.7	29.0	62.9	34.4
6	31.3	25.0	57.8	21.5
Mean ^{2/}	38.4a	33.5a	61.3b	37.5a

^{1/} Measured for 14 days.

^{2/} Means followed by the same letter are not significantly ($P=0.05$) different.

beetle activity. This could amount to about 454 g ammonia per cow per 120-day grazing season or a potential of 57 million kg/yr in the United States.

Many other species of insects are present in cattle dung, and their interaction as it relates to loss of nitrogen and other plant nutrients is not known. This information is needed before we can manipulate the insect fauna in cattle dung in the pasture ecosystem to reduce dipteran pests (horn fly and face fly) and to use the dung to increase soil fertility.

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AUTOMATIC TRAP FOR DETERMINING HOURLY FLIGHT ACTIVITY OF DUNG BEETLES^{1/2/}

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ABSTRACT

The design, construction and operation of a battery-operated, 24-container trap for the automatic collection of bait-attracted dung beetles (Scarabaeidae) on an hourly basis is described. The trap was developed to facilitate studies of the natural flight activity patterns of dung beetles at remote field locations.

INTRODUCTION

As part of ongoing research to enhance biological control of the horn fly, *Haematobia irritans* (L.), detailed information was needed on the daily flight patterns of dung beetles (Scarabaeidae) which are among the important natural competitors and predators of the horn fly (Blume et al. 1973, Anderson and Loomis 1978, and Harris and Oliver 1979). Increased knowledge of the native beetles' flight activities and the effects of environmental factors such as temperature, humidity, wind speed and day length on their behavior is necessary to aid in determining what foreign species of dung beetles might be introduced to aid in suppressing horn fly populations.

To acquire the needed information, we desired a system that would trap the beetles in numbers proportional to their normal flight activity on an hourly basis under conditions as nearly natural as possible. The system required a bait placed at ground level for attracting dung beetles and a buried electro-mechanical device for trapping them. Fincher et al. (1970) found that dung beetles were readily attracted to fresh swine feces, so we selected this material to be used as bait. Design criteria established for the electromechanical trap were that it have capacity for hourly separation of beetle catches over a 24-h period, that it be suitable for direct burial in the soil under the attracting bait, that it be battery operated, and that the batteries have a service life of at least 2 wk without requiring recharging.

Several types of time-interval traps designed for collecting various other species of insects are described in the literature (Hartstack and Hollingsworth 1968, Smith et al. 1973, and Goodenough and Snow 1979). All of these traps were characterized by time-sequencing operation, but none met all the criteria we required for the dung-beetle trap. Fincher and Stewart (1968) described a trap for collecting dung beetles, but it separated hourly catches for only 12 h and it required a 115-vac power source for operation.

This paper describes the design, construction and operation of a battery-operated, 24-container trap for the automatic collection of bait-attracted dung beetles on an hourly basis at remote field locations.

^{1/} Coleoptera:Scarabaeidae.

^{2/} Trade names are used solely for the purpose of providing specific information. Mention of a trade name does not constitute a guarantee or warranty of the product by the U.S. Department of Agriculture or an endorsement by the Department over other products not mentioned.

MECHANICAL DESIGN AND CONSTRUCTION

Trap construction was begun by attaching 24 collection container holders to a roller chain (3.1 m length, 1.3 cm pitch) with 12.7-cm spacing between centers (Fig. 1). The container holders were cylindrical sleeves, 15.2 cm long and 10.2 cm ID, made from polyvinylchloride pipe. The chain was mounted on 2 horizontal 30-tooth sprockets each supported by a 1.3-cm-diameter steel shaft and a pair of pillow blocks attached at each end of an angle iron frame. A 6.4-mm-diameter steel rod was shaped and welded to the base supports of the angle iron frame to serve as a support track for the sleeves. A 12-vac electric motor (electric window lift motor; J. C. Whitney & Co., Chicago, IL), connected to one of the 1.3-cm shafts by means of a V-belt and 3:1 speed reduction pulleys, furnished rotational power to position the sleeves (Fig. 2). The motor was powered by a 12-v, automotive-type lead-acid battery and controlled by an electronic timing circuit described below. Indexing for position control was obtained by micro-switch sensing of 3 cams located with 120° radial spacing on the horizontal surface of the drive sprocket.

Plastic cylinders, 10 cm in diameter and 14 cm deep, with screen bottoms, were used as collection containers within each sleeve. A plastic funnel, 10.2 cm in diameter by 7.6 cm high, was placed in each collection container to facilitate insect capture and to inhibit escape.

The completed mechanism was mounted in a 45.7 X 60.7 X 243.8-cm rectangular enclosure constructed of 1.3-cm plywood. A large metal funnel of 25.4-cm-diameter was inserted through a 20.3-cm-diameter hole in the top of the enclosure. The funnel was supported by a sheet metal neck extension from the enclosure hole so that its 10.2-cm-long stem was positioned 4 cm above one of the collection containers. A screened hole in the floor of the trap enclosure allowed for rainwater drainage.

A cylindrical container (ca. 2 L in volume) with a solid bottom and wire-mesh screen (24 squares/cm) on the sides and top was provided to hold the bait. The container was positioned on a wire support in the center of the large funnel with its bottom recessed about 3 cm into the funnel opening. An annular open space, provided by a clearance of ca. 5 cm between the bottom peripheral edge of the bait container and the funnel walls, allowed attracted insects to fall through the funnel into the collection containers. The screen sides and top of the bait container allowed the odor of the bait (ca. 0.5 kg, positioned in a cone-shaped mass in the center of the container away from contact with any of its sidewalls) to permeate the area in the vicinity of the trap.

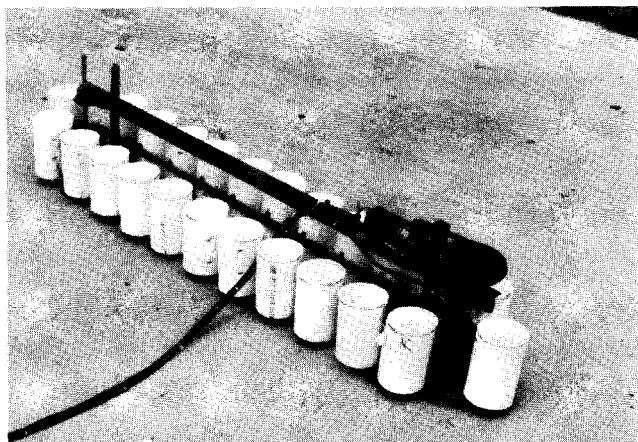


FIG. 1. Basic trap mechanism. See text for description of components.

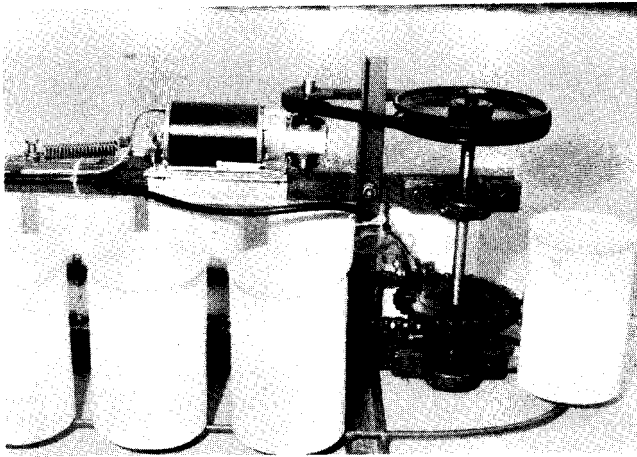


FIG. 2. Detail of trap drive and control mechanisms. One cup holder was removed to facilitate view.

DESCRIPTION OF TIMING AND CONTROL CIRCUITS

The electronic circuit, shown schematically in Fig. 3, was designed and built for controlling the movement and positioning of the collection containers. Integrated circuits A2 and A4 are NOR logic gates connected to form a multivibrator. In normal operation of the complete circuit, the multivibrator functions in a bistable (flip-flop) mode. When the circuit is in the "OFF" state, both inputs to A2 and one of the inputs to A4 are at the low logic level because of the effects of resistors R3, R5 and R6. In this state, the output of A2 is high and the output of A4 is low. Thus, transistors Q1 and Q2 are cut off, the relay is inactivated with its switch open, and no power is delivered to the motor.

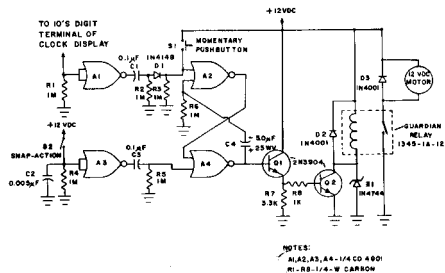


FIG. 3. Schematic of electronic control circuit.

The "ON" or active state for the circuit is triggered when the output from the time clock to the inputs of A1 changes from a high to a low logic level. This transition causes the output of A1 to become high and a positive signal to be transmitted via C1 and D1 to an input of A2. The positive signal at the input of A2 causes the flip-flop, composed of A2 and A4, to change states and the output of A4 to become high. The high output of A4 activates transistors Q1

and Q2. Thus, the relay is activated and power is delivered to the drive motor. After 1/3 revolution of the drive sprocket a cam on the sprocket opens micro-switch S2, thereby allowing low inputs to A3. The low signal at the inputs of A3 causes its output to go high, and a positive signal to be transmitted via C3 to an input of A4. This signal resets the flip-flop, turning Q1 and Q2 off, deactivating the relay and removing power from the motor. The flip-flop is thus returned to its inactive state in which it remains until another timing pulse is delivered to the inputs of A1 by the time clock.

The RC circuit, formed by resistor R6 and capacitor C4, serves a safety function by causing the multivibrator to operate in a monostable mode if no signal indicating that the mechanical repositioning of the containers has been completed is received from S2 within a set time. When the flip-flop is driven to the active state by a timing signal from the clock, it will remain in this state only for a period ca. equal to the RC time constant of $R6 \times C4$ even if no signal is received from S2. Reversion of the flip-flop to its inactive state at the end of the RC time period cuts off the power to the motor and prevents excessive battery drain in the event of electrical or mechanical malfunction. We selected values for R6 and C4 that would allow the resulting RC time to be ca. twice the time required for repositioning of the trap collection containers in normal operation.

The control circuit is manually activated for servicing the trap by momentary closure of switch S1. This push-button switch is mounted in a small chassis separate from the control circuit and is electrically attached by means of a 2-conductor cord of convenient length so it can be operated remotely from an area adjacent to the trap. Closure of S1 furnishes a positive pulse to one input of A2, thereby activating the control circuit. The response of the circuit is the same whether activated automatically by the time clock or manually by momentary closure of S1.

A Model MA1003 12-vdc automotive/instrument clock module (National Semiconductor Corp., Santa Clara, CA.) was used as a source of (clocked) timing pulses for the control circuit. (A data sheet, including circuit diagrams, is furnished by the manufacturer with the clock; therefore, circuit diagrams for the clock are not given here.) The MA1003 clock was not designed for generation of clock pulses, so certain modifications to the original circuitry were necessary. An hourly low-logic-level pulse was obtained, on the hour, for input to NOR-gate A1 by connecting the inputs of A1 to the g-segment drive terminal (pin 10) of the 10's-minute-digit of the display.

For reduction of power consumption during normal operation, the printed circuit of the clock module was modified to allow blanking of the fluorescent display without interrupting the clock pulse output. In this modification (this description refers to the manufacturer's circuit diagram, which is not illustrated here), the printed circuit line for the positive filament supply was broken between the connecting points for one leg of the display switch and diode CR3, and a single-pole, single-throw (SPST) toggle switch was inserted to control the display. With the SPST switch closed, continuity was restored to the filament supply line, and the display was activated for setting the clock and monitoring the timing function. In normal operation, the SPST switch was in the open position; therefore, the filament supply line was open and the display was blanked. The timing control circuit required about 70 ma with the display on and less than 3 ma with the display off.

Although the timing control circuit and the trap drive motor could be powered from the same battery, a separate 12v dry cell was used for the control circuit to prevent interruption of the timing function when the lead-acid battery, used to power the drive motor, was removed for recharging. A NEDA-type 922 12v dry cell powered the control circuit for about 6 months before replacement was required.

The trap drive motor required ca. 3 amp for ca. 5 sec each time the drive mechanism was activated. The 12v lead-acid battery used to power the trap theoretically had capacity to operate for 3-4 wk without recharging, but we

recharged it biweekly to avoid possible damage due to excessive discharge. A marine-type deep-discharge battery should be used if extended periods without recharging are required.

INSTALLATION, OPERATION, AND SERVICING

The enclosed mechanical trap was buried in a cattle pasture with ca. 15 cm of soil and grass sod covering the top to give the trap surroundings a natural appearance. Only the bait container of the trap mechanism was visible above the pasture surface. The battery and control circuit were placed 1.4 m above ground in a rain shelter ca. 3 m from the burial site and were connected to the trap mechanism with the necessary electrical cables.

Beetles that were attracted to the bait approached the trap by flying or walking and fell through the large funnel into a collection container. Once each hour, a different collection container was automatically positioned under the large collection funnel.

For daily trap servicing, the bait container and the large collection funnel were removed from the porthole in the top of the buried trap enclosure to gain access to the collection containers for insect removal. Individual collection containers were removed and the trapped insects were transferred to temporary holding containers for later identification, counting, and subsequent release. After each collection container was replaced, the drive mechanism was advanced 1 step by momentarily depressing remote control switch S1 (Fig. 3). All 24 collection containers were emptied and replaced in a like manner. Lastly, the large collection funnel and bait container with fresh swine feces were set back in place, thus completing the daily service.

Two traps, constructed and used in field experiments for ca. 9 months, have given excellent, near trouble-free, service. With certain modifications, the trap could be useful for trapping other insect species and rodents or for periodic sampling of other phenomena.

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EFFECT OF LOW LEVELS OF GAMMA IRRADIATION ON LONGEVITY
AND STERILITY OF THE BOLL WEEVIL^{1/}A. S. M. Abdul Matin^{2/}, J. E. Wright^{3/}, and T. B. Davich^{3/}Boll Weevil Research Laboratory, AR, SEA, USDA,
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ABSTRACT

Dose-effect studies with a laboratory strain of *Anthonomus grandis* Boheman were conducted with gamma radiation from 2 sources delivered at 2 dose rates. The 6-day-old weevils were less sensitive than the 2- or 3-day-old weevils to the effects of radiation on oviposition. The 2 dose rates of 356 rad/min and 120 rad/min from ⁶⁰Co and ¹³⁷Cs, respectively, were equal in biological effects.

INTRODUCTION

Control or eradication of insects by release of radiation-sterilized males, an idea generally attributed to E. F. Knipling, has had spectacular success in some cases (Bushland 1960). With the boll weevil, *Anthonomus grandis* Boheman, the main problem has been to obtain an effective means of sterilization. Radiation doses low enough to permit adult survival for at least ten days failed to sterilize adults (Davich and Lindquist 1962, Mayer and Brazzel 1966). Chemosterilants have not proven satisfactory when tested against the boll weevil. Lindquist et al. (1964) reported that apholate caused high mortality at doses causing sterility, and that males treated with apholate were not competitive in the field. Busulfan, though effective in laboratory studies, has not been satisfactory in large-scale experiments such as the Pilot Boll Weevil Eradication Experiment conducted in 1972-73 (Lloyd et al. 1976). Boll weevils with lower bacteria content (<500 bacteria/weevil) can tolerate radiation better than those with higher bacteria content (> 100,000 bacteria/weevil) and antibiotics are now used for rearing weevils in the laboratory in large numbers (Sikorowski et al. 1977). The means to produce relatively clean weevils on a large scale and the effects of radiation on sperm and pheromone production as discussed by Earle et al. (1978) have prompted renewed interest in radiation as a means to obtain boll weevil sterility. This paper reports an investigation of the biological effects of a graded series of gamma radiation doses from 2 sources with different energy levels on the longevity and reproduction of adult boll weevils.

MATERIALS AND METHODS

Trays containing advanced stages of pupae about to emerge were obtained from the Robert T. Gast Rearing Laboratory at Mississippi State, MS, and were maintained at about 30°C and 60% RH. The adult weevils were sexed as they emerged and the sexes maintained separately. The sensitivity of male and female boll weevils to lethal effects of gamma radiation was evaluated with 2-, 3-, and 6-day-old weevils with the sexes held separately after treatment.

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Each treatment consisted of 20 insects of each sex with 3 replications. The treatments were 1250, 1570, 1980, 2480, 3140, 4970, and 6800 rads by Fricke dosimetry along with an unirradiated control. The doses were administered at a constant dose rate of 356 rad/min from a ^{60}Co source. Identical sets were exposed to similar radiation doses but they were delivered at a dose rate of 120 rad/min from a ^{137}Cs source to determine if doses given at the 2 dose rates from the 2 radiation sources would differ in the biological effects. The temperature maintained for both radiation sources was $28^{\circ} \pm 2^{\circ}\text{C}$. After treatment, each replicate of 20 insects was held in plastic cages (10 cm x 10 cm) and provided with plugs of adult diet which were changed daily. Mortality data were taken for 21 days, or more.

In addition, male and female 3- and 6-day-old boll weevils were treated with 1250, 1570, 1980, 2480, 3140, or 4970 rads from a ^{60}Co and the ^{137}Cs sources, respectively, and then pair mated (T ♂ x T ♀) to determine the effects of low levels of gamma radiation on fecundity and sterility. Fifteen pair matings for 6-day-old and 30 pair matings for 3-day-old adults were evaluated for each dose, including the respective controls (3 replications). Eggs were extracted from diet plugs as described by Vanderzant and Davich (1974). At 3-day intervals after treatment of the 3- and 6-day-old boll weevils, eggs were taken from each dose, surface-sterilized with 10% formaldehyde solution (Sikorowski et al. 1977), and then implanted in larval medium in petri dishes. The dishes were sealed with cellophane tape to minimize drying and were held for 3 wk before adult emergence was recorded.

Data were subjected to a probit analysis of dose vs mortality at 21 days after treatment according to Finney (1971). Ninety-five percent confidence limits were also computed for mortality.

RESULTS AND DISCUSSION

The effects of gamma radiation for mortality on boll weevils are given in Table 1. The highest of the doses used that would leave more than 50% survival at 21 days posttreatment was 1980 rads for the 2- and 3-day-old weevils and 2480 rads for the 6-day-old weevils. The ^{137}Cs -treated series of 6-day-old boll weevils resulted in higher mortality at 2480 rads (and, in fact, at all dose levels). All 2-day-old weevils treated with 4970 and 6800 rads were dead within 2 wk posttreatment. Survival of the 3-day-old boll weevils treated with radiation doses ranging from 1250 to 4790 rads from ^{137}Cs was not adversely affected at any dose level during the 7-day posttreatment period. Among 3-day-old boll weevils treated with ^{60}Co , females particularly showed increasing susceptibility as the radiation dose increased. Six-day-old weevils appeared to be more tolerant of radiation. After a dose of 2480 rads from ^{60}Co , we observed an average of 63% survival of mixed sexes at 3 wk posttreatment. Doses at the upper levels, however, caused higher mortality during the 2nd wk after irradiation. Thus, within the dose levels tested, 2480 rads was about the highest dose on 6-day-old boll weevils that would allow survival of more than 60% male and female weevils at 3 wk posttreatment.

Following irradiation from the ^{60}Co source, the lethal dose for 50% mortality (LD_{50}) for 2-day-old males was 2241 rads, 2304 rads for those 3 days old, and 2778 rads for 6-day-old boll weevils. The LD_{50} 's for the ^{137}Cs -irradiated 2-, 3-, and 6-day-old boll weevils were 2378, 2776, and 2351 rads, respectively. Statistical comparisons, however, showed that 2-, 3-, and 6-day-old males did not differ from one another in susceptibility to gamma radiation ($P < 0.05$).

TABLE 1. Mortalities of 2-, 3-, and 6-Day-Old Boll Weevils After Irradiation with ^{137}Cs and ^{60}Co . at 21 Days After Treatment.

Age at Irradiation	Dose rate (rad/min)	Male		Female	
		Dose (rad) for 50% Mortality and 95% confidence limit	Dose (rad) for 95% mortality	Dose (rad) for 50% mortality and 95% confidence limit	Dose (rad) for 99% mortality
2 days	120	2378	2300-2460	2265	1596-3454
2 days	356	2241	2052-2576	5899	-----
3 days	120	2776	2590-2992	5014	2407-2794
3 days	356	2304	2037-2854	6722	1799-2365
6 days	120	2351	2267-2438	3232	-----
6 days	356	2778	2684-2881	3805	2539-2765
				2645	4092

a/ ^{137}Cs source.

b/ Doses followed by the same letter are not significantly different ($P > 0.05$).

c/ ^{60}Co source.

TABLE 2. Effects of Low Levels of Gamma Radiation (^{60}Co Source) on Sterility of 3-Day-Old Boll Weevils.

Dose (rad)	Fecundity (no. eggs/?)										Fertility at indicated day after treatment g/					
	day after treatment										0 - 3		3 - 6		6 - 9	
	0-3	3-6	6-9	9-12	12-15	15-18	18-21	No. eggs tested	% Yield adults	No. eggs tested	% Yield adults	No. eggs tested	% Yield adults	No. eggs tested	% Yield adults	No. eggs tested
0	16.2	18.5	21.9	19.2	14.3	16.1	13.4	150	30.0	100	34.0	150	28.0	150	23.3	150
1250	15.8	15.6	14.7	13.5	8.5	9.0	8.4	100	19.0	150	7.3	150	19.0	150	17.0	150
1570	14.4	11.3	11.4	12.7	9.4	11.2	8.6	100	13.0	150	4.7	100	17.3	150	17.3	150
1980	15.4	7.3	6.7	7.5	7.6	8.5	9.3	100	10.0	110	1.8	150	0	24	-----	-----
2480	11.7	4.2	0.8	1.6	1.3	0.8	2.2	100	8.0	102	1.0	71	0	-----	-----	-----
3140	9.8	2.4	0	0	0	0	0	50	2.0	71	0	48	0	-----	-----	-----
4970	7.2	1.6	0	0	0	0	0	150	0.7	48	0	-----	-----	-----	-----	-----

a/ Sterilizing dose for 50% reduction in fertility, $\text{SD}_{50} = 1600$; $\text{SD}_{99} = 6547$.

TABLE 3. Effects of Low Levels of Gamma Radiation (^{137}Cs Source) on Sterility of 6-Day-Old Boll Weevils.

Dose (rad)	Fecundity (No. eggs/♀)							Fertility ^{a/}	
	at indicated	day after treatment ^{a/}						No. Eggs tested	% Yield adult
	0-3	3-6	6-9	9-12	12-15	15-18	18-21		
0	9.4	9.3	6.3	10.5	9.7	10.3	10.9	116	46.6
1250	9.8	9.0	7.9	13.1	11.1	12.4	8.5	96	29.2
1570	6.7	7.3	8.8	11.8	8.6	7.3	10.1	60	16.7
1980	5.7	3.3	1.5	5.3	4.9	4.3	4.0	47	25.5
2480	8.9	1.6	0.4	1.2	3.2	2.3	3.3	60	15.0
3140	7.3	1.2	0	0	0.7	0.5	0.3	56	10.7
4970	8.6	1.3	0	0	0	0	0	55	1.8

^{a/} Sterilizing dose for 50% reduction in fertility, $\text{SD}_{50} = 1581$; $\text{SD}_{99} = 10617$.

There was no difference in the effects between ^{137}Cs and ^{60}Co delivered at 2 dose rates of 120 rad/min and 356 rad/min, respectively ($P < 0.05$). Nauman et al. (1975) reported that increasing dose rates of irradiation are increasingly effective only up to ca. 100 rads/min. The 6-day-old males were more tolerant to the ^{60}Co irradiation than the 2-day-old males (Table 1). The differences between male and female boll weevils, though small, appeared to be consistent in this study. An earlier study by Haynes et al. (1977) did not detect any differential effects of radiation on longevity of male and female boll weevils.

The sterility data for the 3- and 6-day-old weevils are presented in Tables 2 and 3. Again the younger 3-day-old weevils were more affected by radiation than those 6 days old. After the 6th day of the treatment there was a complete cessation in egg laying by 3-day-old females treated with 3140 and 4970 rads. Flint et al. (1966) observed that female boll weevils treated with doses as small as 3200 rads did not produce eggs if they were irradiated as 1- to 3-day-old adults. They also observed that weevils actively laying eggs at the time of irradiation continued to lay eggs for ca. 5 days after treatment.

In this study, the 6-day-old weevils treated with 3140 rads, however, continued egg laying throughout the 21-day posttreatment period, though at a much reduced rate. There was a tendency for 6-day-old weevils treated with intermediate radiation dose levels (i.e., 1980 and 2480 rads) to recover as time passed.

Fertility, as measured by yield of adults from eggs, was more affected by radiation than fecundity, though there was a reduction in oviposition as radiation dose increased. The yield of adults was reduced at all dose levels. The highest dose, 4970 rad, caused a 98 and 96% reduction, respectively, in the emergence of the adult progeny of weevils when they were treated at 3 and 6 days of age. There was no differential sterilization response by 3- and 6-day-old boll weevils to different radiation doses. A 50% reduction in the fertility of 3-day-old weevils resulted from exposure to 1600 rad, compared to 1582 rad for the 6-day-old weevils (Tables 2 and 3). When the fertility of 3-day-old weevils were compared at 3, 6, and 9 days after irradiation with 1250, 1570, and 1980 rads, it was evident that there was some recovery of fertility at 9 days after treatment. Increased fertility with time after irradiation is considered evidence that early stage gametogenic cells are more radiation-resistant relative to those in more advanced developmental

stages (Reimann and Flint 1967). In a similar study with 7- 10-day-old boll weevils, Flint et al. (1966) found a comparable increase in fertility among mixed groups of male and female weevils irradiated with 800, 1600, 2400, and 3200 rads. In the present studies, eggs collected at day 6 after treatment from 3-day-old weevils irradiated with 3140 and 4970 rads did not yield any adult progeny. This agrees well with data provided by Earle and Nilakhe (1977) who observed that spermatozoa maturing at this time probably developed from irradiated spermatids which are about 10 times as sensitive as spermatozoa.

In the boll weevil the survival time does not seem to be independent of dose. However, there is a range from 3140 to 6800 rads through which the effects of doses appear to be relatively slight, and a definite pattern is discernible. This dose-response pattern was characterized by a high rate of mortality after 1 wk and death of a majority of the insects within 2 wk of the treatment.

A similar pattern was reported by Flint et al. (1966). Bartlett et al. (1968) reported that the rate at which boll weevil adults died was almost identical after exposure to 6400 and 12,800 rads. Reimann and Flint (1967) demonstrated that adult death after irradiation was caused by damage to the secretory epithelium in the midgut and disappearance of the regenerative cells of the midgut. As a result of these radiation-induced lesions, the weevils could either die of undernourishment or the material from the lumen of the intestinal tract could leak out into the body cavity. The latter condition could result in an attack of intestinal enzymes on normal body tissue or an electrolyte imbalance, or both (Dieckoff 1972). Also, bacterial contamination could increase and cause mortality.

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CHEMICAL CONSTITUENTS OF PECAN LEAVES AS POSSIBLE INDICATORS
OF SUSCEPTIBILITY TO THE BLACKMARGINED APHID^{1/2/}W. W. Neel^{3/}, P. A. Hedin^{4/} and T. L. Carpenter^{5/}

ABSTRACT

Analyses of leaves collected in June 1979 showed that concentrations of juglone and nitrogen free extracts (NFE) in the leaves of 10 commercial pecan cultivars were positively correlated with infestation levels of the blackmargined aphid (Monellia caryella (= costalis) (Fitch)), whereas fiber percentage was negatively correlated. Analyses of leaf samples collected from these same cultivars in August showed no significant correlations between the content of juglone, NFE, and fiber when compared to aphid infestation levels.

INTRODUCTION

Damage to pecan leaves caused by the yellow aphids, Monelliopsis nigropunctata (Granovsky), and the blackmargined aphid, Monellia caryella (= costalis) (Fitch), has been noted by many investigators in the past. Recently Tedders (1978) reported that damage by aphids increased in severity with leaf age and was most severe in late summer. Apparently no surveys have been made to determine levels of resistance of the common commercial cultivars to these aphids. Gentry et al. (1975) found comparable infestations of M. caryella and M. nigropunctata on 'Stuart' and 'Schley' cultivars throughout the early growing season, but significantly more aphids were found on 'Schley' during the mid-summer (June 14 to August 9) when the infestation was lowest for both cultivars. However, in mid-September the infestations became greater on Stuart and remained greater for the rest of the growing season (September 19 to November 11), presumably because these leaves constituted a more desirable feeding site in that they had experienced less pecan scab fungus damage.

In a recent host plant resistance study Carpenter et al. (1979) demonstrated that 5 pecan cultivars were infested with significantly different levels of M. caryella, but did not show that juglone contributed significantly to resistance. The reasons for investigating juglone as a resistance factor were that it has been isolated from pecan leaves and found to be a possible factor of resistance to pecan scab, Fusicladium effusum Wint. (Hedin et al. 1979), it was described as an allelopathic agent in walnut for other plants (Brooks 1951), and it was found to be a deterrent to feeding by Scolytus multistriatus (Marshall) (Gilbert et al. 1967).

The present study was conducted to compare populations of blackmargined aphids on different cultivars of pecan, and to determine if differences in population levels were related to differences in some important constituents of the leaves.

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MATERIALS AND METHODS

The different cultivars of pecan used for the tests included: Delmas, Frotscher, James, Odom, Pabst, Schley, Stevens, Stuart, Success and Van Deman. We collected 35 compound leaves (ca. 100 g fresh tissue) from 1 tree of each cultivar and made chemical determinations of juglone, protein, fiber, ash, fat, nitrogen free extract (NFE), and sugars. Leaf samples were collected on June 26, and August 10 and 31, 1979. All trees were located in the 2.8-ha Anderson Pecan Grove on the Mississippi State University campus. None of the trees had received insecticide, fungicide, or fertilization treatments during the previous 10-yr period.

Natural *M. caryella* infestation counts were recorded from 25 compound leaves from each of these cultivars on July 2, 1979. In addition, cage studies were also conducted by placing 5 adult aphids that had been collected in the orchard in nylon sleeves (1 compound leaf/sleeve and 3 sleeves/tree) on July 2. The dimensions of the sleeves were 25 x 50 cm (Fig. 1). This procedure was



FIG. 1. Nylon mesh sleeve used to cage aphids during August.

necessary to insure an infestation because of losses due to predation and/or migration of the aphids during July and August. The sleeves were removed and the aphids were counted on August 10. A 2nd sleeve test was initiated on August 10 and final aphid counts were made on August 31.

The leaf samples were dried, ground to a powder, and then subsamples (15 g each) from each cultivar were submitted to the Mississippi State Chemical Laboratory for analyses to determine the percentage protein, fiber, ash, fat, and NFE. Juglone determinations were made by the method of Hedin et al. (1979). These analyses (1) involved extraction of the leaves, (2) separation of plant components by thin layer chromatography, and (3) determination of juglone concentration by spectrophotometric measurement.

The data were analyzed to determine the correlation coefficients (r) and degree of significance (Steel and Torrie 1960) for the relationship between aphid infestations and concentrations of juglone, protein, fiber, ash, fat, and NFE.

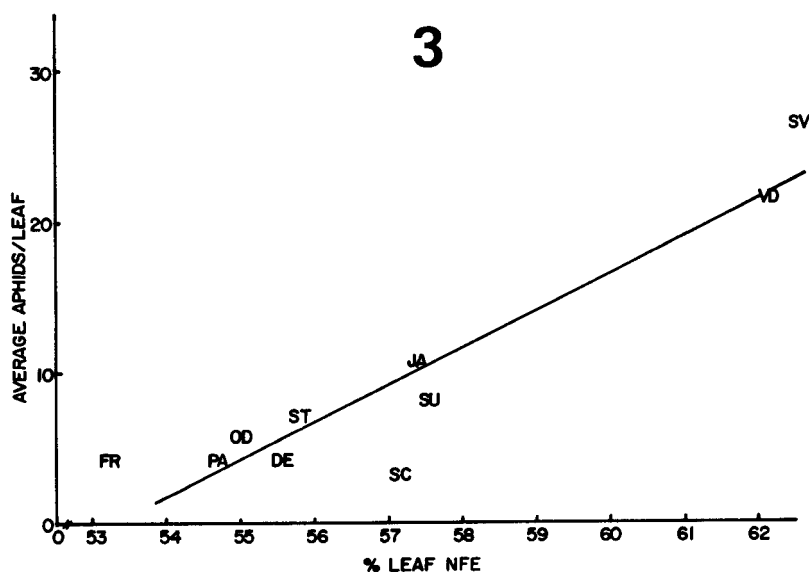
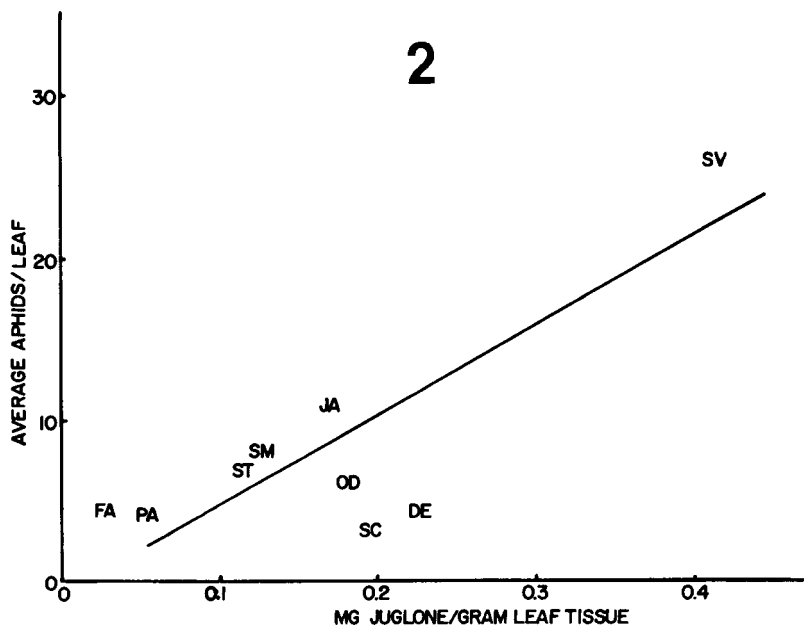
RESULTS AND DISCUSSION

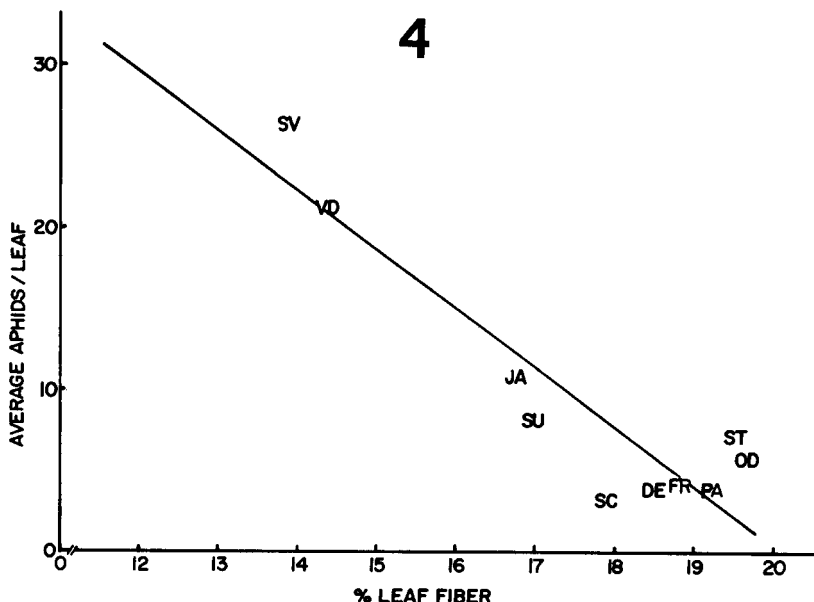
The coefficients of correlation determined on these experimental data indicated that there were significant interactive relationships between leaf fiber and NFE (among the 10 different cultivars) and aphid levels from samples collected on July 2; juglone content was highly correlated but the coefficient was not significant at the 0.05 level of probability (Table 1). The levels of

TABLE 1. Chemical Components of Pecan Leaves from 10 Cultivars and Natural Blackmargined Aphid Population Levels, July 2, 1979, Anderson Pecan Grove, Mississippi State University.

Cultivar	Juglone mg/g	Fiber %	NFE %	Avg. no. aphids per compound leaf
Van Deman	0.35	14.2	62.1	21.9
Stuart	0.12	19.5	55.8	7.0
Frotscher	0.02	18.7	53.1	4.3
Pabst	0.05	19.1	54.8	4.1
Schley	0.19	17.8	57.1	3.0
Success	0.13	17.0	57.5	8.0
James	0.17	16.8	57.5	10.8
Odom	0.18	19.6	55.0	5.6
Delmas	0.22	18.6	55.5	4.1
Stevens	0.41	13.9	62.6	26.3
$r =$	0.78	-0.92	0.93	

protein, ash, and fat were not significantly correlated with aphid numbers. Fiber percentage was negatively correlated ($r = -0.921$), that is the lowest numbers of aphids were found on those cultivars with highest fiber values (Fig. 4). The positive correlations for juglone ($r = 0.78$) and NFE ($r = 0.926$) indicated that the highest numbers of aphids were found on those cultivars with highest juglone and NFE values (Figs. 2 and 3).





FIGS. 2-4. Graphs showing: 2. Positive correlation, leaf juglone and aphids. 3. Positive correlation leaf NFE (nitrogen free extract) and aphids. 4. Negative correlation, leaf fiber and aphids for the following pecan cultivars: Delmas (DE), Frotcher (FR), James (JA), Odom (OD), Pabst (PA), Schley (SC), Stevens (SV), Stuart (ST), Success (SU), and Van Deman (VD). Leaf samples for chemical analyses were taken on June 26, 1979, and aphid counts were made on July 2, 1979.

Aphid counts on August 10 and 31 were made under conditions different from those made on July 2. Since it is likely that enclosure of the compound leaves in sleeves caused a change in environmental conditions, comparisons of data on aphid numbers obtained under natural conditions (July 2) with those obtained under sleeved-leaf conditions (August 10 and 31) probably are not valid. However, the comparison of numbers of sleeved aphids among cultivars does assess the ability of the aphids to feed and reproduce on the respective cultivars, so those data are valid for each individual date. No aphids were found on leaves that were unprotected on the 2 sample dates in August.

The data show that levels of juglone declined slightly for most of the cultivars as the season progressed. Apparently there was an error, either in field sampling or in laboratory procedure, in the juglone analyses of the Van Deman cultivar on July 2 (0.35 mg/g) and the Delmas cultivar on August 31 (0.02 mg/g). Since these juglone levels were considerably out of line with those determined for the same 2 cultivars on the other sample dates, they were omitted in calculating the correlation coefficients on the 1st and last sampling dates. In most cases, an increase in the percentage of fiber of the leaves of the different cultivars was noted as the season progressed. Toughness of the leaves as a result of a high content of fiber can result in resistance (van Emden 1973). Most NFE levels for the different cultivars declined during the same period.

Data for juglone, fiber, and NFE content of leaves from each cultivar, when compared to aphid numbers on August 10 and 31, gave no evidence of significant correlations (Tables 2 and 3). Unmeasured chemical and physiological changes due to leaf aging may account in part for the absence of any correlation between juglone, fiber, and NFE content and aphid population levels on these later dates.

TABLE 2. Chemical Components of Pecan Leaves from 10 Cultivars and Black-margined Aphid Population Levels in a Sleeve Cage Study, August 10, 1979, Anderson Pecan Grove, Mississippi State University.

Cultivar	Juglone mg/g	Fiber %	NFE %	Avg. no. aphids per compound leaf
Van Deman	0.05	17.9	59.9	27.3
Stuart	0.14	22.9	53.1	11.3
Frotscher	0.02	20.7	53.2	20.3
Pabst	0.05	20.3	53.6	7.3
Schley	0.19	22.5	52.2	1.0
Success	0.07	21.6	54.2	0.0
James	0.16	20.6	52.8	27.0
Odom	0.14	19.2	55.7	0.0
Delmas	0.22	21.1	56.4	11.7
Stevens	0.48	19.1	57.1	6.3
r =	-0.25	-0.33	0.19	

TABLE 3. Chemical Components from Pecan Leaves from 10 Cultivars and Black-margined Population Levels in a Sleeve Cage Study, August 31, 1979, Anderson Pecan Grove, Mississippi State University.

Cultivar	Juglone mg/g	Fiber %	NFE %	Avg. no. aphids per compound leaf
Van Deman	0.03	18.6	59.1	23.3
Stuart	0.08	21.7	50.4	54.0
Frotscher	0.03	22.9	51.2	0.0
Pabst	0.06	17.6	57.5	80.0
Schley	0.08	19.3	54.8	0.7
Success	0.04	19.0	55.1	0.3
James	0.03	19.8	53.0	86.3
Odom	0.13	20.7	52.0	0.0
Delmas	0.02	20.9	53.5	2.3
Stevens	0.34	18.3	57.1	54.7
r =	0.25	-0.35	0.19	

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ACTIVITY OF CERTAIN ALKYL AND ARYL PHOSPHORAMIDES
AGAINST HELIOTHIS SPP.^{1/2/3/}D. A. Wolfenbarger^{4/} and J. A. Harding^{5/}

ABSTRACT

Three aryl or alkyl organophosphoramido compounds, BAY 93820 [isopropyl salicylate O-ester with O-methyl phosphoramidothioate], BAY 91273 [isopropyl salicylate O-ester with O-ethyl phosphoramidothioate] and acephate, showed promising activity when applied topically to larvae of the tobacco budworm, Heliothis virescens (F.). Results of several years of field tests conducted in the Lower Rio Grande Valley of Texas showed that applications of acephate at 1.12 kg/ha significantly reduced square and boll damage by Heliothis spp.

INTRODUCTION

Resistance of the tobacco budworm, Heliothis virescens (F.), to organophosphorus (OP) insecticides has been recognized (Wolfenbarger et al. 1973) since 1969. Since the early 1970's, in the effort to find effective insecticides, various phosphoramido compounds have been tested for toxicity against the tobacco budworm. In laboratory tests, Wolfenbarger (1970) demonstrated that 2 such compounds, methamidophos and BAY 65250 (O-ethyl S-methyl phosphoramidothioate) were active against the tobacco budworm and the bollworm, H. zea (Boddie). Similarly in the laboratory, Plapp (1972) found that acephate and methamidophos were ca. equally toxic to the same 2 species and Bull (1979) demonstrated that these 2 phosphoramido compounds were only slightly less toxic to OP-resistant tobacco budworms than to a susceptible strain.

In 1971, extensive field tests of an aryl-substituted phosphoramido, BAY 93820 [isopropyl salicylate O-ester with O-methyl phosphoramidothioate], for control of the Heliothis spp. complex on cotton indicated (Cowan and Davis 1972, McGarr et al. 1972) the compound significantly reduced Heliothis spp. damage in 2 of 3 tests. Although acephate was recently registered for use in controlling Heliothis spp. on cotton, there have been no published reports of its efficacy against those pests in the Rio Grande Valley of Texas. Herein, we report the results of tests of the toxicity of acephate and 2 other phosphoramido compounds to the tobacco budworm in the laboratory, and the results of field tests of acephate in the Lower Rio Grande Valley of Texas during the period 1972-76.

^{1/} Lepidoptera: Noctuidae.

^{2/} This paper reports the results of research only. Mention of a pesticide in this paper does not constitute a recommendation for use by the USDA nor does it imply registration under FIFRA as amended. Also, mention of a proprietary product in this paper does not constitute an endorsement of that product by the USDA.

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MATERIALS AND METHODS

Laboratory Tests. Topical applications of acephate, BAY 93820, and BAY 91273 [isopropyl salicylate O-ester with O-ethyl phosphoramidothioate] were made to 3rd-stage tobacco budworm larvae, and topical, oral, and injected applications of acephate were made to 5th-stage tobacco budworm larvae. The methods for the topical treatments were described by Wolfenbarger and McGarr (1970), the oral treatments by Bull and Lindquist (1966), and the injection treatments by Wolfenbarger et al. (1968). The test insects, 30-50 in each of 2-3 replicates/test were selected at random from an OP-susceptible strain that had been reared continuously in the laboratory since 1970.

Sprays of BAY 93820 and acephate were applied to cotton leaves that had been removed from the plants, and then 3rd-stage larvae were placed on the treated leaves and allowed to feed. The effects of the compounds on the larvae were evaluated according to methods of Wolfenbarger and Redfern (1969). All mortalities were taken 48 h posttreatment.

Field Tests. Between July 12 and 30 of each test year, acephate was applied 2-3 times with a self-propelled high-clearance sprayer (Test 1) to cotton planted April 1 in plots of 2 or 4 rows, 1 m apart and 17-90 m long. A total of 21 L/ha was applied downward from a distance of 0.3 m above the tops of the plants through 3 flat fan nozzles/row.

During the period 1973-75, different amounts of acephate were applied to cotton by aircraft in a total volume of 11-21 L/ha in plots 1.12-4 ha in size. Applications were made 2-5 times between June 6 and July 13.

The effects of sprays were evaluated by inspecting 100 squares or bolls for *Heliothis* spp. damage on 3-7 sampling dates per test. Data were subjected to analysis of variance, and the means for the averages were separated by Duncan's new multiple range test at the 5% level of significance.

RESULTS AND DISCUSSION

Laboratory Tests. As a result of the topical applications to 3rd-stage larvae, the LD₅₀ values for acephate, BAY 93820, and BAY 91273 were 0.041, 0.021, and 0.046 mg/g, respectively. By comparison, earlier tests against 3rd-stage larvae from the same laboratory strain indicated that the topical treatment LD₅₀ values for methamidophos and BAY 65258 were 0.15 mg/g (Wolfenbarger 1970) and for methyl parathion 0.043 mg/g (Wolfenbarger 1972).

When 5th-stage larvae were treated with acephate topically, by injection or orally, the LD₅₀ values were 0.35, 0.062, and 0.19 mg/g, respectively.

At 48 h after tobacco budworm larvae consumed cotton leaves sprayed with acephate at the rates of 0.001, 0.01, and 0.05% wt/vol, mortalities were 30, 40, and 65% for the respective doses; similar applications of BAY 93820 caused mortalities of 0, 10, and 70%.

Field Tests. At the time of the year acephate was evaluated against the *Heliothis* spp. complex, 80% or more of the population was estimated to be composed of the tobacco budworm and 20% or less of the bollworm. In all tests during the period 1973-75, the mean percentages of damaged squares and bolls of cotton sprayed with acephate at 1.12 kg/ha were significantly lower than the mean percentages of damaged squares and bolls in the untreated check (Table 1). In 1976, damage to fruit was significantly lower in plots sprayed with acephate at a rate of 0.84 kg/ha than in plots untreated or treated with acephate at a rate of 0.56 kg/ha. Hence, we conclude that the recommended rate of 1.12 kg/ha should be effective in protecting cotton against the tobacco budworm in the Lower Rio Grande Valley of Texas.

TABLE 1. Field Tests of Acephate Against *Heliothis* Spp. in Cotton, Weslaco, TX.

Mean % damaged ^{a/}								
Squares					Bolls			
1972	1973	1974	1975	1976	1973	1974	1976	
Test	Test	Test	Test	Test	Test	Test	Test	Test
1	1 2	1 2	1 2	1	1	1 2	1	
0.56				74 b				88 b
0.84				57a				73a
1.12	13a	7a	22a 6a 11a 3a		2a	10a 6a		
1.34		2a						
1.68	12a	12a			1a			
Check	20a	34 b 12 b 67 b 24 b 32 b 10 b		95 b	16 b	62 b 17 b 95 b		

^{a/} Means followed by the same letter for each year and test are not significantly different from each other by Duncan's multiple range test at 5% level probability.

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EVALUATION OF THREE SELECTED INSECTICIDES FOR CONTROL OF
THE SOUTHWESTERN CORN BORER^{1/} IN FIELD CORN^{2/}E. D. Bynum, Jr., C. R. Ward^{3/}, and T. L. ArcherTexas Agricultural Experiment Station
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ABSTRACT

In 1976, aerial applications of monocrotophos at rates of 1.12 and 2.24 kg AI/ha and carbaryl in molasses at 1.68 kg AI/ha effectively reduced southwestern corn borer (SWCB) infestations on tasseled corn. Plots treated with monocrotophos had significantly fewer larval infested, girdled, and lodged plants than plots that were treated with carbaryl or untreated. In 1977, larval densities were reduced by ground applications of diflubenzuron at 0.13 kg AI/ha and by carbaryl in molasses at 1.68 kg AI/ha. All chemically treated plots had fewer plants lodged than untreated plots.

INTRODUCTION

The southwestern corn borer (SWCB), *Diatraea grandiosella* (Dyar), is an internal stalk feeder that often severely damages corn in the southern U.S. The internal tunneling damage by 1st generation SWCB larvae to whorl stage corn and 2nd generation larvae to tasseled corn have been reported to directly reduce individual plant yield by 20% and 9%, respectively (Scott and Davis 1974). The extent of this plant damage to the entire crop production depends on SWCB infestation levels incurred at each growth stage. Throughout the Texas High Plains, observations reveal < 4% of the plant population may be infested with 1st brood larvae and 20% yield reduction to each infested plant would result in a relatively small per acre yield loss (< 1%). Therefore, the greatest potential for heavy yield losses is from heavier 2nd generation infestations and from the girdling activity by these larvae preparing to overwinter. Fall girdling activity by southwestern corn borer larvae weakens corn plants, causing plants to lodge and results in a yield reduction from corn ears that cannot be harvested by conventional machinery (Chada et al. 1965, Henderson and Douglas 1967).

Presently, insecticide applications provide adequate protection from 2nd generation SWCB. However, multiple applications of currently registered insecticides for SWCB control may enhance secondary outbreaks of spider mites infesting corn (unpublished data). Therefore, the present studies were conducted to determine and compare the effects of carbaryl, monocrotophos, and the insect growth regulator diflubenzuron.

METHODS AND MATERIALS

Test 1. Funk G-4503 field corn was planted April 23, 1976, on 1 m beds at a plant population of ca. 66,718/ha. Two applications of monocrotophos

^{1/}Lepidoptera: Pyralidae.

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and carbaryl were aerially applied to tasseled corn on August 12 and 19, 1976. Each treatment was made over an area 45 m wide X 61 m long.

Data were collected from 4 randomized plots (2 rows wide X 15 m long) within each treatment area. Whole-plant counts (10 plants/plot) were made to determine SWCB egg and larval infestations prior to and after application. Pre-treatment counts were made at 7 and 2 days prior to the 1st insecticide application and posttreatment counts at 6 (August 18) and 14 days (August 26) after the 1st application. On November 9, 25 plants/plot were monitored for lodging. Also, each plant was split to determine if SWCB larvae were present and to assess girdling damage. Corn ears were hand harvested from two, 4 linear row m/plot for subsequent yield determinations.

Test 2. Pioneer variety 3369A corn was planted on May 3, 1977. Plots (4 rows wide X 15 m long) were randomly arranged within 4 replicated blocks. Di-flubenzuron and carbaryl were applied July 21 and 28 to tasseled corn, with a hand-operated, CO₂-pressured sprayer calibrated to deliver 7.6 liters/ha of insecticide and water mixture.

The pre- and posttreatment SWCB egg counts (on 3 leaves above and 3 below the ear) were made on each of 25 plants in each treated and check plot July 19, 27, and August 3. Infestation levels, damage evaluation (girdling and lodging), and yields were determined from plants in two, 4 linear row m/plot on September 23.

An analysis of variance and Duncan's new multiple range test was used for each test to determine differences in chemical efficacy to SWCB and for yields in different plots.

RESULTS AND DISCUSSION

Test 1. Pretreatment counts showed SWCB egg numbers/plant and % of total plants infested were low and had not reached the action threshold (35% egg and/or small larvae) recommended by the Texas Agricultural Extension Service (Table 1). Even combining 7- and 2-day pretreatment counts, the highest plant

TABLE 1. Control of SWCB on Field Corn: Egg Numbers and % of Plants Infested with Eggs (I) Prior to and After Aerial Application of Insecticides, August 1976.

Chemical	Rate kg AI/ha	Day pretreatment ^{a/}		Days after 1st treatment					
		2		6		14			
		\bar{X} no. eggs	%I	\bar{X} no. eggs	%I	\bar{X} no. eggs	%I	\bar{X} no. eggs hatched	
Monocrotophos ^{b/}	1.12	0.33a	17.5a	0.63ab	17.5b	0.23b	12.5a	0.73a	
Monocrotophos ^{b/}	2.24	0.40a	15.0a	0.50b	22.5b	0.28b	15.0a	0.45a	
Carbaryl ^{b/}	1.68	0.23a	10.0a	0.38b	17.5b	0.43b	17.5a	0.10a	
Check	----	0.28a	12.5a	1.38a	47.5a	1.08a	30.0a	0.55a	

^{a/} Figures in a column followed by the same letter are not significantly different at the 5% significance level, Duncan's new multiple range test.

^{b/} Monocrotophos formulated as a water miscible solution containing 597 g AI/L, and carbaryl formulated as a flowable solution with molasses containing 478 g AI/L.

infestation was 27.5%. Six days after application, egg infestations had increased to 47.5% in the untreated area and all treated plots were significantly lower in egg numbers per plant and % plants infested. Posttreatment counts at

14 days showed no differences in % of plants infested with eggs, but egg numbers per plant were significantly reduced in insecticide treated plots.

All insecticide applications resulted in significant reductions in infested, girdled, and lodged plants when compared to untreated corn (Fig. 1).

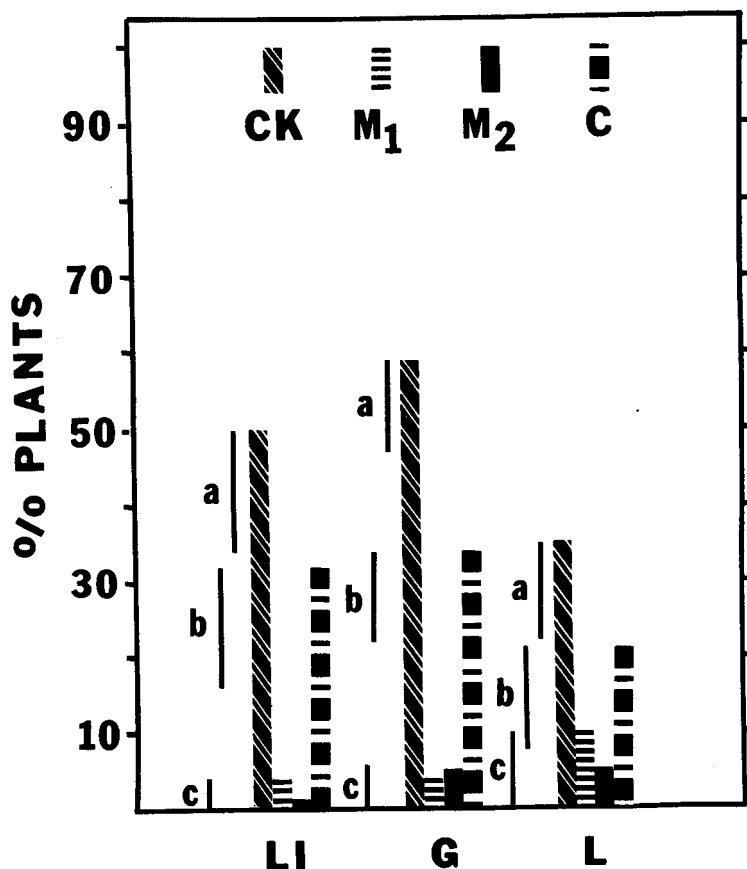


FIG.1. Percentage of plants larval infested (LI), girdled (G), and lodged (L) by SWCB in the untreated check (CK) and treatments with monocrotophos at 1.12 (M₁) and 2.24 kg AI/ha (M₂) and carbaryl at 1.68 kg AI/ha (C). Duncan's new multiple range test ($P=0.05$) is illustrated by vertical lines with small letters.

Girdling and lodging were lowest in plots treated with monocrotophos, thus demonstrating monocrotophos provided better control than carbaryl. Monocrotophos has the additional advantage of controlling mites as well as SWCB (Baranowski 1976, Daniels and Chedester 1977, and C. R. Ward, unpublished data). Yields obtained in the untreated check and treatments with monocrotophos at 1.12 and 2.24 kg AI/ha and carbaryl were statistically alike (7213, 6272, 6272, and 7150 kg/ha, respectively).

Test 2. Egg infestation levels in all plots were low (2%-7% infested plants) and appeared to remain static throughout the 2nd generation SWCB ovipositional period. Even after the insecticide applications no significant difference in either egg numbers/plant or percentage of plants infested with eggs occurred among treated and untreated plots.

SWCB densities in 1976 and 1977 never reached the action threshold for egg and small larvae infestations that are used in commercial practice. But, the subsequent heavy larval infestations and lodging in untreated plots at harvest indicate a need exists for 1) a more accurate threshold level before recommending chemicals and 2) better techniques for sampling SWCB eggs.

All treatments significantly reduced plant lodging (Fig. 2). Although

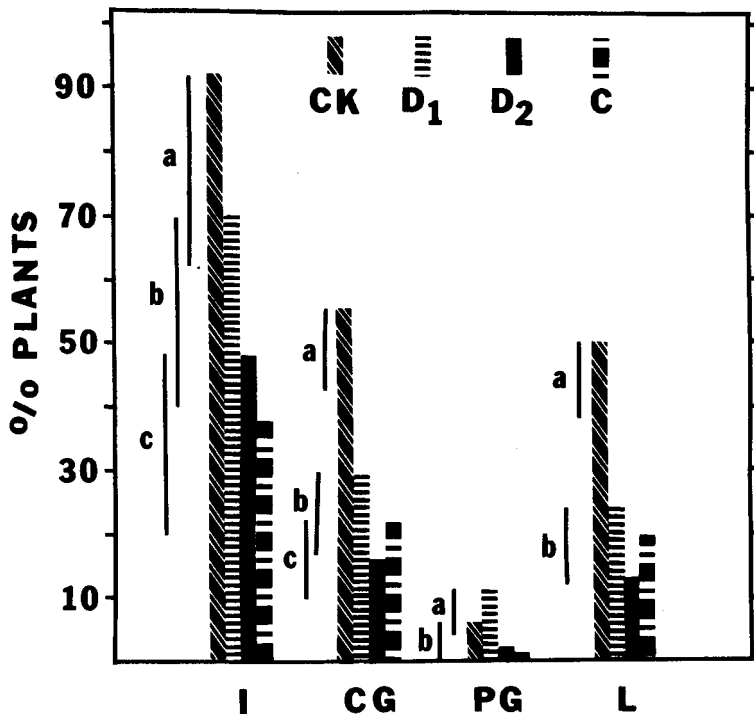


FIG. 2. Percentage of plants infested (I), completely girdled (CG), partially girdled (PG) and lodged (L) by SWCB in the untreated check (CK) and treatments with diflubenzuron at 0.07 (D₁) and 0.13 kg AI/ha (D₂) and carbaryl at 1.68 kg AI/ha (C). Duncan's new multiple range test ($P = 0.05$) is illustrated by vertical lines with small letters.

carbaryl treated plots were lowest in infested plants, the plots treated with the high rate of diflubenzuron were lowest in plants completely girdled and lodged. Diflubenzuron also has been shown not to be injurious to many important beneficial insects (Ables et al. 1977 and Wilkinson et al. 1978).

Although the number of infested plants was greatest in plots treated with diflubenzuron at 0.07 kg AI/ha, the number of plants completely girdled or

lodged was comparable to the other chemical treatments. This suggests that diflubenzuron may alter girdling activity of larvae preparing to enter diapause. Further studies need to be conducted on whether diflubenzuron concentrations consumed by larvae may directly or indirectly modify the diapausing hormone which regulates the SWCB larval activity to girdle plants. No statistical differences were noted for yields in the check and treatments of diflubenzuron at 0.07 and 0.13 kg AI/ha, and carbaryl (4641, 5143, 4453, and 4955 kg/ha, respectively).

Monocrotophos and diflubenzuron were effective in reducing SWCB larval infestations and/or inhibiting the prediapause girdling activity. The use of either monocrotophos or diflubenzuron for SWCB control may not readily cause secondary spider mite outbreaks. Significant differences were noted among larval infestations, but when corn ears from lodged and nonlodged plants were hand harvested no yield differences were obtained among treatments for either test. This suggests that tunneling damage from 2nd brood larvae may be nominal in reducing yields. Therefore, the greatest potential for yield loss would occur when corn ears could not be mechanically harvested from plants that lodged because of larval girdling.

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EFFECT OF DIFLUBENZURON FORMULATIONS ON THE EGG
PARASITE TRICHOGRAMMA PRETIOSUM^{1/2/3/}

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ABSTRACT

A mixture of diflubenzuron (25% WP, 70 g AI/ha), crop oil (Savol[®], 4.7 L/ha) and water applied to control the boll weevil, Anthonomus grandis Boheman, apparently had an adverse effect on the level of parasitism of Heliothis spp. eggs by Trichogramma pretiosum Riley. Laboratory studies subsequently demonstrated that application of diflubenzuron alone did not affect levels of parasitism; however, application of mixtures of crop oil and diflubenzuron or of crop oil alone significantly reduced the levels of parasitism of Heliothis spp. eggs by T. pretiosum.

INTRODUCTION

The insect growth regulator (IGR) diflubenzuron has been reported effective in controlling several phytophagous pests such as the gypsy moth, Lymantria dispar (L.), on apple (Granett et al. 1976); velvetbean caterpillar, Anticarsia gemmatilis Hübner, on soybean (Turnipseed et al. 1974); alfalfa weevil, Hypera postica (Gyllenhal), on alfalfa (Neal 1974); boll weevil, Anthonomus grandis Boheman, on cotton (Taft and Hopkins 1975); and citrus rust mite, Phyllocoptruta oleivora (Ashmead), on citrus (McCoy 1978). Diflubenzuron apparently has excellent potential in integrated pest management (IPM) programs directed against these and other pests because it effectively controls the target organism while causing relatively little harm to many species of parasites and predators (Ables et al. 1975, 1977; Granett and Weseloh 1975; Granett et al. 1976; Keever et al. 1977; Wilkinson et al. 1978).

Recently, interest has been renewed in use of the egg parasite Trichogramma pretiosum Riley to control Heliothis spp. in cotton (Ables et al. 1979, Ridgway et al. in press). Ables et al. (1977) demonstrated that this parasite and other entomophages were not adversely affected when exposed to diflubenzuron formulated with water. However, the results of Wilkinson et al. (1978) suggest that mixtures of diflubenzuron with emulsified paraffinic crop oils may cause significant mortality among some species of parasites and/or predators (Trichogramma was not tested). Since results of some of our field studies in 1978 suggested that T. pretiosum was adversely affected by treatments with a standard diflubenzuron-oil formulation, we initiated additional studies in the laboratory and greenhouse to determine the factors involved; these studies are reported here.

1/ Hymenoptera: Trichogrammatidae.

2/ In cooperation with the Texas Agricultural Experiment Station, Texas A&M University, College Station, TX 77843.

3/ This paper reports the results of research only. Mention of a pesticide in this paper does not constitute a recommendation for use by the USDA nor does it imply registration under FIFRA as amended. Also, mention of a commercial or proprietary product in this paper does not constitute an endorsement of this product by the USDA.

METHODS AND RESULTS

Field Tests. We conducted a field test in 1978 near Navasota, TX, in a 20-ha cotton field separated from the nearest commercial cotton by ca. 8 km. This test involved the use of microbial insecticides, including *Bacillus thuringiensis* (as Dipel®) and the *Heliothis* nuclear polyhedrosis virus (as Elcar®), in conjunction with *Trichogramma* releases to control *Heliothis* spp. In addition, diflubenzuron (25% WP, 70 g AI) was mixed with 4.7 L of crop oil (as Savol®) and water in a final volume of 46.7 (aerial) or 93.6 (ground) L/ha and applied 6 times at ca. 5-day intervals to control the boll weevil (Bull et al. 1979). During the early part of this test, we observed that diflubenzuron treatments seemed to cause a reduction in the levels of parasitism of naturally occurring *Heliothis* spp. eggs by *Trichogramma* (5-44% decrease within 2-3 days post application). Also, the percentage of parasitized eggs that produced adult parasites was consistently lower in treated areas than in the untreated control.

Following these observations, we divided a 10-ha portion of the field into 2 equal plots. One plot was treated with diflubenzuron with a high-clearance ground sprayer at the previously described rate (3 treatments), and 1 plot was left untreated. Four aerial releases of *Trichogramma* were made on both plots (avg. 110,000 parasites/ha) at 3- to 5-day intervals. We determined the extent of parasitism and subsequent adult emergence from parasitized eggs by collecting tan *Heliothis* eggs (ca. 1 day old) daily and holding them in the laboratory for observation. The dates of parasite releases and diflubenzuron-Savol treatments and parasitism of eggs in treated and untreated plots appear in Fig. 1. These data

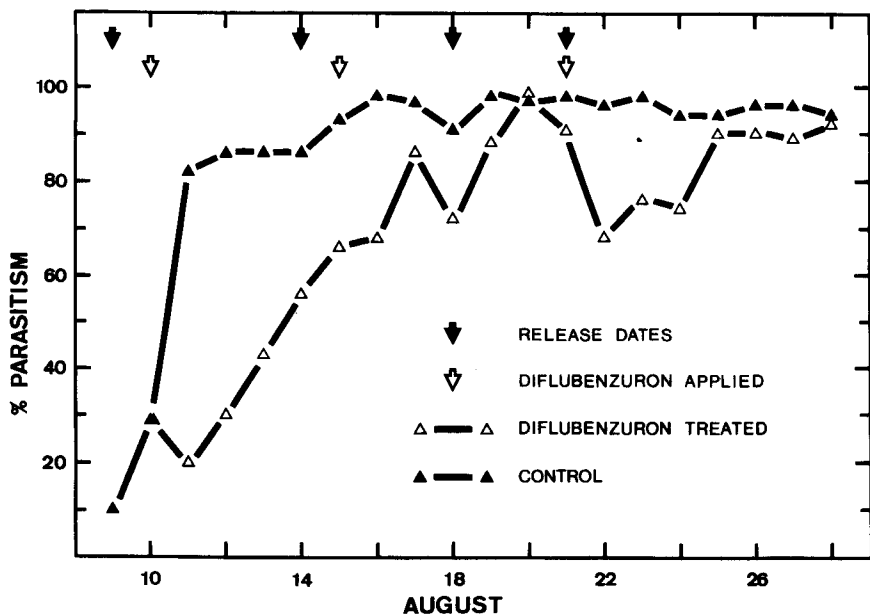


FIG. 1. Dates of *Trichogramma* releases and diflubenzuron-Savol treatments and mean percentage of parasitized *Heliothis* eggs in treated and untreated plots of cotton.

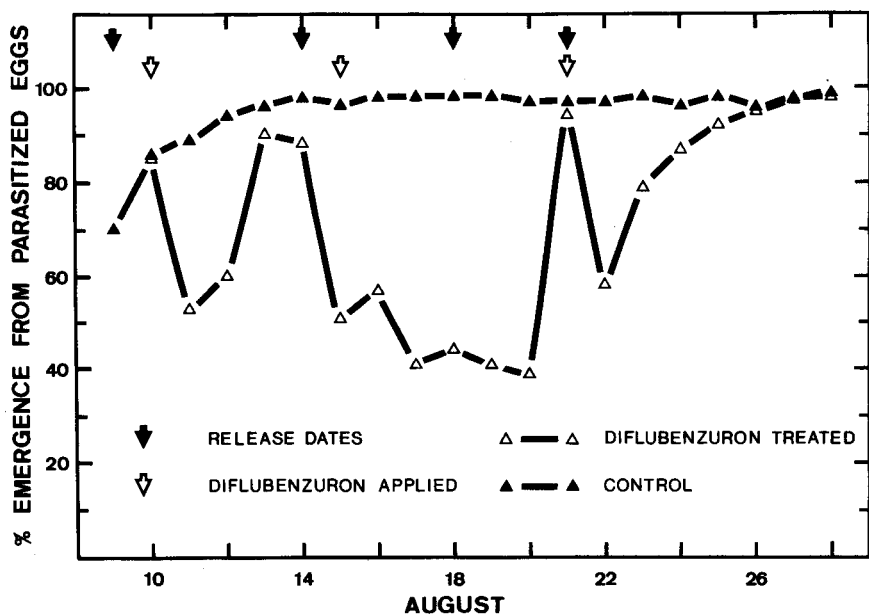


FIG. 2. Percentage of parasitized *Heliothis* eggs producing adult *Trichogramma* after collection from diflubenzuron-treated and untreated cotton plots.

strongly suggest that the diflubenzuron-Savol treatments reduced parasitism in the treated plot. *Heliothis* egg density was similar in both plots and averaged 69,000/ha during the test period (August 10-28). Results shown in Fig. 2 describe percentage of the same apparently parasitized eggs that actually produced adult parasites (parasitized eggs turn dark and show no larval development and are thus easily identified). These data revealed additional adverse effects on the parasite in that the number of parasitized eggs that produced adult parasites was consistently lower in the treated plot than in the untreated plot. Parasitism and emergence of adult parasites appeared to be similar among eggs collected from either treated or untreated plots after the diflubenzuron-Savol treatments were terminated.

Laboratory Tests. In laboratory tests of the effect of diflubenzuron on parasitism, eggs (6-24 h old) of the tobacco budworm, *Heliothis virescens* (F.), were obtained on oviposition substrates (paper toweling). Each egg group was then divided into 4 equal portions and sprayed with aqueous solutions of diflubenzuron alone, Savol alone, or with diflubenzuron + Savol. Diflubenzuron was applied at a rate equivalent to 70 g AI/ha and Savol at a rate equivalent to 4.7 L/ha; the final equivalent volume of aqueous mixtures sprayed was 93.6 L/ha. Treated eggs were placed in a standard petri dish (4 treatments/dish) and exposed for 15 min to adult parasites (collected from laboratory colony at 2 h postemergence). Parasites were removed and the eggs placed individually in compartmented holding cards (Hoffman et al. 1970) and held 10-12 days at $27 \pm 1.5^\circ\text{C}$ for observation.

Data presented in Table 1 indicate essentially no differences in the levels of parasitism of eggs that were untreated or treated with diflubenzuron alone. However, eggs treated with Savol alone or with Savol + diflubenzuron showed a significant ($P < 0.05$) reduction in levels of parasitism by *Trichogramma*. The pattern and

TABLE 1. Effect of Different Treatments on Parasitism of Tobacco Budworm Eggs by Trichogramma.

Treatment	Test 1 ^{a/}			Test 2 ^{b/}		
	No. eggs	% parasitism	% adult emergence	No. eggs	% parasitism	% adult emergence
H ₂ O untreated	629	60 a	95 a	685	72.7 a	95 a
DiFlubenzuron						
+ H ₂ O	696	51 a	89 a	732	75.9 a	98 a
Savol + H ₂ O	720	15 b	91 a	707	71.1 a	96 a
DiFlubenzuron						
+ Savol	552	19 b	92 a	640	72.1 a	93 a

^{a/}Test 1: eggs treated and then exposed to parasites; Test 2: eggs exposed to parasites and then treated.

^{b/}Data represent averages of 8 or more replicates; means followed by same letters are not significantly different ($P < 0.05$) "according to Duncan's multiple range test." Figures for % emergence adults are based on those eggs designated parasitized.

extent of parasitism in each test, although highly variable among replicates, was consistent. Parasitism was always substantially lower among eggs treated with Savol alone or in combination than in those untreated or treated with diflubenzuron alone. However, in contrast with results of field studies, we found no apparent differences among treatments in the percentage of parasitized eggs that produced adult parasites.

A 2nd series of laboratory tests was conducted to investigate the effect of the same treatments on previously parasitized Heliothis eggs. Eggs were parasitized as described and then treated and held under the same conditions. The results (Table 1), which indicated that none of the treatments adversely affected emergence of adult parasites, suggest that the effect of the oil is simply to inhibit oviposition by the parasite.

A 3rd series of laboratory tests was conducted to evaluate the effect of different oils of plant origin on the parasitism of tobacco budworm eggs by Trichogramma. Oils used were food grade and contained no preservatives. Treatments included an emulsifier used alone or in combination with corn oil, cottonseed oil, or soybean oil. An untreated check was included, as were treatments with Savol. Eggs were treated, parasitized, and held for observation as described. Data presented in Table 2 reaffirm results of previous tests, which indicate that oil adversely affected parasitism by Trichogramma, but again there was little or no effect on the percentage of parasitized eggs that subsequently produced adult parasites. Thus, the field and laboratory tests were in agreement on the apparent adverse effects of oils on parasitism by Trichogramma, but we were unable to demonstrate in the laboratory the reductions in percentage of parasitized eggs producing parasites.

DISCUSSION

As mentioned before, reports of the effects of diflubenzuron on parasitic Hymenoptera have been somewhat variable. For example, when host insects were treated with the chemical in a water formulation, some parasites of house fly, Musca domestica L., pupae (Ables et al. 1975) and T. pretiosum (Ables et al. 1977) were unaffected but Pediobius foveolatus (Crawford) and Apanteles melanoscelus (Ratzeburg) failed to complete development in their respective hosts, Epilachna varivestis Mulsant and Lymantria dispar (L.) (McWhorter and Shepard 1977, Granett and Weseloh 1975). Topical applications of aqueous suspensions of diflubenzuron

TABLE 2. Effect of Selected Oils on Parasitization of Tobacco Budworm Eggs and Subsequent Emergence of Adult Trichogramma.

Treatment ^{a/}	No. eggs	% parasitism	% adult emergence \pm SE ^{b/}
0.1% Triton X-100	846	39 \pm 32.4	97 \pm 2.5
Corn oil in 0.1% Triton X-100	294	2 \pm 1.4	80 \pm 15.0
Cottonseed oil in 0.1% Triton X-100	192	7 \pm 5.0	79 \pm 14.3
Soybean oil in 0.1% Triton X-100	246	0 \pm 0	--
Savol	707	1 \pm 2.8	100 \pm 0
Untreated check	786	45 \pm 31.1	97 \pm 2.4

^{a/}All applied at rate equivalent to 4.7 L oil plus 89.9 L H₂O/ha.

^{b/}Figures for adult parasite emergence are based on those eggs designated parasitized.

to those adult parasites studied thus far or to their substrates apparently have not been detrimental (Ables et al. 1977, Hassan 1977, Wilkinson et al. 1978). Although Wilkinson et al. (1978) reported that diflubenzuron formulated with oils + water had no effect on Apanteles marginiventris (Cresson), our results demonstrated that such formulations definitely had an adverse effect on T. pretiosum.

Thus, available evidence suggests that diflubenzuron is less detrimental to certain parasitic Hymenoptera and to other entomophages than conventional insecticides (Ables et al. 1975, 1977; Keever et al. 1977; Wilkinson et al. 1978). However, additional research is needed on different formulation and application techniques for this IGR to ensure optimum use of the chemical in IPM. Diflubenzuron is generally recommended for use with oil for maximum efficacy against the boll weevil. However, mites on citrus (McCoy 1978), gypsy moth on apples (Granett et al. 1976), and several pests of soybeans (Turnipseed et al. 1974) may be satisfactorily controlled with formulations of diflubenzuron in water. When possible, oils and/or emulsifiers should be reduced or eliminated not only for potential conservation of natural enemies but also for reduction of total treatment costs. Additionally, proper timing of the spray applications in relation to development of populations may be important to the survival of entomophages (Granett et al. 1976, Ables et al. 1977).

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CHITIN SYNTHESIS IN *HELIOTHIS ZEA* (BODDIE) PUPAE
AND INHIBITION BY CHITIN SYNTHESIS INHIBITORS^{1/}Heidi M. Kaska^{2/}, Richard T. Mayer^{3/}, Blair A. Sowa^{4/},
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ABSTRACT

Chitin synthesis in *Heliothis zea* (Boddie) was determined by measuring in vitro incorporation of ¹⁴C-labeled N-acetyl-D-glucosamine during pharate adult development. Two periods of chitin synthesis were observed, both associated with increases in total chitin content of the cuticle. The 1st period was of short duration, peaking at 3 h after larval-pupal ecdysis and lasting 1-2 h. The 2nd period of synthesis activity occurred from days 5 to 11 postecdysis, peaking at day 10. Two chitin synthesis inhibitors, diflubenzuron and Polyoxin D, were assayed in vitro for their effect on precursor incorporation on day 8. Addition of these compounds to the incubation media resulted in a 75% reduction in precursor incorporation.

INTRODUCTION

The present project was initiated to examine periods of chitin synthesis in the pupa and pharate adult of *Heliothis zea* (Boddie) as part of our effort at establishing physiological profiles of various developmental events in this insect. In the past, information regarding temporal patterns of chitin synthesis in insects was limited to estimates provided by measurements of dry weight of chitosan or cuticular thickness. These measurements, though helpful, provide an incomplete picture of chitin synthesis as they do not clearly differentiate between activity of chitinase and chitin formation. Similarly, assays by Marks and Sowa (1975) and Oberlander et al. (1973) show that chitin synthesis can be measured in vitro, but these assay systems preclude temporal studies as they involve addition of exogenous hormone. However, recent development of in vitro assays by Vardanis (1976) and Mayer et al. (1979, 1980), which utilize no exogenous hormone, facilitates examination of chitin synthesis activity during the course of an insect's development. A modification of these assays was used in our study to measure incorporation of labeled precursor in *H. zea* cuticle. The assay was also tested for its efficacy in screening compounds which might affect chitin synthesis rates.

^{1/}Mention of a proprietary product in this paper does not constitute an endorsement of this product by the U.S. Department of Agriculture.

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EXPERIMENTS

Experimental Insects. *Heliothis zea* larvae were reared in the laboratory on a synthetic diet at 25°C under a 12:12 light-dark cycle. Insects were weighed 1 h after larval-pupal ecdysis and thereafter held at 27°C (12:12) until they were used in specific experiments. Developing pharate adults were used at day 8 after larval-pupal ecdysis for all experiments except the temporal study, which required insects ranging in age from 0 to 11 days postecdysis.

Sources of Materials. Both the N-acetyl-D-glucosamine (1.7 mM; specific activity 58.18 mCi/mM; ^{14}C -labeled at the C-1 position of the D-glucosamine moiety) and the Bray's (dioxane-based) scintillation cocktail were purchased from New England Nuclear, Boston, MA. Chitinase, prepared from *Streptomyces griseus*, was purchased from Sigma Chemical Co., St. Louis, MO. The Polyoxin D was a gift of Dr. H. Saito, Kaken Chemical Co., Ltd., Tokyo, Japan and the diflubenzuron (DFB) was donated by Thompson-Hayward Chemical Co., Kansas City, KS. The 50 mM sodium phosphate buffers ranged in pH from 6.0 to 9.5 and contained a final concentration of 11 mM K^{+1} (from KCl) and 128 mM Na^{+1} (difference made up with NaCl). The acetate buffer (pH 5.5) contained 300 mM sodium acetate and 300 mM NaCl.

Test Procedures and Results. Chitin synthesis was determined by incubating insect cuticle with the ^{14}C -labeled chitin precursor, NAGA, according to Vardanis (1976) and Mayer et al. (1979, 1980). Abdominal halves were incubated in test tubes containing 1 ml phosphate buffer and various amounts of 200 mM NAGA and/or 1.7 mM ^{14}C -NAGA at 27°C for 4 h or more, after which the incubation media was discarded and 2 ml of 50% (w/w) KOH were added to stop the reaction. The incubated abdomens were then heated in the KOH for $1\frac{1}{2}$ h to digest nonchitinous materials and the resulting cuticle was recovered on a glass filter and rinsed with 100 ml each of distilled water, ethanol, and distilled water. Each filter bearing the cuticular residue was then placed in Bray's scintillation cocktail and counted by standard liquid scintillation techniques.

Six experiments were conducted in the course of this research: 3 studies to determine the effect of pH, precursor concentration, and incubation time on incorporation; a chitinase digestion study; a temporal study to determine periods of chitin synthesis in pupae of various ages; and a test of the inhibitory effects of 2 chitin synthesis inhibitors, DFB and Polyoxin D. Results for each of the experiments follow.

Effect of pH on Precursor Incorporation. Abdominal halves were incubated for 4 h in phosphate buffers containing 1 mM NAGA (sum of labeled and unlabeled precursors); pH values of different buffers were 6.0, 6.5, 7.0, 7.5, 8.0, 8.5, 9.0, and 9.5. Results of 3 replicate experiments revealed mean incorporations (\pm S.E.) of 0.088 ± 0.25 , 2.35 ± 0.89 , 3.32 ± 0.83 , 4.01 ± 0.91 , 3.85 ± 2.12 , 2.77 ± 0.52 , 2.15 ± 0.53 , and 0.62 ± 0.17 nmoles, respectively, indicating that a pH of 7.0-8.0 should be used for the incubation media.

Effect of Substrate Concentration on Precursor Incorporation. Abdominal halves were incubated in phosphate buffer, pH 7.0, for 4 h along with 6 different concentrations of NAGA, 3.4, 200, 400, 600, 800, and 1000 μM . Results of this experiment revealed mean incorporations of 0.032 ± 0.004 , 1.285 ± 0.257 , 2.571 ± 0.677 , 3.117 ± 0.812 , 4.158 ± 0.777 , and 2.961 ± 0.473 nmoles, respectively, indicating maximum precursor incorporation at 800 μM NAGA.

Effect of Incubation Time on Precursor Incorporation. The effect of incubation time on NAGA incorporation was determined. Abdominal halves were incubated in phosphate buffer (pH 7.0) with a total NAGA concentration of 1 mM at 27°C for 4, 8, and 24 h. The study revealed that mean

incorporation at 4, 8, and 24 h was 2.77 ± 0.76 , 2.04 ± 0.59 , and 2.54 ± 0.47 nmoles, respectively, indicating that little additional incorporation occurred after 4 h.

Chitinase Digestion of Samples. To confirm that the NAGA precursor was being incorporated into chitin rather than some other material, prepared abdomens were halved and $\frac{1}{2}$ was subjected to chitinase digestion. After incubation with NAGA and hot KOH treatment, cuticle was placed in test tubes with 5 mg chitinase in 1 ml 0.3 M acetate buffer and incubated at 27°C for 48 h (Mayer et al. 1979, 1980). The cuticle was then rinsed, refiltered, and placed in vials containing scintillation fluid for counting. Results of this study showed that when the chitinase-treated abdominal halves were compared to the control halves, an average of $77 \pm 8\%$ of the labeled material was released from the cuticle.

Incorporation Rates During Pharate Adult Development. Results of the NAGA incorporation study during pharate adult development are shown in Fig. 1. Each point represents the mean of 10 individual pupal abdomens. When NAGA was used as a chitin precursor, 2 peaks of synthesis activity were seen: one during the 1st 24 h following larval-pupal ecdysis and one during pharate adult development at 10 days postecdysis. Refinement of the 1st 6-h interval was made by examining incorporation rates at 0, 1, 2, 3, 4, and 6 h post larval-pupal ecdysis. Results of these studies showed that peak incorporation during this period occurred at 3 h postecdysis. Activity at this 3-h point was as intense as at the 10-day point in terms of nmoles NAGA incorporated. However, the initial activity was relatively short-lived, compared with that which occurred later in adult development.

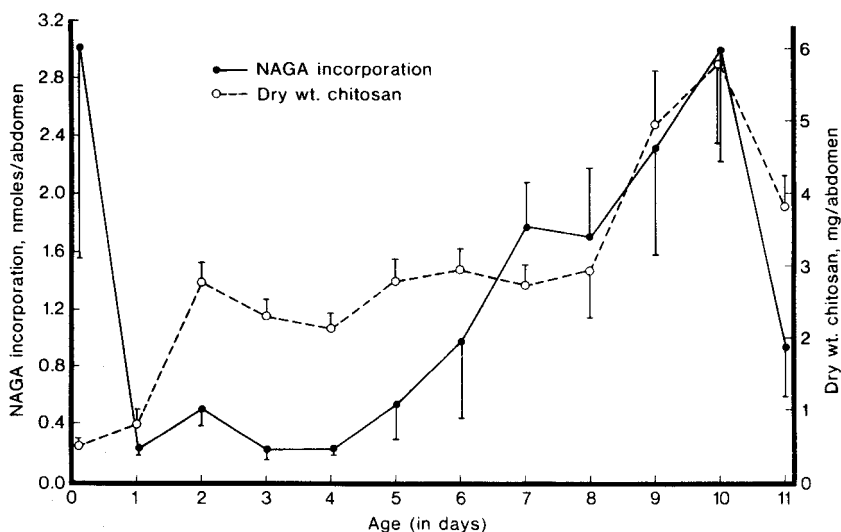


FIG. 1. *In vitro* incorporation of ^{14}C -NAGA into chitin when pupal abdomens were incubated at intervals after larval-pupal ecdysis with 1 mM NAGA for 4 h. Gravimetric measurements of treated cuticle made following NAGA incorporation and hot KOH digestion are also shown.

Gravimetric measurements of cuticle from each of the individuals used in the temporal incorporation study were made prior to placement in scintillation vials and are also shown in Fig. 1. These measurements reveal that an increase in dry weight of the KOH-treated cuticle (chitosan) occurred at 2 periods within the pupal stage: the 1st during the 1st 2 days postecdysis and the 2nd from 8 to 10 days after larval-pupal ecdysis. These dry weight results correlate with the incorporation data which indicate substantial chitin synthesis activity at about these same times.

Diflubenzuron and Polyoxin D Inhibition of Chitin Synthesis. Polyoxin D and DFB were tested at 2 concentrations to determine the efficiency of the assay in screening substances which affect chitin synthesis. Substrate and inhibitors were added to the incubation media containing ^{14}C -labeled NAGA immediately preceding addition of the abdominal halves. The mean percentage inhibitions of NAGA incorporation in DFB-treated samples were $57 \pm 7\%$ at the $0.02 \mu\text{g/ml}$ concentration and $82 \pm 7\%$ at the $0.2 \mu\text{g/ml}$ concentration. When Polyoxin D was tested at a concentration of $10 \mu\text{g/ml}$, inhibition averaged $79 \pm 5\%$.

DISCUSSION

The temporal study of chitin synthesis during pharate adult development revealed 2 peaks of activity: one at 3 h after larval-pupal ecdysis, before sclerotization of the exocuticle was complete, and a 2nd at 10 days postecdysis, 2 days before eclosion. The 2nd period of chitin synthesis activity began at 5 days postecdysis, $\frac{1}{2}$ day after the 20-hydroxy-ecdysone peak reported by Holman and Meola (1978), and is, therefore, in accord with the classical concept of chitin synthesis activity being initiated by and/or associated with ecdysone production (Locke 1964, Willis 1974, Ryerse and Locke 1978, Nardi and Willis 1979).

The 1st peak of chitin activity at 3 h post larval-pupal ecdysis was followed over a 2-day period by a small increase in total dry weight of chitin. This peak probably represents postecdysial endocuticle formation similar to that reported in other insects (Zacharuk 1976). The 2nd period of chitin synthesis activity occurred between 8 and 11 days postecdysis and was also associated with an increase in dry weight of chitinous material in the cuticle.

Results of the temporal study, in addition to supplying information about the nature of chitin synthesis in *H. zea*, provided data necessary for the assay of chitin synthesis inhibitors. Under the conditions defined in this study, at a temperature of 27°C and a total NAGA concentration of 1 mM , optimum incorporation of precursor occurred when cuticle was incubated at pH 7.5 for 4 h.

In order to evaluate the usefulness of this assay system for testing the effect of various compounds on chitin synthesis, 2 known chitin synthesis inhibitors, DFB and Polyoxin D, were added to the initial incubation media before incubation commenced. Since the temporal study revealed that the main period of chitin synthesis occurs between days 8 and 11 postecdysis, cuticle used for these studies was obtained from pharate adults on day 8 postecdysis.

The results of these tests showed an inhibition of synthesis for each compound (at the highest levels tested) of over 75%. Polyoxin D inhibition in our system is comparable to that of the milkweed bug (Hajjar and Casida 1979) and stable fly (Mayer et al. 1980) systems but is less effective than that reported in the cockroach leg regenerate system. The values obtained in our system for DFB inhibition of chitin synthesis agree with those obtained in cockroach leg regenerates (Sowa and Marks 1975) and stable fly

abdomens (Mayer et al. 1980), showing comparable inhibition at DFB concentrations 1-2 orders magnitude lower than reported for the milkweed bug system (Hajjar and Casida 1979). Thus, despite reports that DFB was ineffective in controlling *Heliothis* spp. (Wolfenbarger et al. 1977, Sowa and Meola, unpublished data), we conclude that *H. zea* pupal chitin synthesis shows no unusual insensitivity when the compound is delivered to intact cuticle-forming tissues in vitro.

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FEED-THROUGH EFFICACY OF CGA-19255 AND CGA-72662 AGAINST
MANURE-BREEDING FLIES^{1/} AND OTHER ARTHROPODS AND RESIDUES
IN FECES, EGGS, AND TISSUES OF LAYING HENS^{2/}

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ABSTRACT

CGA-72662 (N-cyclopropyl-1,3,5-triazine-2,4,6-triamine) was found to be ca. 2X more effective than CGA-19255 (6-azido-N-cyclopropyl-N'-ethyl-1,3,5-triazine-2,4-diamine) as a poultry feed-through to inhibit development of immature forms of the house fly, *Musca domestica* L., and the little house fly, *Fannia canicularis* (L.). Levels of 1.25 and 5.0 ppm of CGA-72662 gave >99% total mortality of the house fly and little house fly, respectively. CGA-19255 is metabolized to CGA-72662 in chickens, and residues of CGA-72662 were found in eggs, liver, and muscle of chickens fed either compound.

INTRODUCTION

It has previously been reported that the insect growth regulator, Ciba-Geigy CGA-19255 (6-azido-N-cyclopropyl-N'-ethyl-1,3,5-triazine-2,4-diamine) fed to hens at a level from 2.5 to 5 ppm prevented development of larvae of the house fly, *Musca domestica* L., in manure (Breedon and Turner 1977, Christensen and Knapp 1976, Miller et al. 1977). Miller et al. (1977) showed that this compound also inhibited development of larvae of the little house fly, *Fannia canicularis* (L.), but 10 ppm was needed for complete inhibition. They further reported that when the compound was fed to chickens at levels as high as 40 ppm in the ration, no CGA-19255 was found in the eggs. We report here results of further tests of CGA-19255 and tests of CGA-72662 (N-cyclopropyl-1,3,5-triazine-2,4,6-triamine).

METHODS AND MATERIALS

In an initial experiment, CGA-19255 and CGA-72662 were each mixed into a commercial-type poultry ration at levels of 0, 1.25, 2.5, and 5.0 ppm. Rations containing each of these levels were fed to 8 mature White Leghorn hens for 21 days. Eight times during the feeding period, manure droppings were collected from under the treated and control hens and bioassayed with house fly and little house fly larvae as previously described (Miller et al. 1970).

1/ Diptera: Muscidae.

2/ This paper reports the results of research only. Mention of a pesticide does not constitute a recommendation for use by the USDA nor does it apply registration under FIFRA as amended. Also, mention of a proprietary product does not constitute an endorsement of this product by the USDA.

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In a 2nd experiment, which started June 15, 1978 and continued until September 27, 1978, 12 White Leghorn hens were placed in a wire battery suspended above the floor in a small poultry house and fed a ration containing 1.5 ppm of CGA-72662. Hens housed in a separate building and fed untreated ration served as a control. Manure was allowed to accumulate under the cages. Twice a week, manure from both groups of hens was sampled and put into paper souffle cups, which were placed over sand in plastic containers covered with nylon stocking material and held for 3 wk. At this time, the containers were opened and adult insects were put into vials for later counting and identification.

In a 3rd experiment, 4 hens were fed CGA-19255 or CGA-72662 at 1 of 4 dosages (2.5, 12.5, 25, and 125 ppm), plus a control treatment, for 5 wk. During wk 3, 4, and 5 of the experiment, manure samples were collected from each group of hens and analyzed for CGA-19255 and CGA-72662. At the end of 5 wk, all hens were sacrificed by cervical dislocation and samples of fat, liver, and breast muscle were taken, frozen at -20°C, and later analyzed for both compounds.

The extraction procedures for CGA-19255 and CGA-72662 were the same as those previously described (Miller et al. 1977); however, the acetonitrile extracts were chromatographed on 5 g of silica gel (3405 Baker analyzed). The CGA-19255 was eluted from the silica gel column with 150 ml of 10% acetone in methylene chloride and the CGA-72662 with 125 ml of methanol. The eluants from the silica gel column were evaporated on a rotary evaporator and the final volumes adjusted to 20 ml with methanol and submitted to high pressure liquid chromatography (HPLC) for detection of residue.

The HPLC was operated with the following parameters: instrument-laboratory-assembled, Constametric II G Pump (Laboratory Data Control, Riviera Beach, FL), Schoeffel SF-770 Spectroflow Monitor (Schoeffel Instrument Corporation, Westwood, NJ); Zorbax-CN (E. I. du Pont de Nemours & Co., Inc., Wilmington, DE), 25-cm stainless steel; flow rate - 0.6 ml/min; mobile phase - 50% methanol, 50% water; detector - UV photometer at 220 nm; sample size - 20 µl methanol extract; temperature - ambient. Under these conditions, the method can detect and measure 0.02, 0.04, and 0.1 ppm of CGA-19255 or CGA-72662 in eggs, tissues, and feces, respectively.

Mortality and residue data were analyzed with analysis of variance and differences in means were tested for by Duncan's new multiple range test.

RESULTS AND DISCUSSION

CGA-72662 was more effective against the house fly than CGA-19255 by killing larvae directly or by preventing eclosion of adults from pupae (i.e., >99% total mortality at 1.25 ppm of CGA-72662 and at 2.5 ppm of CGA-19255) (Table 1). As we previously reported (Miller et al. 1977), the little house fly is less susceptible than the house fly to these compounds; however, >99% mortality was obtained at the 5-ppm feeding level of CGA-72662.

TABLE 1. Mortality^{a/} of House Flies and Little House Flies Seeded as Larvae into Manure from Chickens Fed CGA-19255 and CGA-72662 (Experiment 1).

Compound	Level in feed (ppm)	House fly		Little house fly	
		% Larval mortality	% Total mortality	% Larval mortality	% Total mortality
CGA-19255	1.25	8 a ^{b/}	78 a	2 ab	1 a
	2.5	76 b	>99 b	1 a	2 a
	5.0	>99 c	100 b	6 ab	26 b
CGA-72662	1.25	95 c	>99 b	2 ab	4 a
	2.5	100 c	100 b	15 b	62 c
	5.0	100 c	100 b	80 c	>99 d

a/ Corrected for control mortality with Abbott's formula.

b/ Means in a column not followed by a common letter are significantly different at the 5% level according to Duncan's new multiple range test.

TABLE 2. Numbers of Insects Emerging from Manure^{a/} of Control Hens and from Those Fed 1.5 ppm of CGA-72662 During Summer of 1978 (Experiment 2).

	June		July		August		September	
	Control	Treated	Control	Treated	Control	Treated	Control	Treated
Coleoptera								
Hydrophilidae	0	0	0	0	9	0	20	0
Staphylinidae	2	0	3	0	2	0	0	0
Histeridae	0	1	0	3	1	1	0	0
Diptera								
Calliphoridae	0	0	0	0	1	0	0	0
Muscidae	972	0	213	0	100	0	13	0
Drosophilidae	0	0	0	1	19	8	11	0
Sphaeroceridae	412 ^{b/}	0	501	0	89	0	1	0
and Milichidae	0	0	21	0	2	0	0	0
Sepsidae								
Hymenoptera								
Braconidae	34	0	42	1	22	0	0	0

a/ Total number of insects emerging from 4 samples in June, 8 in July and 9 in August and in September.

b/ Approximately 17% of totals of the 2 families were Sphaeroceridae.

Few insects emerged from the manure of hens fed CGA-72662 (Table 2). These insects included not only Dipteran species but also species of Coleoptera and Hymenoptera that may be parasitic or predatory on fly larvae or pupae. We did not find any manure-inhabiting mites such as *Macrocheles muscadomesticae* (Scopoli) or *Fuscuropoda vegetans* (De Geer) in manure from either control or treated hens. Axtell (1970) reported that these mite species are found in poultry manure and are predaceous on immature stages of the house fly. He showed that larviciding with organophosphorus insecticides destroyed these mite populations. Since, under some circumstances, certain insects and mites are beneficial in holding down fly populations in poultry houses, further work on the effect of CGA-72662 on beneficial species needs to be conducted.

TABLE 3. Residues of CGA-72662 in Eggs, Tissues, and Feces of Chickens Fed CGA-19255 and CGA-72662 in Their Rations at Indicated Levels^{a, b/} (Experiment 3).

Level in feed(ppm)	Compound	Eggs	Liver	Muscle	Feces
<hr/> μg/g wet weight <hr/>					
2.5	CGA-19255	0.02 a ^{c/}	0.03 a	0.03 a	0.20 a
	CGA-72662	0.10 a	0.00 b	0.00 b	0.27 a
12.5	CGA-19255	0.06 A	0.10 a	0.08 a	0.90 a
	CGA-72662	0.20 B	0.10 a	0.10 a	1.33 a
25	CGA-19255	0.13 A	0.12 a	0.10 A	2.17 a
	CGA-72662	0.28 B	0.18 a	0.20 B	3.17 a
125	CGA-19255	0.50 A	0.75 a	0.50 A	11.67 a
	CGA-72662	2.45 B	0.77 a	0.90 B	23.33 a

a/ Lower limit of detection 0.02, 0.04, and 0.1 μg/g for eggs, tissues, and feces, respectively. Values in table lower than these result from 0 amounts in average.

b/ Within a criteria and level, means not followed by a common letter are significantly different (capital letter $P < 0.01$, lower case letter $P < 0.05$) according to Duncan's new multiple range test.

c/ Residues in eggs at 2.5 μg/g level were significantly different at $P < 0.10$.

Table 3 shows the residues of CGA-72662 in the eggs, liver, muscle, and feces of hens fed CGA-19255 and CGA-72662 at 4 different levels. Hens fed CGA-19255 had no residues of the parent compound in any of the tissues or feces analyzed, which confirms our earlier work with this compound (Miller et al. 1977). Hens fed either compound had significant residues of CGA-72662 in the eggs; however, at each feeding level, hens fed CGA-72662 had greater residues than hens fed CGA-19255. Residue levels in eggs of hens fed 2.5 ppm of CGA-72662 were, however, only 0.10 ppm, and this level of feeding is 2X more than that shown necessary to give >99% total mortality of house flies.

CGA-72662 does not appear to be fat-soluble since none (0.04 ppm) was detected in the body fat of hens fed any level of either compound. Levels detected in the liver were similar for both compounds except at the lowest feeding level, at which residues from hens fed CGA-72662 were below detection level (<0.04 ppm).

The amount of residue in the breast muscle was similar to that found in eggs except that no (<0.04 ppm) CGA-72662 was found in the muscle of hens fed this compound at the lowest level tested.

The amounts of CGA-72662 found in the feces of hens fed CGA-72662 were 1.4-2X higher than those from hens fed CGA-19255, but because of large day-to-day variation, these differences were not significant.

Our results indicate that CGA-72662 is highly effective against immature stages of the house fly and the little house fly when administered to hens in the ration. The previously reported effectiveness of CGA-19255 (Miller et al. 1977) appears to be through the action of CGA-72662. It would appear that a major proportion of CGA-19255 is converted to CGA-72662 in the digestive tract of the hen since only CGA-72662 is found in the feces. The converted CGA-72662 is then probably absorbed into the body and metabolized by the hens as if they had been fed CGA-72662 directly.

An area that should be further investigated is the possible deleterious effects of these compounds on nontarget and possibly beneficial insect and mite fauna in the droppings of hens fed the compounds.

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THE ONTOGENY OF BLISTER BEETLES (COLEOPTERA, MELOIDAE)
IV.- PYROTA INSULATA LeCONTE

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ABSTRACT

The larval stages of Pyrota insulata LeConte develop at a faster rate than other species of the genus. Coarctate larvae break diapause in late summer and the adults remain in the pupal cells until early spring, when they emerge in synchronization with the bloom of mesquite trees. Larvae had the ability to revert to the diapausing coarctate phase up to 4 times, indicating a strong adaptation to extremes of drought and temperature.

RESUMEN

El desarrollo de los estados larvales de Pyrota insulata LeConte es relativamente rápido comparado con otras especies del género. Las larvas coarctadas rompen la diapausa a finales del verano y los adultos permanecen en sus celdas pupales hasta comienzos de la primavera, cuando emergen en sincronización con la floración de los árboles de mesquite. Las larvas tienen la habilidad de regresar al estado diapáusico de la fase coarctada hasta 4 veces en su vida mostrando una gran adaptación a temperaturas y sequías extremas.

INTRODUCTION

This paper is the 4th in a series of papers dealing with the postembryonic ontogeny of blister beetles. Selander and Mathieu (1964) reared 3 species of the genus Pyrota, using provisions and larvae of the honey bee. The same authors (1969) reared 7 species of Epicauta as part of a study on the ecology and behavior of these beetles. Selander and Weddle (1969, 1972) manipulated the age of triungulins and temperature during larval phases of Epicauta segmenta (say), and their study provided a sound base to explain the adaptive patterning of hypermetamorphosis in blister beetles.

This report describes the ontogeny of Pyrota insulata LeConte from the time of oviposition to the emergence of the last adult.

MATERIALS AND METHODS

Large numbers of P. insulata were collected as they responded to light at Apodaca, Nuevo León, México on the nights of March 10 and 20, 1967. They were also collected from mesquite during the day on March 17, 1967.

Adults were kept in plastic cages with 4 cm of fine sand on the bottom and were fed daily with tender leaves and blooms of mesquite. Females began oviposition by the usual way of digging a tunnel in the soil. Eggs were removed daily, and records were made of the time of hatching and survival of unfed triungulins.

The insects were reared with the technique described by Selander and Mathieu (1964), except that bee larvae were not added to the diet. Twenty triungulins from the same egg mass were used to initiate each study. Terminology used is the same suggested by Selander and Mathieu (1964): T₁, triungulin phase or 1st larval instar; FG, 1st grub phase (instars 2-5); C, coarctate phase (instar 6); SG, 2nd grub phase (instar 7); P, pupal stage (instar 8); A, adult stage (instar 9). Glass tubes (2.5 cm in length) with cotton stoppers on both ends

were used for feeding phases T₁ and FG; new pollen and water were given daily to all larvae. When any mold developed on the food, larvae were transferred to new vials. The experiment was done at room temperature (22-25°C), following the natural climatic fluctuation of Monterrey, Nuevo León.

RESULTS AND DISCUSSION

Egg and 1st Instar Larval Stage. The average duration from oviposition to eclosion in *P. insulata* was 10.24 ± 1.13 days (SD) (N=50) at $24^\circ\text{C} \pm 1^\circ\text{C}$. This incubation period is comparable to the range reported by Selander and Mathieu (1964). The mean number of eggs/mass oviposited by females of this species was 884 ± 261.4 (SD) (N=14). Among triungulins maintained without food in vials with moist cotton stoppers, 50% mortality was reached 6 days after eclosion and total mortality after 11 or 12 days. Females of this species lay as many as 6 egg masses, potentially ovipositing over 5000 eggs in their life time. Thus, this species has one of the largest biotic potentials among winged species of Meloidae with free walking triungulins. Because 1st instar larvae must find their host bee nest within a few days of eclosion, mortality must be very high during the dispersal phase of development.

First Grub to Adult Stages. Results of studies of the duration of the 1st part of the ontogeny from T₁ to FG₅ are presented in Table 1.

TABLE 1. Duration (in Days) of the T₁ and FG Phases of *P. insulata*.

Instar	Mean	SD	Range	N
T ₁	5.28	2.67	2-9	14
FG ₂	2.57	0.65	2-4	14
FG ₃	2.21	0.80	1-4	14
FG ₄	2.43	0.94	1-4	14
FG ₅	$17.42^{1/}$			

^{1/} The FG₅ larvae were fed 10 days and then transferred to a vial that contained only soil.

TABLE 2. Duration in Days of the Feeding Grub Stages of 3 Species of *Pyrota*^{1/}

Species	\bar{x}	Range	SD	N
FC ₂ to FC ₄				
(a)				
<i>P. insulata</i>	7.21	6-9	1.01	14
<i>P. nigrovittata</i>	8.30	6-13	1.60	22
<i>P. palpalis</i>	8.20	4-14	1.80	33
T ₁ to FC ₅				
(b)				
<i>P. insulata</i>	29.33	26-34	2.05	12
<i>P. nigrovittata</i>	38.40	31-42	--	8
<i>P. palpalis</i>	31.70	24-42	--	18

^{1/} Data shown for *P. nigrovittata* and *P. palpalis* are from Selander and Mathieu (1964).

TABLE 3. Duration (in Days) of the Larval Phases of P. insulata After the Feeding Period.

Larval phases	Short Diapause (N=5)			Long Diapause (N=5)			Combined Diapause (N=10)		
	Mean	Range	SD	Mean	Range	SD	Mean	Range	SD
C ₆	52.40	41-58	6.62	412.40	382-437	20.04	--	--	--
SG ₇	13.60	9-18	3.36	14.00	7-20	5.83	13.80	7-20	4.49
P ₈	15.20	10-18	3.90	17.60	9-23	5.18	16.40	9-23	4.50
A	122.80	54-188	62.30	109.20	59-160	39.48	116.00	54-188	49.69

The triungulin, after feeding begins, is the most variable stage (CV=50.6%) and the FG₅ stage is the least variable (CV=8.7%).

In comparing the data on the duration of the feeding-grub phase of P. insulata with that of P. nigrovittata (Haag-Rutenberg) and P. palpalis (Champion) reported by Selander and Mathieu (1964), it is evident that there is a difference in the time of development from the 2nd to 4th instars. (Table 2a).

Pyrota insulata developed faster, even though larvae were reared at a lower temperature and without bee larvae. The duration of the entire larval phase (Table 2b) appeared to be shorter for P. insulata, although no statistical comparisons were made.

The majority of the larvae (N=10) followed the usual ontogenetic pattern for meloids; that is, T₁ - FG₂₋₅ - C₆ - SG₇ - P₈ - A. Studies of the duration of the postfeeding period of P. insulata indicated that a group of 5 coarctates broke diapause when moisture was added to the vials where larval cells had been formed (Table 3). Coarctates were kept in glass tubes, after being removed from their earthen cells, to facilitate future observations. In July 1967, these coarctates molted to the SG₇ stage, and then pupated and emerged as adults. Several adults lived up to 6 months in a lethargic state without feeding. This suggests that individuals of this species develop to the adult stage in earthen cells in the soil, or in bee nests in the fall, and then spend the winter in a lethargic state and emerge in early spring when mesquite sprouts and blooms. Selander and Weddle (1972) found that thermal stimulation terminated diapause in coarctate larvae of E. segmenta; 15°C was the temperature causing 100% termination of diapause after 60 to 90 days exposure.

Another group of 5 larvae had a longer period of diapause, going into a 2nd yr as coarctates. After diapause was terminated, the fate of these individuals was very similar to that observed for the 1st group (Table 3). Two larvae from the latter group of coarctates had a greatly prolonged period of diapause (1111 and 1149 days) but then completed the SG₇, P₈ and adult stages within the normal range of duration.

One male larva departed from the normal pattern by leaving and reverting to the diapause stage 4 times (Table 4). The period of time from the day the triungulin began feeding to the day the adult emerged covered ca. 4 1/2 yr. When this individual developed to the adult stage, it fed well and lived normally for 47 days; this adult was small but no smaller than some others observed in nature. A similar reversion by E. segmenta larvae to the coarctate phase was reported by Selander and Weddle (1972). Thus, this behavior may be common to Meloidae.

The geographical distribution pattern of Meloidae reveals that many of the species have been successful in the arid lands of the world. Hypermetamorphosis is certainly an extraordinary adaptation for survival under prolonged climatic stress. Synchronization of the larval and adult hosts is achieved through this ontogenetic plasticity, where triungulins, coarctates, and adults play the major adaptative roles.

TABLE 4. Extended Ontogenetic Pattern Shown by 1 Individual of P. insulata.

<u>Phase</u>	<u>Days duration</u>	<u>Days accumulated</u>
T ₁ -C ₆ -----	412 -----	412 -----
SG ₇ -----	8 -----	420 -----
C ₈ -----	428 -----	848 -----
SG ₉ -----	10 -----	858 -----
C ₁₀ -----	406 -----	1264 -----
SG ₁₁ -----	19 -----	1283 -----
C ₁₂ -----	412 -----	1695 -----
SG ₁₃ -----	12 -----	1707 -----
P ₁₄ -----	19 -----	1726 -----

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ACTIVITY OF THREE CYCLOHEXADIENE ANALOGUES
AGAINST THE BOLL WEEVIL^{1/} AND TOBACCO BUDWORM^{2/3/4/}

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ABSTRACT

Three analogues of cyclohexadiene (I) 2,6-dibromo-4-hydroxy-4-methyl-2,5-cyclohexadiene-1-one, (II) 2,6-dibromo-4,4-dimethyl-2,5-cyclohexadiene-1-one, and (III) 2,6-dibromo-4-hydroxy-4-methyl-2,5-cyclohexadiene-1-one acetate were tested for activity against the boll weevil, Anthonomus grandis Boheman, and the tobacco budworm, Heliothis virescens (F.). Only compound III was effective against the boll weevil, but all 3 compounds showed activity against the tobacco budworm. Spray applications of compound III on cotton in the field apparently caused some reduction in the development of boll weevils in cotton squares and, when incorporated into artificial larval diet, the chemical reduced adult emergence ca. 90% at concentrations as low as 0.001% (w/v). Applications of compound III as a foliar spray to potted cotton plants completely inhibited the hatch of tobacco budworm eggs. Topical applications of the chemical were also effective against 1st stage tobacco budworms. In addition, topical applications of compound I significantly affected larval development, pupation, and adult eclosion of this insect in the laboratory.

INTRODUCTION

Insect growth regulators (IGR's) have great potential for use in the selective control of different pests. An IGR that could be used effectively in the selective control of the boll weevil, Anthonomus grandis Boheman, and Heliothis spp. on cotton could represent an invaluable tool for use in pest management programs that seek to minimize current dependence on conventional insecticides. In laboratory studies, Guerra et al. (1973) demonstrated that certain juvenile hormone (JH) analogues were active against the tobacco budworm, Heliothis virescens (F.), but concluded that none was effective enough for practical use. Guerra (1975) also conducted laboratory tests of combinations of 5 oral chemo-sterilants and gamma irradiation and found that none of the chemicals alone significantly affected development of tobacco budworms. Wolfenbarger et al. (1977) demonstrated that the IGR diflubenzuron was active against the tobacco budworm in the laboratory when it was incorporated into artificial diets; however, diflubenzuron is generally considered to be ineffective against Heliothis spp. in the field at the recommended use rates. Since the boll weevil and Heliothis spp. are perhaps the most destructive pests of cotton, and there is an urgent need to reduce the current heavy use of broad spectrum insecticides for their control, it is important that we develop effective alternative biocontrol

^{1/}Coleoptera: Curculionidae.^{2/}Lepidoptera: Noctuidae.^{3/}In cooperation with the Texas Agricultural Experimental Station, Texas A&M University, College Station, TX 77843.^{4/}This paper reports the results of research only. Mention of a pesticide does not constitute a recommendation for use by the USDA nor does it imply registration under FIFRA as amended.

techniques for managing codistributed populations of these pests. Herein, we report the results of laboratory and field cage tests conducted at Brownsville, TX to evaluate the biological activity of 3 experimental analogues of cyclohexadiene (generally classed as IGR's) against the boll weevil and tobacco budworm.

METHODS AND MATERIALS

The test compounds, obtained from Rhone Paulenc-Rhodia, Monmouth Junction, NJ included: (I) 2,6-dibromo-4-hydroxy-4-methyl-2,5-cyclohexadiene-1-one, (II) 2,6-dibromo-4,4-dimethyl-2,5-cyclohexadiene-1-one, and (III) 2,6-dibromo-4-hydroxy-4-methyl-2,5-cyclohexadiene-1-one acetate. Boll weevils used in the test were obtained from the Boll Weevil Research Laboratory, Mississippi State, MS and the tobacco budworms were from a Brownsville strain reared on a casein-wheat germ larval diet as described by Guerra (1970).

Laboratory Tests. For tests with the boll weevil, the 3 compounds were each incorporated into an artificial diet at concentrations ranging from 0.0001% to 0.1% (w/v) and then poured into 21-ml transparent plastic cups (15 ml each); untreated diet was used as check. The test diets were infested the next day with 20 one-day-old eggs/cup, 11-16 cups in each of 4 replicates/treatment. Test cups were held under a controlled daily photophase of 14 h at ca. 26.7°C until adult emergence took place (ca. 14 days). A topical dose of 10 µg in 1 µl acetone/insect was selected arbitrarily and doses of each compound were applied with a microapplicator to the dorsal tergites of the thorax of tobacco budworm larvae. The larvae used for these treatments ranged in age from 0 to 6 days (1st-3rd instar); control larvae of the same ages were treated with acetone only. Each bioassay was replicated twice and involved the treatment of at least 100 larvae. First and 2nd instar larvae were placed on filter paper during treatments to prevent their drowning in the excessive volumes of the test solutions. Thus, for these 2 larval stages much of the applied chemical was lost immediately after administration. After treatment, larvae were held individually in cups of diet under the same conditions of photoperiod and temperature. Insects that developed to the pupal stage were removed from diet cups and held in similar but empty cups (same light conditions but a temperature fluctuating between 23.3 and 25.6°C) until adults emerged.

Test solutions were prepared by dissolving the compound in 4 ml of redistilled acetone. Potted cotton plants were placed on a turntable in a spray chamber in the laboratory and treated with each chemical at a rate equivalent to 1.12 kg/ha. Leaves of check plants were sprayed with an equivalent volume of acetone. After each treatment, leaves were clipped from the plant and placed individually in petri dishes where they were exposed to a single 8-day-old tobacco budworm larvae (4th instar). At least 100 leaves were bioassayed from each treatment and tests were replicated twice. Surviving larvae were transferred to cups of diet 72 h later and handled as in the topical treatments.

Latent effects of the materials on development and reproduction were determined by setting up reciprocal crosses of single pairs of the treated and untreated adults that developed from the test larvae. Pairs were placed in 1-L glass jars provided with cheesecloth covers and resting sites and were fed on a solution of 5% sucrose. Temperature and light conditions were the same as for pupae. Fertility was estimated from observations of eggs oviposited during the period from day 2 to 7 of adult life.

Field-Cage Tests. Compound III was dissolved in acetone and applied at a rate of 1.12 kg AI in 57 L/ha with a hand-sprayer to squaring cotton (the equivalent of ca. 98,765 squares/ha) that was divided with plastic screening into individual plots 6 rows (1 m apart) wide and 5.8 m long. To insure infestation of all test plots, 4234 (400-2240/day) tobacco budworm moths were released May 16-18 and 20-22. Also, May 16-19 and 22, 23, and 27, 8500 (500-2000/day) boll weevils were released. Sprays were applied May 19, 21, 23, 27, and 30 and June 2, 5, 9, and 13. Whole-plant examinations were made prior to each treatment.

In addition, boll weevil damaged squares were collected and held for observations of adult emergence. Squares from which no adults emerged were dissected for observations of possible larval development. Tobacco budworm feeding damage to squares and bolls was recorded. Larvae infesting the treated plants were counted and categorized as small, medium, or large in size: small larvae were in the 1st or 2nd stage; medium larvae were 3rd-4th stage; and large larvae were older than 4th stage.

RESULTS

Laboratory Tests. Preliminary tests (not shown here) showed that compounds I and II had no effect on the development of boll weevil larvae when incorporated into the diet at the aforementioned concentrations. However, compound III demonstrated good activity; adult emergence was reduced more than 90% at concentrations of 0.001 and 0.0001% and was totally inhibited at concentrations of 0.05% or greater (Table 1). Results with treated leaves (Table 2) indicated that pupation of tobacco budworm larvae was reduced more by compounds II and III (35 and 24%, respectively) than by compound I (13%).

TABLE 1. Percentage Emergence of Adult Boll Weevils After Larvae were Fed Diet Containing Compound III.

Concentration (%)	No. of emerged adults	% emergence
0.1	0	0
0.075	0	0
0.05	0	0
0.025	11	1
0.01	19	2
0.001	52	5
0.0001	44	5
0	460	54

TABLE 2. Effects of 3 Cyclohexadiene Analogues on Tobacco Budworms Fed Treated (1.12 kg/ha) Cotton Leaves.

Compound	% pupation	% adult eclosion ^{a/}	% hatch
Untreated	71	85	72
I	62	31	23
II	46	22	9
III	54	56	0

^{a/} Based on number of insects that pupated.

On the other hand, adult eclosion was reduced more by compounds I and II (64 and 74%, respectively) than by compound III (34%). None of the test compounds showed any antifeeding properties.

Fecundity (eggs/♀/lifetime) was comparable in all treatments, but egg hatch was dramatically reduced by compounds I and II (68 and 87%, respectively) and completely inhibited by compound III. Although no detailed observations were made to determine why egg hatch was inhibited, some embryonic rudiments could be observed in the unhatched eggs indicating that fertilization took place but normal development was affected.

When applied topically to larvae of tobacco budworms of different ages (Table 3), all 3 compounds significantly affected larval development, pupation and adult eclosion; the order of toxicity was I>II>III. All the compounds significantly reduced adult emergence compared to the untreated check; however, compound I had significantly less effect than compounds II and III. These chemicals exhibited effects similar to those described by Guerra et al. (1973) for some mimics of the JH of the tobacco budworm. For example, when larvae were treated with these types of compounds, fewer adults emerged than did those from the untreated check. All 3 compounds acted as chemosterilants of the tobacco budworm, and % egg hatch of treated pairs was significantly less than hatch of eggs in the untreated check.

TABLE 3. Effect of Topical Applications of 3 Cyclohexadiene Analogues (10 µg/Insect) on Tobacco Budworm Larvae of Different Ages.^{a/}

Larval age (days)	% larval mortality				% adult emergence ^{b/}				% hatch			
	CK	I	II	III	CK	I	II	III	CK	I	II	III
QC ^{c/}	8	86	78	46	80	4	4	2	77	11	15	56
1	22	90	84	50	78	6	8	28	80	16	28	47
2	24	68	70	82	72	12	30	18	75	31	38	46
3	20	86	70	54	80	12	22	12	77	52	50	48
4	12	76	58	46	76	18	32	18	79	46	59	72
5	17	76	60	44	77	14	18	32	80	42	42	41
6	17	84	58	42	77	2	32	26	72	55	30	31
Mean ^{d/}	17d	81a	68b	52c	77c	10a	21b	18b	77b	36a	37a	49a

a/ 2 replicates, 100 insects/replicate.

b/ Calculations based on the number of surviving pupae.

c/ Neonate larvae.

d/ Means followed by the same letter for % larval mortality, adult emergence, and hatch are not significantly different from each other at the 5% level of probability by Duncan's multiple range test.

Larval development (14 days), pupal development (9 days), and pupal weight (214 mg) were comparable to those of the control for those insects that were not killed by the treatments (data not shown here). Fertility was reduced when reciprocal crosses were made between surviving treated insects and untreated moths, but differences among treatments were not significant (data not shown for reciprocal crosses). Also, effects were generally more pronounced when the larvae were less than 1-day-old at the time of treatment.

Field-Cage Test. Results of tests of the effects of spray applications of compound III on boll weevils and tobacco budworms in the field indicated that seasonal averages for populations of small larvae in treated (41,659/ha) and untreated (39,864/ha) plots were about equal. However, populations of medium and large larvae were reduced 26% and the number of damaged squares was reduced 23% in treated plots when compared with control plots (28,553 larvae/ha/sampling date and 104,872 damaged squares/ha/sampling date).

Although the populations of boll weevil larvae in treated and untreated squares were equal as indicated by dissection of the squares (data not shown here), we found a 61% reduction from the check in populations of adult boll weevils that emerged from 1/3-grown treated squares. Populations were not reduced in full-grown squares.

Our results indicated that compound III was ca. equally active against the boll weevil and the tobacco budworm in laboratory and field cage studies. Although the field treatments were not unusually effective against these pests, we believe that our results are generally encouraging and that compounds such as these tested may have sufficient potential to merit further studies for possible use in pest control.

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FIRST FINDINGS OF COTTON LEAFWORM LARVAE IN THE
UNITED STATES, 1922-79^{1/}

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ABSTRACT

The cotton leafworm, Alabama argillacea (Hübner), was a major cotton pest before the organic insecticides came into general use in the late 1940's. First findings of larvae were important because they indicated when population buildup and spread to other areas by subsequent generations might be expected. In recent years it has reentered the cotton insect picture. Date of 1st findings of larvae in Texas from 1922 to 1979, where they were found, and who made the findings are presented.

INTRODUCTION

Parencia (1978) states that "The cotton leafworm, Alabama argillacea (Hübner), has four stages in its life cycle - egg, larva, pupa, and adult. The female moth lays bluish-green eggs, singly on the undersides of cotton leaves. They hatch in 3 or 4 days. Newly hatched larvae are pale, dingy yellow. They feed only on the undersides of cotton leaves.

There are five larval stages. In the summer the larvae usually become full grown in about 2 weeks and are about 1 1/2 inches long. Their color varies with some being yellowish green, without prominent stripes, while others have a broad, black stripe and a fine yellow stripe down their backs. All cotton leafworm larvae have a distinguishing characteristic - on the top of each segment are four black dots that form a square.

When a larva has completed its feeding, it 'webs up' in the fold of a leaf and becomes a pupa. Other larvae may eat away the leaf, leaving the pupa hanging by a thread from the leaf vein or stem.

The pupal stage lasts about 1 week in mid-summer, but it may last up to 4 weeks in the fall. The moth, which is olive tan, has a wing-spread of about 1 1/2 inches. It is a strong flier and, frequently, is found in the northernmost states and in Canada. In these areas, it feeds on ripe fruits, such as peaches, grapes and cantaloupes.

The life cycle of the cotton leafworm is completed in about a month with two to eight generations in a year. Female cotton leafworm moths lay their eggs only on cotton in this country. However, in the mid-1960's, Hampea sp. was discovered as a host in Mexico."

Although the cotton leafworm was of little or no economic importance from 1950 to the late 1970's, it was a major pest up to the late 1940's and received as much attention as the bollworm, Heliothis zea (Boddie), in J. H. Comstock's report in 1880. Parencia et al. (1962) stated that "Before the organic insecticides came into general use in the late 1940's, the cotton leafworm often was

^{1/}This paper presents the results of research only. Mention of a pesticide does not constitute a recommendation for use by the USDA nor does it imply registration under FIFRA as amended. Also, mention of a commercial product does not constitute an endorsement of this product by the USDA.

one of the most destructive cotton pests. In outbreak years, supplies of insecticides, largely calcium arsenate, used to control it were usually exhausted before the end of the season. Since then, the widespread use of organic insecticides for the control of other cotton insects in southern Texas usually delays population buildup so that migration to other areas occurs too late to be of much importance."

Parentia and Rainwater 1964 stated that "The cotton leafworm is a tropical species that does not overwinter in the United States. Every year the moths migrate into this country from Central or South America. Each year since 1922 cotton leafworm larvae have been found first in southern Texas. Usually they were found in May but in some years in April, June or July. The cotton leafworms spread out from southern Texas. By the end of a cotton growing season they usually are present in every cotton producing state from Arizona eastward. Larvae have never been reported from California or Nevada.

In the years before and through World War II, 1st findings of the pest in the United States were of considerable significance. Usually economic damage and migration to other areas could be expected from subsequent generations. The planting period of the cotton crop in Texas ranges from February 1 in the Lower Rio Grande Valley to June on the High Plains. Thus population buildups in southern areas often resulted in injurious infestations in all other areas of the State as well as in Oklahoma and other States as the cotton growing season progressed. Tremendous amounts of insecticides were often required to prevent defoliation of the crop. It was of utmost importance that industry be prepared to make sufficient quantities of insecticides available to meet the growers' needs."

DISCUSSION

In 1978, the cotton leafworm reentered the cotton insect picture. The 1st cotton leafworm larva of the season was found in the southern tip of Texas on May 17, 1978, which was earlier than in any year since 1963. Subsequent reports of cotton leafworms in various State Extension Pest Management Reports were more numerous than in many years. They defoliated late-growing fields in Texas and an untreated field at Tifton, GA. They were controlled in mid-August in previously untreated fields in north central Louisiana and in 1 field in Mississippi, with adults being caught in large numbers late in the season in light traps at Stoneville, MS. Infestations were also reported from Alabama.

In 1979, the cotton leafworm again made its appearance in the mid-south and southeast. Infestations were controlled in a portion of a field in southern Arkansas and larvae were found in Stoneville, MS, for the 1st time in many years. It has been many years since cotton leafworms have been reported from states other than Texas. Apparently, changes in the use of insect control practices are causing the cotton leafworm to reemerge as a problem pest in cotton production.

First findings of cotton leafworm larvae in the United States from 1922 through 1979 were compiled from office files and are given in Table 1. The earliest finding was made on April 7, 1943, by R. L. McGarr and the author, near San Benito, TX. The latest finding was made on December 10, 1965, in Calhoun County, TX by the author. Reports of larval findings were not received only in 1928 and 1976 of the 58-yr period.

The author has been associated with research on cotton insects since 1933 when he was a seasonal assistant at the Cotton Insects Laboratory in Port Lavaca, Texas. He recalls that reports of 1st findings of cotton leafworm larvae had to be telegraphed to Division Headquarters in Washington, DC. Larvae were forwarded to the taxonomic group for verification of identification. This procedure was followed through 1953 when the author was at the Waco Cotton Insects Laboratory, which was responsible for conducting the Cotton Insect Survey for Texas and Oklahoma. Thereafter, he continued his interest in the 1st

TABLE 1. First Findings of Cotton Leafworm Larvae in the United States, 1922-79.

Date	Location (all in Texas)	Collected or reported by
July 3, 1922	Atascosa County	
June 8, 1923	Cameron County	T. C. Barber
July 1, 1924	do	do
May 22, 1925	do	do
May 18, 1926	Wharton County	W. L. Owen
July 5, 1927	Nueces County	
Apr. 29, 1929	Cameron County	T. C. Barber
May 20, 1930	do	F. L. Thomas
June 27, 1931	Nueces County	G. A. Maloney
July 11, 1932	Aransas County	
June 27, 1933	Calhoun County	K.P. Ewing & R.L. McGarr
May 24, 1934	do	do
May 23, 1935	do	do
May 5, 1936	do	do
May 27, 1937	Nueces County	F. L. Thomas
May 2, 1938	Calhoun County	K. P. Ewing
May 4, 1939	Cameron County	H. C. Massey
May 27, 1940	do	R. L. McGarr
June 6, 1941	Calhoun County	C. R. Parencia
Apr. 30, 1942	Hidalgo County	R.L. McGarr & C.A. Richmond
Apr. 7, 1943	Cameron County	R.L. McGarr & C.R. Parencia
May 25, 1944	Jim Wells County	F. I. Jeffrey
June 23, 1945	Cameron County	A. M. Thompson
June 7, 1946	do	L. M. Conn
June 21, 1947	Nueces County	D. H. Alexander
June 20, 1948	Refugio County	L. F. Greer
July 18, 1949	Calhoun County	do
May 10, 1950	Zapata County	H. L. Bales
July 17, 1951	Cameron County	Douglas Early
June 3, 1952	do	G. L. Smith
June 9, 1953	Calhoun County	C.E. King & N.G. Land
May 31, 1954	Cameron County	A. J. Chapman
June 9, 1955	San Patricio County	L. F. Greer
June 21, 1956	do	do
June 10, 1957	Cameron County	C. A. Richmond
June 10, 1957	Calhoun County	Edward Migura
June 10, 1958	do	C. L. Cook
June 3, 1959	Matagorda County	George Davis
May 31, 1960	Calhoun County	C. L. Cook
June 5, 1961	do	do
June 10, 1962	do	do
Apr. 29, 1963	Cameron County	O. T. Robertson
Apr. 29, 1963	Refugio and San Patricio	L. F. Greer
Counties		
June 18, 1964	Refugio County	do
Dec. 10, 1965	Calhoun County	C. R. Parencia
June 28, 1966	Refugio County	L. F. Greer
Oct. 21, 1967	do	do
Aug. 8, 1968	Cameron County	J. E. Houghtaling
July 7, 1969	Calhoun County	Gilbert Heideman
Aug. 3, 1970	Cameron County	J. E. Houghtaling
Oct. 1, 1971	Maverick County	M. J. Lukefahr
Aug. 25, 1972	Cameron County	Roy Parker
Nov. 6, 1973	Fort Bend and Wharton	C. L. Cole
Counties		

TABLE 1. First Findings of Cotton Leafworm Larvae in the United States, 1922-79.

Date	Location (all in Texas)	Collected or reported by
July 24, 1974-----	Hidalgo County-----	Byron Koenig-----
July 29, 1975-----	Cameron County-----	Nieves Hernandez-----
June 27, 1977-----	Walker County-----	Allen Dean-----
May 17, 1978-----	Cameron County-----	J. W. Davis-----
June 3, 1979-----	do-----	Ed Gage-----

findings, while remaining at the Waco Laboratory, in his assignment in Beltsville, MD, and since he has been in Stoneville, MS. The 1st findings are reliable since they were either made by entomologists or identifications were verified by entomologists. Findings by persons unknown to the author were verified by their supervisors.

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TOXICITY OF MONOCROTOPHOS AND CERTAIN RELATED COMPOUNDS
TO DIFFERENT STRAINS AND CROSSES OF THE TOBACCO BUDWORM^{1,2,3/}

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ABSTRACT

Topical applications of monocrotophos and Shell Development Co. compound SD-4092 (benzyl 3-hydroxycrotonate dimethyl phosphate) were equally toxic to larvae of a susceptible (S) strain of the tobacco budworm, Heliothis virescens (F.), but they were 4 and 2X less toxic, respectively, to a resistant (R) tobacco budworm strain. The E isomer of monocrotophos was the most effective of 10 related compounds tested, but the Z isomer was inactive. In 1969 and 1971, respectively, there were 6- and 27-fold differences in the LD₅₀ values for R- and S-strain insects treated with monocrotophos. In the same years the LD₅₀ values for the SXR progeny were ca. 2-fold greater than were those of the RXS progeny, and although the differences were not significant, they showed a trend for the patroclinous effect.

INTRODUCTION

In 1968 and 1969, certain strains of the tobacco budworm, Heliothis virescens (F.), were found to be resistant to monocrotophos (Wolfenbarger and McGarr 1970, Cantu and Wolfenbarger 1970, Wolfenbarger 1973) in the Lower Rio Grande Valley of Texas. However, this insecticide is currently registered for use against Heliothis spp. in cotton, and it was effective against field populations of these pests in tests conducted by Davis et al. (1975) and Harding et al. (1977).

Studies were conducted in 1971 and again in 1976 to determine whether there were any changes in the monocrotophos tolerance of tobacco budworms collected in the same area. In 1976, the E and Z isomers and several other related compounds were tested for their toxicity to R- and S-strains of the tobacco budworm and to compare their activity with that of monocrotophos. Also, to determine the mode of inheritance of the factor(s) of resistance R- and S-strains were crossed.

MATERIALS AND METHODS

In addition to monocrotophos, the following compounds were tested: crotoxyphos, Ciba Corp. experimental compounds C-776 (dimethyl phosphate ester of 2-chloro-N-ethyl-3-hydroxycrotonamide) and C-768 (dimethyl phosphate ester of 2-chloro-3-hydroxy-N-methylcrotonamide), dicrotophos, and Shell Development Co. experimental compounds SD-4092 (benzyl 3-hydroxycrotonate dimethyl phosphate), SD-11370 [(E)-3-hydroxy-N-octyl crotonamide dimethyl phosphate], SD-11374 [(E)-3-hydroxy-N-propylcrotonamide dimethyl phosphate], SD-11392 [(E)-N-hexyl-3-hydroxy-crotonamide dimethyl

^{1/} Lepidoptera: Noctuidae

^{2/} In cooperation with the Texas Agricultural Experiment Station, Texas A&M University, College Station, TX 77843.

^{3/} This paper reports the results of research only. Mention of a pesticide does not constitute a recommendation for use by the USDA nor does it imply registration under FIFRA as amended. Also, mention of a commercial or proprietary product does not constitute an endorsement for use by the USDA.

phosphate], and SD-41045 [α -methyl-p-(methylthio)benzyl 3-hydroxycrotonate dimethyl phosphate]. Except for the E and Z isomers of monocrotophos, all compounds were of technical grade.

The test insects were second-third stage (25-35 mg) larvae of the S-strain of tobacco budworms that had been reared at the Brownsville laboratory since 1969, and the progeny of moths of 3 different R strains collected as larvae from field cotton in the falls of 1969 and 1971 at Mante and Cuauhtemoc, Tamaulipas, Mexico, respectively, and in the summer of 1976 at Brownsville, Texas. Mante and Cuauhtemoc are ca. 83 km apart and each is ca. 670 km south of Brownsville.

Tests with the field-collected strains were conducted within 4-5 generations after the cultures were established in the laboratory. In 1969 and 1971, LD₅₀ values were determined for the R- and S-strains; reciprocal crosses among these insects provided the respective F₁ generations.

Neonate larvae were placed individually in 22-ml plastic cups containing about 10 ml of artificial diet on which they fed for 5 days. Graded concentrations of each compound were applied in 1 μ l of acetone to the dorsal thorax of each larvae. Approximately 200-500 insects were used for 2-8 replications of each dose. Mortalities were recorded after 48 h, and the LD₅₀ values (expressed as μ g/larva) were calculated with a log dose-mortality computer program. Differences of at least 5X in LD₅₀ values were considered statistically significant because therein the fiducial limits, which are about equal to 95% confidence intervals, did not overlap.

RESULTS AND DISCUSSION

Toxicity of Monocrotophos and Analogues. SD-4092 (the benzyl homologue of crotoxyphos) and monocrotophos (82% E isomer) were about equally toxic to the S- and R-strains tested, and crotoxyphos was active against the S-strain of the tobacco budworm (Table 1). The remaining compounds showed little or no activity. Our observations of the inactivity of compounds with 6 or 8 carbons on the N-alkyl moiety (SD-11370, SD-11374, and SD-11392) were comparable to those of Sun and Johnson (1969) in their studies with houseflies, *Musca domestica* L., and roaches, *Dictyoptera* sp. The E isomer of monocrotophos was highly toxic to both strains, with only a 3-fold difference between the LD₅₀ values for the R- and S-strains. Also, the E isomer was 13X and 18X more active than monocrotophos against the S- and R-strains, respectively. The Z isomer was totally inactive.

TABLE 1. LD₅₀ Values (μ g/larva) for Larvae of S- and R-Strains of the Tobacco Budworm. (1976)

Compound	LD ₅₀ (fiducial limits), 48-h posttreatment	
	S	R
Monocrotophos (82% <u>E</u> isomer)	1.9 (1.4-2.5)	7.2 (5.6-10.2)
Monocrotophos (<u>E</u> isomer)	0.15 (0.1-0.2)	0.4 (0.3-0.5)
Monocrotophos (<u>Z</u> isomer)	a/	a/
SD-4092	1.9 (1.5-2.3)	4.4 (3.1-6.7)
Crotoxyphos (SD-4294)	3.4 (2.5-5.0)	32.4 (15.9-126.9)
SD-11374	19.4 (12.0-95.7)	16.4 (12.3-29.9)
SD-14045	113.7 (35.2- ∞)	185.5 (73.0-1,400)

a/ Unable to determine because no more than 17% mortality occurred at the highest dose tested. Compounds SD-11370, SD-11392, C-776, and C-768 were also inactive.

Strains and Crosses of Strains. The LD₅₀ value for monocrotophos treatments of the Cuauhtemoc R-strain in 1969 was 6X greater than for the S-strain (Table 2). The LD₅₀ values for the Mante R-strain in 1971 (Table 2) and for the Brownsville R-strain in 1976 (Table 1) were 27X and 4X greater than the S-strain, respectively.

In 1969, the LD₅₀ values of the F₁'s of the reciprocal crosses (Table 2) were more like those of the R-strain than those of the S-strain; in 1971 (Table 2) they were intermediate, therein suggesting the incomplete dominance of the character for resistance. But, even though the LD₅₀ values of the F₁ RXS did not differ significantly from those of the SXR, in both years the resistance to monocrotophos appeared to be patrocinous. No other information is available on the patrocinous effect for monocrotophos, but Whitten (1978) did not show a patrocinous effect in his resistance studies with methyl parathion against the tobacco budworm.

TABLE 2. Mortality Data for Progeny of R-Strain Tobacco Budworms Collected in the Field at Cuauhtemoc (1969) and Mante (1971), Tamps., Mexico and a Laboratory S-Strain from Brownsville, TX and for F₁ Crosses Among These Strains.

LD ₅₀ (fiducial limits), 48-h posttreatment ^{a/}			
S	R	F ₁	
		RXS	SXR
<u>Cuauhtemoc (1969)</u>			
1.2 (0.7-1.9)	6.7 (4.3-10.1)	4.7 (1.3-8.3)	10.2 (5.6-21.1)
<u>Mante (1971)</u>			
3.7 (2.8-4.8)	99.7 (56.2-248.9)	16.2 (11.5-24.1)	27.9 (22.0-36.6)

a/ In data for crosses, female parent is listed first.

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NEW AND IMPROVED TECHNIQUES FOR MASS REARING BOLL WEEVILS^{1,2/}J. G. Griffin^{3/} and J. Roberson^{4/}

ABSTRACT

A new system was developed for handling rearing trays for planting through adult emergence of mass reared boll weevils, Anthonomus grandis grandis Boheman. Use of the new system resulted in reduced cost for equipment, and considerable savings in time and labor for handling and allowed for fumigation of each development and emergence unit after each production cycle. Experimental results indicated that the yield of adult weevils with the new system was equal and possibly superior to that obtained with the standard system.

INTRODUCTION

The standard procedure for mass rearing the boll weevil, Anthonomus grandis grandis Boheman, in the laboratory was described by Griffin et al. (1979). Although this procedure was more efficient than earlier ones described by Gast and Davich (1966) and Griffin and Lindig (1978), further improvement was still needed in the use of time and labor, especially in the preparation of trays for weevil emergence and in collection of the weevils. The weevil development part of the operation is not as labor intensive as the tray preparation for emergence and, the weevil collection after emergence. This paper describes new equipment and procedures developed to increase the efficiency and reduce the cost for mass rearing boll weevils.

DESCRIPTION OF NEW TECHNIQUES AND PROCEDURES

One major consideration of the total rearing concept involved compartmentalization within the larvae-holding and adult-emergence rooms with the objective of reducing 1) labor requirements, 2) construction cost for adult-emergence equipment, 3) microbial contamination when larval rearing tray covers were removed for adult emergence, and 4) environmental variation during larval development and adult emergence.

To provide the compartmentalization, we constructed cubicles, each with inside dimensions of 2 m wide x 2.45-m long x 1.85-m high, with a 1.06-m x 1.75-m door in one wall, (Fig. 1) in one of the regular development rooms and in the emergence room. Compartments this size accommodated a day's supply of the standard, infested rearing trays when production was ca. 7-8 million weevils/wk. Although the development and emergence rooms were already environmentally controlled, each cubicle was equipped with a thermostatically controlled electric heating unit, an air circulating fan, and an exhaust fan to increase the uniformity of air movement and temperature control around the trays. Air temperatures were maintained at 30° ±1°C and 31° ±1°C for the development and emergence cubicles, respectively, and their RH was maintained at 50±5%. The front (door) wall of the emergence cubicle contained 28 holes, each with a diameter of 5 cm. They were located to form 4 columns of 7 holes each (Fig. 2). Additionally, a 5-cm hole was cut in metal lids from 100-mm-mouth plastic jugs (3.8-L size), and one of these lids was centered and anchored over each hole in the wall with the top side of the lid against the

^{1/}Coleoptera: Curculionidae.

^{2/}Mention of a proprietary product in this paper does not constitute an endorsement of this product by the USDA. This research was done in cooperation with the Miss. Agric. and Forestry Exp. Stn., Mississippi State, MS 39762.

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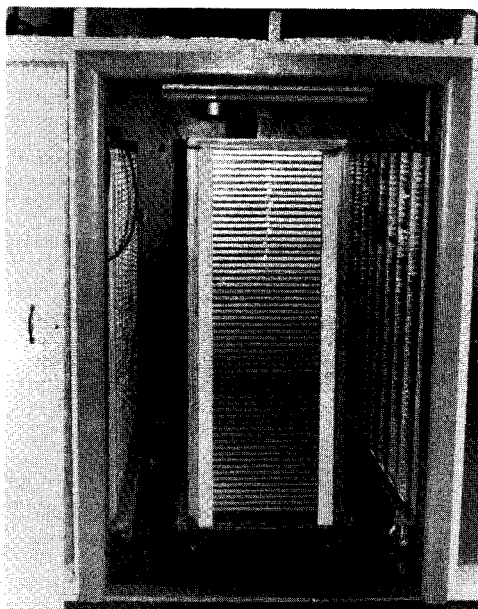


FIG. 1. Inside view of a development cubicle with rackveyors in place.

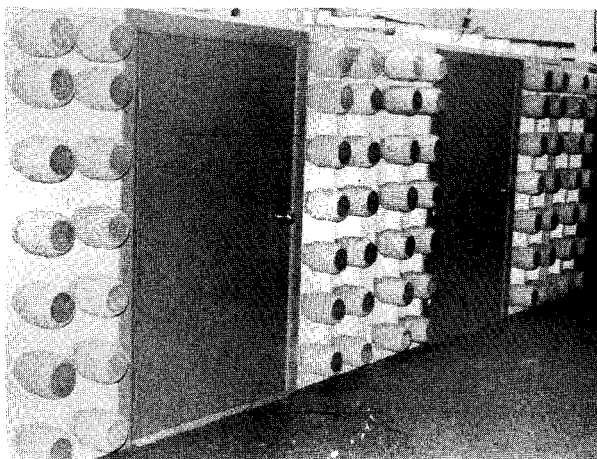


FIG. 2. Outside view of emergence cubicles, showing collecting jugs attached to wall.

wall's surface. A 10.2-cm-diameter hole was cut in the bottom of the jugs and then covered with a piece of 14-mesh stainless steel hardware cloth to make them weevil-proof. A jug was screwed in each lid on the wall, thereby enclosing the holes. The jugs were painted on the outside with a fluorescent lemon-yellow paint, the color being used for weevil traps.

METHOD FOR EVALUATING YIELD PERFORMANCE

The standard procedure and materials described by Griffin et al. (1979) were used to prepare and infest all of the rearing trays for the tests. Each day the infested trays were divided into 2 groups. One group of trays was stacked in a rackveyor (Griffin 1979), and the other group was stacked on a standard stacking cart. The loaded rackveyor was then rolled into a development cubicle in the development room, and the loaded cart was rolled into the regular development room. On the 13th day after the trays were infested, the loaded rackveyor and cart were rolled to the tray preparation room in the adult emergence area of the facility. A random sample (125-150) of trays was removed from the rackveyor, the covers were removed, and the opened trays were placed in a standard emergence box (Griffin et al. 1979). The box with trays was then rolled into the emergence room. Adult emergence holes ca. 1 x 2.5 cm were burned with an electrically heated rod into 1 end of the remaining trays on the rackveyor, and then the rackveyor was rolled into an emergence cubicle in the emergence room. The trays on the standard stacking cart were removed from the cart, the covers were removed, and the trays were placed in a standard emergence box. The loaded box was then rolled into the emergence room.

The emerged weevils were collected daily from each of the collecting systems. After collection, they were weighed and the number collected was determined from a standard number/weight table. The weevils from each group were collected over a 6-day period.

The test was replicated 10 times and the results were analyzed (Duncan's new multiple range test) to compare the number of adults emerged and collected per tray from: 1) trays handled by the standard rearing procedure (control), 2) trays held on the rackveyor in the development cubicle for weevil development and then placed in the standard emergence box for emergence (Treatment A) and 3) from trays held on the rackveyor both for development in the development cubicle and for emergence in the emergence cubicle (Treatment B).

RESULTS AND DISCUSSION

The average numbers of weevils/tray/treatment were as follows: 1) control - 410, 2) treatment A - 391, and 3) treatment B - 464. Although treatment B produced the highest number of weevils/tray, there was no significant difference ($P = 0.05$) between the treatments. Because the covers did not have to be removed from the trays in treatment B, this method of handling the trays saved labor and time and reduced possible contamination in the tray preparation room. The covered trays that were placed in the cubicle for weevil emergence had (visually) less contamination of the growing medium than the trays with the covers removed. Also, the covered trays maintained the medium in a better condition for adult emergence. The most important cost saving found from the study was for the emergence equipment. The cubicles were less expensive in construction cost per unit capacity than were the regular emergence boxes, and also required less labor for handling, cleaning, and sanitizing. It is estimated that the savings in equipment cost through use of the cubicle units could amount to \$100,000 or more for a facility that rears 18 to 20 million weevil/wk.

The infested trays were easier to place in the rackveyors than on the regular cart. Additionally, spacer screens as used on the cart were not necessary on the rackveyor.

The cubicles were constructed light-tight, and therefore essentially air-tight except for the openings for the collecting jugs. An air exhaust system vented through the top of the cubicles and the roof of the rearing building allowed the fumigation of each cubicle to kill microorganisms after each use. Plastic bags were fastened over the emergence jugs for fumigation.

Seven of the emergence cubicles were constructed and used in the production run for the North Carolina-Virginia Boll Weevil Eradication Trial program during the summer of 1979. Operational results from the new system were very satisfactory, and this type of unit is now used as the standard method for rearing the weevils. Time was not available to construct the quantity of development cubicles needed for the 1979 summer production run. Therefore, mass production tests are needed to check these units for their maximum potential.

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**OVIPOSITION, EGG HATCH, AND LARVAL SURVIVAL OF LONE STAR TICKS
HELD AT DIFFERENT TEMPERATURES AND HUMIDITIES**

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ABSTRACT

Engorged female lone star ticks, Amblyomma americanum (L.), held under different combinations of constant temperature (10-35° C) and relative humidity (RH, 35-95%) did not produce eggs at low humidities in low and moderate temperatures and were inefficient in egg production at 35° C and 35-65% RH. Females did not oviposit when held for 5-9 wk at 10° C, but then laid normal batches of eggs, which hatched, when transferred to 27° C and 95% RH.

At 30° C, no eggs laid at 35 or 55% RH hatched, and only 19% hatched of those laid at 65% RH. At 20-30° C with 75-80% RH, 64-81% of the eggs hatched, but with 90-95% RH > 90% hatched. No eggs hatched at 35° C at any RH.

Larvae were susceptible to humidities below 85% RH at any temperature but especially vulnerable at high temperatures. Maximum larval longevity in this study was 91-98 days at 20° C and 95% RH.

INTRODUCTION

Laboratory and field survival of the lone star tick, Amblyomma americanum (L.), depends on suitable temperature and on fairly high humidity (Lancaster 1957, Hair and Howell 1970). In fact, Sacktor et al. (1948) found a relative humidity (RH) of 85-100% most favorable which corresponds to the critical equilibrium humidity of 84-85% RH as reported by Sauer and Hair 1971. Nevertheless, replete female lone star ticks are fairly tolerant of both constant low RH (Lancaster and McMillan 1955, Sonenshine and Tigner 1969) and varying temperatures and RH (Lancaster 1958, Sauer and Hair 1971). Although oviposition occurs over a relatively wide range of temperatures and RH, eggs hatch only at higher RH. Patrick and Hair (1979) showed that field-released engorged females had only limited capacity to select oviposition sites most favorable for egg hatch.

Although unfed lone star tick larvae are known to survive about 10 wk in the laboratory at 21° C and high RH (85-100%) (Sacktor et al. 1948) and up to 9 months in the field under a wide range of conditions (Hooker et al. 1912), an exposure of only 2 days to low RH of 51 and 59% resulted in complete mortality of unfed laboratory-reared larvae (Lancaster and McMillan 1955). The latter authors therefore suggested that RH in some natural habitats, for example pastures, is too low to permit larval survival. Patrick and Hair (1979) found larval survival shorter in a meadow habitat (10-19 days) than in a wooded habitat (33-106 days), but the influence of temperature could not be separated from that of RH.

The purpose of the present study was to quantitate under laboratory conditions the effects of temperature and RH on egg production, hatching, and larval survival of the lone star tick. Thus, we determined the number and % hatch of eggs from engorged females held at different temperatures and RH. Also, we measured larval longevity at the same conditions since larvae have limited mobility and in nature would probably remain in the habitat where oviposition occurred.

1/ Acarina: Ixodidae.

METHODS AND MATERIALS

Recently colonized (F_2 or F_3) female lone star ticks were allowed to feed to repletion on stanchioned cattle, and then they were weighed and placed in individual 8-dr shell vials with the open end covered with organdy (for ventilation). Groups of 7-10 vials of females were then placed in circular glass chambers (3.78 L) containing different saturated salt solutions (Winston and Bates 1960) which produced different specific relative humidities. These chambers were held in controlled-temperature cabinets maintained at 10, 20, 30 and 35° C in constant darkness or at the standard rearing room temperature of 27° C with a 12:12 LD photocycle on countertops. All temperatures were measured with a conventional thermistor thermometer.

Although we obtained slightly different RH's for some of the salts than were reported by Winston and Bates (1960), relative humidities of 35, 55, 65, 75, 80, 85, 90, and 95% were maintained and checked periodically with conventional RH meters. Females held at 10° C were moved to 27° C and 95% RH after 5-9 wk since no oviposition occurred.

Unfed larvae from the same colony were exposed to the same temperatures and RH (groups of 150-200/4-dr vial with 5 vials/treatment). Larvae were considered dead if they could not be induced to move by warming the vials in the hands for a few moments while exhaling into the open vial.

Reproductive efficiency of the females was determined by using the Index of Reproduction Efficiency (REI = no. of eggs/g females) established by Drummond and Whetstone (1970). Since the REI for lone star ticks is not correlated with the weight of the engorged female (Drummond et al. 1971) but the number of eggs and the weight of the engorged female is highly correlated, reproductive efficiency at the different temperatures and RH was based on the REI and not on the number of eggs laid. Egg masses were weighed 20 days after oviposition began and the number of eggs was estimated by dividing by the known average egg weight for that particular set of conditions as determined by subsampling several egg masses. Minimum incubation time of eggs was based on the number of days from 1st oviposition to 1st hatch.

RESULTS AND DISCUSSION

Oviposition and Egg Hatch. No eggs were laid by females held for 5-9 wk at 10° C and 35, 65, 80, or 95% RH. However, when these ticks were transferred to 27° C and 95% RH, all laid average numbers of eggs (Drummond et al. 1971) that hatched. Thus at low temperature, RH had little influence on ovipositional success. Lancaster (1958) was able to prevent oviposition by lone star ticks for at least 85 days by holding them at 10° C, and Loomis (1961) reported that engorged females could be held up to 4 months at 5° C without oviposition.

The results obtained when the engorged females were held at 20, 30, and 35° C and 35, 65, 80, or 90% RH are reported in Table 1. At 20° C average preoviposition time ranged from 10.9 to 12.4 days, number of eggs from 5631 to 6637, and the REI from 9178 to 10,170. There was no apparent relationship between these parameters and % RH. Drummond et al. (1971) reported the REI at 27° C and 60-90% RH as 9943-10,334. However, we found no eggs hatched at 35 and 65% RH. Hatch was 65.4% at 80% RH and 92.4 at 95% RH. Hatch began from 45.1 to 51.7 days after the females began oviposition.

At 30° C, preoviposition time ranged from 6.0 to 7.6 days. Fewer eggs were laid and reproductive efficiency was slightly less with this temperature and all RH than at 20° C. Egg hatch at 30° C was similar to that observed at 20° C except that hatch was only 18.8% for eggs held at 65% RH. Eggs took only 25.5-28.8 days to begin hatching.

At 35° C, preoviposition time averaged 5.6-6.0 days. The fewest eggs were laid at this high temperature, and the REI average was only 5744 at 35% RH and 5440 at 65% RH. The average REI at 95% was 8099, a result which suggests that a

high RH can offset some of the adverse effects of higher temperatures on egg production. Since no eggs hatched at any RH, 35° C was above the upper temperature tolerance level for egg hatch.

TABLE 1. Number of Eggs, Preoviposition Time, Index of Reproductive Efficiency (REI), Egg Hatch, and Minimum Incubation Time for Engorged Lone Star Tick Females Held at Indicated Temperature and RH.

% RH	Engorged wt. (g)	Preoviposition time (days)	Average ^{a/}		REI	% Hatch	Minimum incubation time (days)
			Temp. °C	Number of Eggs			
35	0.6387	10.9	20	6246	9779	0	-
65	0.5866	10.9		5966	10170	0	-
80	0.6935	11.0		6637	9469	65	51.7
95	0.6155	12.4		5631	9178	92	45.1
35	0.6601	7.1	30	5874	8801	0	-
65	0.5983	7.0		5635	9367	19	28.0
80	0.5443	6.0		4949	9221	64	28.8
95	0.6487	7.5		5309	8258	90	25.5
35	0.6849	6.0	35	3962	5744	0	-
65	0.5924	5.6		3531	5440	0	-
80	0.6302	5.6		4890	7571	0	-
95	0.5170	6.0		4112	8099	0	-

^{a/} Each treatment was applied to 7-10 females.

At 27° C and 12:12 LD, the preoviposition time, number of eggs produced, and reproductive efficiency were independent of RH's between 55 and 95% (Table 2). No eggs hatched at 55 and 65% RH, but at higher RH's hatchability increased dramatically. Egg incubation time was not influenced by RH. Sonenshine and Tigner (1969) also found little effect of RH between 45 and 95% on oviposition at 27° C and little egg hatch below 85% RH, and Lancaster and McMillan (1955) found normal egg hatch above 73% RH. Our data indicated that the difference between 65 and 75% RH is critical for egg hatch at 27° C.

TABLE 2. Number of Eggs, Preoviposition Time, Reproductive Efficiency, Egg Hatch, and Minimum Incubation Time for Engorged Lone Star Tick Females Held at Indicated RH and 27° C.

RH%	Engorged wt.(g)	Preoviposition time (days)	Average ^{a/}		REI	% Hatch	Minimum incubation time (days)
			Number of eggs				
55	0.6194	6.6	5734	9257	0	-	
65	0.5559	5.6	5640	10146	0	-	
75	0.7156	6.3	6733	9409	81	34.3	
85	0.7264	7.0	6844	9422	85	31.3	
90	0.4843	6.3	4410	9106	92	31.0	
95	0.6023	6.3	6050	10045	96	31.7	

^{a/} Each treatment was applied to 7-10 females.

Larval Survival. The survival of larvae at 75 and 95% RH and 10, 20, 30, or 35° C is reported in Table 3. Higher temperatures shortened longevity at 75% RH, probably because of increased desiccation. All ticks held at 75% RH died after 3 days at 35° C; only 1% died of those held at 10° C. At 95% RH larvae lived somewhat longer at 35° C than at 10° C. Maximum longevity (91-98 days) occurred at 20° C and 95% RH. This compares with 70 days at 21° C reported by Sacktor et al. (1948).

TABLE 3. Survival of Lone Star Tick Larvae Held at Indicated Temperatures and RH.

Days	% Survival ^{a/}							
	10° C		20° C		30° C		35° C	
	% RH		% RH		% RH		% RH	
	75	95	75	95	75	95	75	95
1	100	100	95	100	90	98	75	99
2	100	98	83	100	61	95	10	99
3	99	97	70	100	31	95	0	99
4	98	97	37	100	15	95		99
5	97	97	15	100	0	95		97
6	94	90	2	100		93		97
7	90	85	0	100		93		97
14	2	54		99		92		85
21		26		97		88		78
28		20		84		82		63
35		5		78		69		43
42		0		74		49		27
49				68		22		22
56				65		6		14
63				61		0		8
70				54				0
77				49				
84				33				
91				19				
98				0				

^{a/} Based on 750-1000 larvae/treatment

The importance of maintaining a high RH at rearing temperatures of 27° C is emphasized by the data shown in Table 4. The length of time larvae survived increased from only 2-3 days at 55 and 65% RH to 84-91 days at 95% RH. A RH of 80% or higher was necessary for any substantial larval survival after 7 days.

TABLE 4. Survival of Lone Star Tick Larvae Held at Indicated RH and 27° C.

<u>% Survival at Indicated % Relative Humidity ^{a/}</u>							
Days	55	65	75	80	85	90	95
1	92	100	100	100	100	100	100
2	70	83	94	96	100	100	100
3	17	29	35	96	100	100	100
4	0	0	27	96	100	100	100
5			18	96	95	98	100
6			8	90	94	98	97
7			8	84	93	95	96
14			0	39	90	94	95
21				24	83	94	94
28				1	77	93	94
35				0	67	92	94
42					54	92	94
49					24	91	94
56					0	91	93
63						88	93
70						84	90
77						71	81
84						41	65
91						0	44
98							24
105							6
112							0

^{a/} Based on 750-1000 larvae/treatment

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COMPARISON OF SCREWORM^{1/} CAPTURES IN LIVER- AND SWORMLURE-
BAITED TRAPS IN A TROPICAL AREA OF SOUTHERN MEXICOJ. P. Spencer^{2/}, C. J. Whitten^{3/}, J. R. Coppedge^{3/}, and J. W. Snow^{4/}Screworm Research, AR, SEA, USDA, P. O. Box 986
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ABSTRACT

In a field comparison conducted in a coastal area of Chiapas, Mexico, screen traps baited with liver and wind-oriented traps baited with swormlure-2 caught equal numbers of screwworms, Cochliomyia hominivorax (Coquerel). However, the liver-baited screen traps caught significantly more C. macellaria (F.), sarcophagid species, and other miscellaneous flies than the wind-oriented traps, which greatly increased the time required for fly identification. In a 2nd test, plastic wicks and cotton wicks used as swormlure-2 dispensers were equally effective in attracting female screwworms, but plastic wicks were significantly more effective in attracting screwworm males as evidenced by trap captures.

RESUMEN

En una prueba de campo conducida en un area costanera de Chiapas, Mexico, trampas de malla cebadas con hígado y trampas orientadas por el viento cebadas con swormlure-2 capturaron moscas del gusano barrenador Cochliomyia hominivorax (Coquerel), en iguales numeros. Sin embargo, las trampas de malla cebadas con hígado capturaron significativamente mas C. macellaria (F.), y mas moscas de la familia Sarcophagidae y otros dipteros que las otras trampas y asi grandemente aumentan el tiempo gastado en la identificacion de moscas. En la segunda prueba, mechas de plástico y mechas de algodón usadas como dispensadores de swormlure-2 fueron iguales en la captura de hembras del gusano barrenador. No obstante, las mechas de plastico capturaron significativamente mas machos del gusano barrenador.

INTRODUCTION

Until 1978, a modified version of the Bishopp screen blow-fly trap (Bishopp 1916) baited with decomposing liver served as the standard screwworm fly survey trap for trapping native adults and sterile flies released by the Southwestern and Mexico-American Screwworm Eradication Programs. Jones et al. (1976) developed a synthetic attractant for adults of the screwworm, Cochliomyia hominivorax (Coquerel), that was later named swormlure. Coppedge et al. (1977) subsequently developed a new and more effective formulation of the attractant, which was designated swormlure-2 (SL-2). It has since replaced liver and swormlure as the standard attractant for screwworm flies. The availability of this attractant permitted greater flexibility in trap design and led to the development of a wind-oriented trap (WOT) (Broce et al. 1977) which, when used with SL-2, was much more efficient than the Bishopp style trap in capturing native

1/ Diptera: Calliphoridae.

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and sterile-release screwworm flies. The increase in efficiency plus ease of handling and lower cost of the WOT resulted in the establishment of an extensive screwworm survey system in the United States and northern Mexico. However, since SL-2 and the WOT were developed in semi-arid areas of the southwestern United States, we thought it was important to evaluate the effectiveness of the new system in the humid tropics of southern Mexico, an area geographically and climatologically different. Although Broce (unpublished data) conducted a preliminary comparison of SL-2 and liver in Veracruz, Mexico, a definitive evaluation was lacking. We report here an evaluation in southern Mexico of the WOT with SL-2, and 2 types of SL-2 dispensers.

METHODS AND MATERIALS

Six sites between the towns of Arriaga and Pijijiapan near the Pacific coast of the Mexican state of Chiapas were selected for the trap comparisons. This area is approximately 16 km wide and 100 km long with a mountain range on the northeastern side and the Pacific Ocean on the southwestern side. The sites were located 4-26 km apart along this strip of coastal plain. The screwworm population in this area has not been disturbed by the release of sterile screwworm adults; consequently, all screwworms captured were native adults.

For the 1st test, one screen trap and one WOT were suspended from trees 50m apart at each of the 6 sites. One kg of decomposing beef liver that had been "aged" for 7 days, and enough water to cover the liver were added to each screen trap. This mixture was changed every 2 wk. One 50-ml bottle of SL-2 was placed in each WOT. The SL-2 was dispensed with a 1.00-cm-diam, 12-cm-long braided cotton wick that protruded ca. 2.0 cm from the bottle. SL-2 was added as needed and the remaining attractant and cotton wicks were replaced every 3 wk. Flies were collected and traps at each site were rotated twice weekly. The comparison was conducted from July to September 1978.

A 2nd test to compare the effectiveness of attractant dispensers was conducted from October 1978 to March 1979 using the same 6 sites described above. The procedure used was similar to that used by Broce et al. (1979). Two WOT were positioned at each site. Preliminary testing indicated that traps baited with a single cotton wick captured more screwworm flies than traps baited with 2 cotton wicks while the opposite trend was indicated with plastic wick configurations. Therefore, one trap was baited with one 50-ml bottle of SL-2 with a single 1.00-cm-diam, 12-cm-long cotton wick. The 2nd trap was baited with two 50-ml bottles of SL-2, each bottle with a single 0.66-cm OD plastic wick. The plastic wicks had a polyester center with a propylene covering. The cotton wicks were placed in the bottles with ca. 2.0 cm protruding from the top. The plastic wicks were placed through closely fitting holes drilled in the bottle cap; these wicks also protruded ca. 2.0 cm. Attractant was added to the bottles with cotton wicks as needed with the SL-2 and wicks being completely changed every 3 wk. The plastic wicks and SL-2 in the other bottles were changed after 12 wk. Flies were collected from the traps and the traps were rotated twice weekly.

The numbers of screwworm flies captured during each collection period were small, therefore the data from each test were pooled by trap type on a weekly basis. The wide variation (0-8 in the 1st test and 0-26 in the 2nd test) of the pooled data necessitated the use of a non-parametric technique (Conover 1971) for statistical analysis.

RESULTS AND DISCUSSION

In the 1st test, both trap types captured screwworm flies at each site during the 11-wk test period. The SL-2 baited traps captured screwworm flies 10 of the 11 wk, and the liver baited traps 9 of 11 wk. Although more flies were caught in the SL-2 baited traps than in the liver baited traps (33 vs 28), there

was no demonstrable statistical difference in the capture rate on a weekly basis (Table 1). However, as expected, the number of flies other than screwworms

TABLE 1. Comparison of Fly Captures in Screen Traps Baited with Liver and those of Wind-Oriented Traps Baited with Swormlure-2.

Insect	Average no. of flies captured/wk ^{a/}		Statistical/difference ^{b/} between traps
	Swormlure-2	Liver	
Screwworm male	0.4	0.3	NS
Screwworm female	3.0	2.5	NS
<u>C. macellaria</u>	150.4	4285.8	Sig
Sarcophagidae	84.5	2595.1	Sig
Other Diptera	6.4	141.6	Sig

a/ Averages of 11 replicates.

b/ Statistical comparison made on weekly captures using the Sign Test;

NS = non significant and Sig = Significant at the P = .05 level.

caught in the traps baited with liver significantly exceeded the number caught in traps baited with SL-2. These results are similar to those of other researchers (Broce et al. 1977, Coppedge et al. 1977) who also reported that SL-2 was more selective than decomposing liver for attracting screwworms in arid and semi-arid climates. This selectivity is one of the principal advantages of the use of SL-2 with the WOT, since fly identification is expedited by a reduction in the number of unwanted or "trash" flies. This test was conducted in the rainy season during which a total of 800.4 mm or an average of 10.96 mm/day of rain fell in the test area. (Rainfall data were acquired from a meteorological station of the Secretaria de Agricultura y Recursos Hidraulicos located in the test area.)

In the 2nd test, traps with cotton wicks and plastic wicks caught equal numbers of female screwworm flies (Table 2). The traps with plastic wicks captured

TABLE 2. Comparison of Captures in Wind-Oriented Traps Baited with Plastic or Cotton Wicks.

Insect	Average no. of flies captured/wk ^{a/}		Statistical differences ^{b/} between traps
	Plastic	Cotton	
Screwworm male	1.0	0.4	Sig
Screwworm female	6.0	6.0	NS
<u>C. macellaria</u>	81.1	118.2	NS
Sarcophagidae	92.0	59.9	NS
Other Diptera	41.9	18.7	NS

a/ Averages of 23 replicates.

b/ Statistical comparison made on weekly captures using the Sign Test;

NS = non-significant and Sig = significant at the P = .05 level.

male screwworm flies 13 of 23 wk; the traps with cotton wicks captured male screwworm flies 5 of 23 wk. The capture of male screwworm flies was significantly higher in the traps with plastic wicks. Although traps with cotton

wicks caught greater numbers of C. macellaria (F.) and traps with plastic wicks caught higher numbers of sarcophagids and other flies, these differences were not significant. No rain fell during this test. Broce et al. (1979) compared the effectiveness of the 2 types of wicks in south Texas in a test of trap pairs, one trap utilizing 2 plastic wicks and the other 2 cotton wicks. While their data did not differentiate between the responses of the 2 sexes or that of native and sterile-release populations, the results indicated no significant differences in total screwworm fly captures between the wick types. Significantly more C. macellaria were captured in those traps with the cotton wicks in the Texas study, and the authors proposed that this difference may be attributed to the 10-fold increase in the amount of attractant dispensed by the cotton wicks. In our test, the single cotton wick dispensed about 4 X more SL-2 than did the 2 plastic wicks (Whitten, unpublished data).

These tests demonstrated that although SL-2 and the WOT did not appear to be as effective for trapping screwworm adults in humid regions as they were when tested in drier areas of the United States (Broce et al. 1977, Coppedge et al. 1977), their combined effectiveness was still equal to that of the liver-baited Bishopp trap. However, use of the new system in more humid areas is justified by its ease of operation and reduced captures of unwanted flies. Use of the plastic instead of the cotton wick is justified by the resultant increase in captures of male screwworms, and, as previously pointed out by Broce et al. (1979), the reduction in cost due to lower requirements of SL-2 (less evaporation/unit of time) as well as the reduction in hazard to personnel due to the lessened possibility of spilling attractant from bottles fitted with plastic wicks.

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EGG PRODUCTION EFFICIENCY OF FEMALE LONE STAR TICKS
OF DIFFERENT ENGORGEMENT WEIGHTS

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ABSTRACT

Female lone star ticks, *Amblyomma americanum* (L.), were allowed to feed to repletion on stanchioned cattle. After detachment, engorged females were grouped by weight and allowed to oviposit. Weight of eggs produced per female was significantly ($P = < 0.01$) linearly correlated with the weight of engorged females, but the relationship between weight and the efficiency of ticks in converting body weight to weight of eggs (Index of Conversion Efficiency (CEI): g eggs/g females) was curvilinear. Light females (0.09-0.12 g) had a CEI of 0.220 compared to 0.559 for median range females (0.54-0.57 g) and 0.449 for heavy females (0.99-1.15 g).

During oviposition, engorged females steadily decreased in egg production efficiency (no. of eggs laid/reduction in body weight), which dropped from > 14.0 during the 4 days after oviposition commenced to < 5.0 from day 20 to completion. About 96.3% of the eggs were laid during the 1st 20 days after oviposition commenced.

INTRODUCTION

Females of the lone star tick, *Amblyomma americanum* (L.), ingest a relatively large bloodmeal (avg 0.74 ml) (Sauer and Hair 1972) over a period of ca. 6-13 days (Lancaster 1973), then drop from the host and oviposit egg masses that are directly proportional in weight to the engorged weight of the tick (Drummond et al. 1971). The weight of the engorged female is therefore an important factor when egg production is used to determine the population dynamics of this species.

Engorgement weights of lone star tick females from cattle and other hosts in southeastern Oklahoma vary widely. We therefore conducted this study to examine egg production efficiency in relation to weights of engorged female ticks. We report here calculated daily and total egg production efficiencies for this species.

MATERIALS AND METHODS

Female lone star ticks (F_2 or F_3 ticks initially collected near Poteau, OK) were allowed to feed on stanchioned cattle according to the method of Gladney and Drummond (1970). Females that had engorged and detached were weighed, and then placed individually into small glass vials and held at 27°C and a 12:12 photocycle in glass chambers maintained at 93% RH (Winston and Bates 1960).

We divided 1 group of 190 ♀ ticks into 19 subgroups (10 ♀/subgroup) by weight (0.09-0.12, 0.14-0.17, 0.19-0.22... > 0.99 g). Females were allowed to oviposit to completion. Egg masses and females were then weighed, and females were subsequently returned to their vials for possible additional egg production.

We randomly selected a 2nd group of 20 females, ranging in weight from 0.1934 to 1.1812 g, to determine daily egg production efficiency. Females were weighed and transferred to fresh vials every 24 h during the ovipositional period.

1/ Acarina: Ixodidae.

The Index of Conversion Efficiency (CEI = g eggs/g females) described by Drummond and Whetstone (1970) was used to calculate the efficiency of the ticks in converting body weight to weight of eggs. For calculations of daily reproduction efficiency, we determined the number of eggs produced per female in relation to reductions in weight.

RESULTS AND DISCUSSION

Table 1 lists mean engorgement and egg mass weights, mean CEI, and % of weight lost during preoviposition and oviposition by females in the 19 weight ranges. Ovipositional success and % hatch were not correlated; all females laid eggs, nearly all of which hatched.

TABLE 1. Mean Engorgement Weight, Egg Mass Weight, Egg Production Efficiency (Index of Conversion Efficiency = CEI), and % Reduction in Body Weight Used During Preoviposition and Oviposition in Female Lone Star Ticks of Indicated Engorgement Weights.

Range	Mean engorgement wt (g) ^{a/}	Mean egg mass wt (g)	Mean CEI (g eggs/g ♀)	Mean reduction in body wt
0.09-0.12	0.1023	0.0220	0.215	36
0.14-0.17	0.1611	0.0620	0.385	50
0.19-0.22	0.2110	0.0818	0.388	48
0.24-0.27	0.2618	0.1121	0.428	56
0.29-0.32	0.3132	0.1475	0.471	62
0.34-0.37	0.3547	0.1830	0.516	66
0.39-0.42	0.4128	0.2341	0.567	70
0.44-0.47	0.4505	0.2626	0.583	71
0.49-0.52	0.5094	0.2848	0.559	68
0.54-0.57	0.5571	0.2947	0.529	67
0.59-0.62	0.6018	0.3108	0.508	64
0.64-0.67	0.6593	0.3567	0.541	66
0.69-0.72	0.7125	0.3848	0.540	67
0.74-0.77	0.7637	0.3925	0.514	64
0.79-0.82	0.8114	0.4114	0.507	63
0.84-0.87	0.8625	0.4390	0.509	64
0.89-0.92	0.9137	0.4486	0.491	61
0.94-0.97	0.9512	0.4756	0.499	62
0.99-1.15	1.0549	0.4737	0.449	57

^{a/} Based on 10 engorged ♀/range.

The weight of eggs produced per female was significantly linearly correlated ($r = 0.987$, $P = < 0.01$) with weight. Gladney and Drummond (1970) and Drummond et al. (1971) found a highly significant correlation between the number of eggs produced per female and the weight of the female. We found females in the light range (0.09-0.12 g) laid ca. 425 eggs, those in the median range (0.54-0.57 g) ca. 5670 eggs, and those in the heaviest range (0.99-1.15 g) ca. 9100 eggs.

The relationship between body weight and the efficiency of females in converting body weight to weight of eggs was curvilinear. Fitting the data to a 2nd-degree polynomial curve produced a highly significant correlation ($r = 0.908$) between CEI and mean weight. Females in the lightest range converted only 22% of their body weight to eggs compared to 53% for females in the median range and 45% for those in the heaviest range. Ticks that weighed 0.34-0.87 g converted 50-59%

of their body weight to eggs. This result was similar to the 60% average weight conversion reported by Drummond et al. (1971) for ticks in the same species and weight range.

The % of body weight lost during preoviposition and oviposition was also significantly ($r = 0.992$) linearly correlated to the CEI. The lightest females lost only 36% of their body weight compared to 68% for females in the median range and 45% for those in the heaviest range.

Table 2 shows the mean daily efficiency of 20 females in converting body weight to weight of eggs. Efficiency on day 1 could not be determined because females were weighed only after oviposition commenced. Efficiency of all females decreased steadily after the 1st few days of oviposition. Some of the heaviest females continued to oviposit after day 20, and their efficiency continued to decline each day. Most of the eggs (96.3%) were laid within 20 days after oviposition commenced; however, 1 female continued to lay eggs for 38 days.

TABLE 2. Reproductive Efficiencies (RE) of 20 ♀ Lone Star Ticks^{a/} at Indicated Days after Initiation of Oviposition.

Days	b/ RE	Days	b/ RE
1	--	11	11.9
2	14.5	12	10.8
3	14.2	13	9.7
4	14.0	14	9.8
5	13.4	15	8.9
6	13.9	16	8.3
7	13.1	17	8.4
8	13.1	18	6.8
9	13.0	19	5.2
10	12.4	20	4.6
		21-38	< 4.6

a/ Engorged females weighed from 0.1934 to 1.1812 g.

b/ RE = mean no. eggs laid/change in body weight (g).

Females lived for several days after oviposition ceased but continued to lose weight. About 33% of all females died within 1 wk after oviposition ceased compared to 46% after 3 wk, 74% after 5 wk, and 97% after 7 wk. Those ticks surviving longer than 3 wk after oviposition ceased lost an additional avg of 13.3% of their weight with no additional egg production.

These studies show that the Oklahoma strain of the lone star tick is nearly as efficient in egg production as the Kerrville laboratory strain (Drummond et al. 1971), and that egg production efficiency is dependent on the engorged female weight. The number of eggs produced daily by females decreases rapidly after the 1st few days of oviposition, as does egg production efficiency. We plan to use the results of this study in estimating population densities resulting from eggs produced by females of different engorgement weights. Also, the ability to determine when females are most efficient in egg production should help us understand the reproduction cycle of this species and benefit our rearing programs.

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EFFICACY OF PERMETHRIN EAR TAGS AGAINST
FACE FLIES^{1/} AND HORN FLIES^{1/} ON PASTURED CATTLE^{2/}

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ABSTRACT

Two different ear tags, polyvinylchloride (PVC) and polyurethane (PU), both containing 5 and 10% permethrin, were placed in each ear of cows and heifers. Face flies on the cattle were reduced throughout the test period by an average of 21 and 74% with the 5 and 10% PVC tags and 49 and 59% with the 5 and 10% PU tags, respectively. No horn flies were observed on any of the treated cattle during the 1st 12 wk of the test period.

RESUMEN

Se utilizaron "marcadores" de cloruro de polivinilo (PVC) o de poliuretano (PU) con un contenido de 5 y 10% de permetrina, colocadas en ambas orejas de vacas y terneras. A lo largo del experimento, la mosca de la cara, Musca autumnalis De Geer, se redujo, en promedio, 21 y 74% al usar PVC al 5 y 10% respectivamente, y 49 y 59% al emplear PU. No se observó la mosca del cuerno, Haematobia irritans (L.), en ninguna de las reses tratadas durante las 12 primeras semanas del experimento.

INTRODUCTION

Harvey and Brethour (1970) 1st reported achieving control of the horn fly, Haematobia irritans (L.), by attaching resin strips containing dichlorovos to the ears of cattle. Since then other workers have reported similar results with other insecticides. Stirofos impregnated ear tags were successfully used against the Gulf Coast tick, Amblyomma maculatum Koch, (Gladney 1976) and against the horn fly (Ahrens 1977, Sheppard 1980). Cattle ear tags containing fenvalerate also were found to be effective against the horn fly (Ahrens and Cocke 1979).

This paper reports on the efficacy of a permethrin (Atroban[®]) impregnated ear tag against the face fly, Musca autumnalis De Geer, and the horn fly.

^{1/}Diptera: Muscidae.

^{2/}The investigation reported in this paper (No. 80-7-121) is in connection with a project of the Kentucky Agricultural Experiment Station and is published with approval of the Director.

MATERIALS AND METHODS

Two cattle ear tag matrices, polyvinylchloride (PVC) and polyurethane (PU), each containing 5 or 10% permethrin, were tested against face flies and horn flies on pastured cattle from June 15, 1979 through October 15, 1979.

University of Kentucky white face Angus cow/calf herds were used in the test. The treatments and herd sizes were assigned randomly as follows:

- 5% PVC tags - 32 cows, 30 calves and 2 bulls
- 5% PU tags - 32 heifers, no calves and 2 bulls
- 10% PVC tags - 27 cows, 24 calves and 2 bulls
- 10% OY tags - 41 cows, 35 calves and 2 bulls
- Untreated - 23 cows, 15 calves and 1 bull

The cows and heifers were tagged with 1 tag/ear placed between the 1st and 2nd rib from the top of the ear halfway between the tip of the ear and the head. Bulls and calves were not tagged and bulls were removed from all the treated herds in mid-August. Each treatment herd was placed on similar 60-ha partially wooded fescue pastures.

The number of flies on 10 randomly selected animals from each treated and untreated herds were counted in the morning of the same day for each count. Non-tagged cattle in the treated herds were not used for fly counts. At the end of the test all treated cattle were examined for ear damage and tag loss. Analysis of variance tests were performed on the data and, where F values were significant, Duncan's multiple range test was applied.

RESULTS AND DISCUSSION

The 10% permethrin matrices resulted in the greatest face fly reduction throughout the test (Table 1). Fly reductions averaged 74% (range of 56 to 85%) for the 10% PVC tags and 57% (range of 21 to 75%) for the 10% PU tags. There were no significant differences between the effectiveness of these 2 tags except during wk 1, 3 and 11. The 5% PU averaged 49% reduction (range of 19 to 59%) for the season; however, this treatment did not differ significantly from either of the 10% formulations at wk 1, from the 10% PVC at wk 9, 13 and 14 or from the 10% PU at wk 3, 7 and 11. The 5% PVC was the least effective treatment against the face fly with an average of 21% reduction for the season (range of 0 to 48%). All treatments had significantly fewer face flies at each count than on the untreated cattle except for the 5% PVC at wk 9 and 13. The low reduction of face flies in the 5% PVC test may have been due to only 52% of the cattle being treated as compared to 100% of the cattle treated in the 5% PU test. We have obtained the same results in similar tests.

Complete horn fly control (Data not shown) was obtained with all treatments 1 wk after tagging and continued until the 13th wk of the test when the horn fly population increased and ranged from 0.7 to 5 flies/head for all treatments. During the 14th wk horn fly counts ranged from 1.5 to 11.5, at which time the test was terminated due to cool weather. Cattle with the 5% PVC and 5% PU tags had the most horn flies while those with the 10% PVC had the least. Horn fly counts on the untreated cattle averaged slightly more than 300/animal throughout the test. Complete horn fly control was achieved on the untagged bulls during the 2 months they were with the tagged cattle.

No necrosis or skin irritation of the ear area around the tags was found on any of the tagged cattle. Two 5% PVC tags and one 10% PVC tag were lost during the test.

TABLE 1. Efficacy of Permethrin Ear Tags Against Face Flies on Cattle.

Treatment		Mean no. flies at indicated wk posttreatment ^{a/}									
Concen- tration	Formu- lation	0	1	3	5	7	9	11	13	14	
10%	PVC	16.6 c	3.8 a	4.6 a	5.0 a	7.0 a	3.9 a	4.9 a	6.5 a	3.2 a	
10%	PU	10.7 b	11.2 b	16.1 b	8.0 a	9.0 ab	4.6 a	7.5 b	6.8 a	4.5 a	
5%	PVC	19.0 d	19.3 c	27.6 d	14.6 b	12.4 b	20.3 b	12.2 c	12.4 b	9.3 bc	
5%	PU	8.6 a	7.5 ab	18.9 bc	13.0 b	13.1 b	7.4 a	8.9 bc	7.5 a	5.8 ab	
Untreated		17.0 c	24.0 d	23.2 d	20.4 c	24.0 c	16.6 b	16.3 d	15.7 c	12.6 c	

^{a/} Means (within columns) followed by different letters differ significantly at the 5% level.

The reduction of face flies by use of permethrin ear tags appears promising and may be a breakthrough in face fly control. If the treated tags are applied early in the season in order to reduce the emerging overwintering or 1st generation of face flies (as early as May 1 in Kentucky), face fly control could possibly be greater.

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FREE AMINO ACIDS IN PSOROPTIC MITES^{1/ 2/}Fred C. Wright^{3/}, H. R. Crookshank^{4/}, and J. F. Cooper^{4/}

ABSTRACT

Free amino acids and related compounds were determined with an amino acid analyzer in extracts of the common scabies mite, Psoroptes ovis (Hering), and the ear canker mite, P. cuniculi (Delafond), of rabbits. The major components of the free amino acid pool of both species were alanine, glycine, asparagine-glutamine, serine, arginine, leucine, valine, and threonine (comprising ca. 74% of the total for P. ovis and ca. 71% for P. cuniculi). The quantity of total free amino acids in P. cuniculi was 1.2 times that found in P. ovis. All amino acid components necessary for the existence of the urea cycle were present in both species. Tryptophan, one of the amino acids essential for man and animals, and for insects associated with animal parasitism, was not found.

INTRODUCTION

The primary aim of the Scabies and Mange Research Unit of the U. S. Livestock Insects Laboratory at Kerrville, TX, is the eradication of the sheep scab mite, Psoroptes ovis (Hering), which causes scabies in both cattle and sheep. Often we use a surrogate, the ear canker mite, P. cuniculi (Delafond), of rabbits in our studies. Research thrusts include trying to find a better means by which to distinguish between the species of psoroptic mites, to determine exactly what parasitic mites feed upon, and to develop an artificial diet for the in vitro rearing of P. ovis in the laboratory. Data on the amino acid concentrations in the free amino acid pool found in mites would be very beneficial in this research.

The literature abounds in reports of free amino acids in insects; however, there is very little information on the distribution of these chemicals in mites. Rodriguez and Hampton (1966) used radiolabeled glucose to study essential amino acids in the twospotted spider mite, Tetranychus urticae Koch, and Bactor and Rasmy (1979) used paper chromatography in their study of free amino acids in the adult citrus brown mite, Eutetranychus orientalis (Klein). Zein-Edlin and Scott (1958) chromatographically determined free amino acids in the tropical rat mite, Ornithonyssus bacoti (Hirst). However, apparently there is no published information on the free amino acids in parasitic mites of cattle, sheep, or rabbits. The objective of this study was to determine and compare the free amino acids and related compounds in 2 species of psoroptic mites, P. ovis from cattle and P. cuniculi from domestic rabbits.

MATERIALS AND METHODS

Psoroptes ovis mites were obtained by scraping infested areas on cattle and P. cuniculi mites by collecting scabs from the ears of infested rabbits. The mites were then separated from the hair and scabs according to the method of

^{1/}Acarina:Psoroptidae.

^{2/}Mention of a proprietary product in this paper does not constitute an endorsement of this product by the USDA.

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Wright and Riner (1979). Three collections were made of *P. ovis* and 1 of *P. cuniculi*. After transfer to heat-sealable rice paper bags, the mites were washed with distilled water in an ultrasonic bath to remove external contamination and debris, and then held in a freezer at -60°C to await analysis. Extracts for physiological or free amino acid determination were prepared by homogenizing 250-mg samples of mites in 20 ml of 1% picric acid solution with a Brinkmann Polytron® homogenizer (Brinkmann Inst. Inc., Westbury, NY). The extracts were centrifuged, and an 18-ml aliquot of the supernatant was passed through a Dowex 2-X8 anion exchange resin column (200-400 mesh) (Bio-Rad Laboratories, Richmond, CA). The resin was rinsed with three 10-ml rinses of 0.02N HCL. The appropriate fraction was collected, carefully evaporated to dryness, and the residue taken up in 5 ml of 0.2N citrate buffer (pH 2.2). This solution was filtered through a 0.2 µm Millipore® filter, and 0.5-ml aliquots were analyzed for 41 amino acids and related compounds with a Beckman Model 121 automatic amino acid analyzer (Beckman Instrument Co., Palo Alto, CA) interfaced with an Infotronics Model CSI-210® digital integrator (Columbia Scientific Ind. Corp., Austin, TX). Amino acid calibration standards, (P-AN and P-B obtained from Hamilton Co., Reno, NV), were used to identify and convert the sample concentrations to micromoles (µM) of amino acid/100g of sample. Asparagine and glutamine elute together, and the reported values were based on an asparagine standard.

RESULTS AND DISCUSSION

The 22 conventional amino acids detected and their calculated values are shown in Table 1. Concentrations of other amino acids and related compounds in the free amino acid pool are given in Table 2. Five compounds not detected in either species were tryptophan, sarcosine, carnosine, anserine, and hydroxylysine.

TABLE 1. Conventional Free Amino Acids in *Psoroptes ovis* and *P. cuniculi*.

Amino Acid	P. ovis		P. cuniculi	
	µM/100g ^{a,b/}	% of total	µM/100g ^{a/}	% of total
Alanine	1950	19.33	1760	14.00
Glycine	1350	13.38	1910	15.19
Asparagine ^{c/} Glutamine ^{c/}	1170	11.60	1640	13.05
Serine	710	7.04	850	6.76
Arginine	610	6.05	990	7.88
Leucine	600	5.95	610	4.85
Valine	570	5.65	610	4.85
Threonine	550	5.45	520	4.14
Glutamic acid	420	4.16	460	3.66
Proline	390	3.87	490	3.90
Isoleucine	320	3.17	340	2.70
Tyrosine	270	2.68	390	3.10
Lysine	260	2.58	540	4.30
Methionine	250	2.48	230	1.83
Phenylalanine	200	1.98	260	2.07
Hydroxyproline	180	1.78	400	3.18
Histidine	170	1.68	310	2.47
½-Cystine	50	.50	130	1.03
α-Aminobutyric acid	30	.30	20	.16
Aspartic acid	20	.20	60	.48
Ornithine	10	.10	30	.24
Citrulline	10	.10	20	.16
TOTALS	10090	100.03	12570	100.00

^{a/} Calculated per 100g of mites.

^{b/} Average of 3 samples collected at different times.

^{c/} Asparagine and glutamine eluted together and reported values are based on the asparagine standard.

In earlier work on phytophagous mites, Rodriguez and Hampton (1966) detected 21 amino acids in young adult T. urticae females fed on bean plants, and Bector and Rasmy (1979) reported 12 amino acids and 2 amides in phytophagous adult E. orientalis females fed on seedlings of sour orange. Also, Zein-Edlin and Scott (1958) reported 12 amino acids in starved parasitic adult O. bacoti. The free amino acids reported in these species of mites were also detected in both P. ovis and P. cuniculi in this study (Table 1). We, however, detected additional amino acids or related compounds not reported by these earlier researchers (Table 2). Such detection could have resulted from improved techniques and instrumentation. Our sampling method enabled us to collect primarily nymphal and adult mites; however, we made no attempt to avoid collecting larval mites, so our samples (>250 mg each) should have represented a cross-section of the population at the time of collection. Thus, the concentration of amino acids and related compounds found in this study did not represent any one particular life stage. Other researchers have found changes in the concentrations of free amino acids due to metamorphosis in insects (Hackman 1956, Patterson 1957, Bursell 1963, Nowosielski and Patton 1965, Levenbook and Dinamarca 1966, Bector 1975).

TABLE 2. Other Amino Acids or Related Compounds in Psoroptes ovis and P. cuniculi.

Compound	$\mu\text{M}/100\text{g}$ of mites	
	<u>P. ovis</u> ^{a/}	<u>P. cuniculi</u>
Creatinine	4290	5150
Urea	2370	2140
Ammonia	300	400
Ethanolamine	150	170
Taurine	70	350
Phosphoserine	80	90
Phosphoethanolamine	40	70
Cystathionine	50	50
γ -Aminobutyric acid	30	40
α -Aminoadipic acid	20	10
β -Alanine	20	40
β -Aminoisobutyric acid	10	40
1-Methylhistidine	10	40
3-Methylhistidine	10	ND ^{b/}

^{a/} Average of 3 samples collected at different times.

^{b/} None detected.

In our study, alanine, glycine, asparagine-glutamine, serine, arginine, leucine, valine, and threonine (Table 1) were the major components of the free amino acid pool, comprising 74.45% in P. ovis and 70.72% in P. cuniculi. The pattern of the decreasing amounts of the amino acids of the pool was not the same for both species; however, this is not unexpected, as Micks and Gibson (1957) have reported variations in amino acid patterns among different species of both insects and ticks. Also, the amount of the amino acids and related compounds in P. cuniculi is about 1.2 times that of P. ovis, possibly a result of differences in feeding habits and food sources (host animal). For example, P. cuniculi has been shown to ingest erythrocytes during feeding (Wright and DeLoach 1980), whereas P. ovis has been reported to feed on lymph and serous fluids. Alanine was found in the greatest concentration in P. ovis. Glycine, the simplest amino acid, was found in the greatest concentration in P. cuniculi.

Rodriguez and Hampton (1966) suggested the presence of the 'ornithine cycle' (urea cycle) in the twospotted spider mite but had insufficient evidence with which to confirm their suggestion. In our work, all components necessary for the urea cycle (excluding cofactors and enzyme systems) were present in both species, namely arginine, ornithine, citrulline (Table 1), and urea (Table 2). Ammonia was also present. This compound, a toxic substance in most instances, is used in the formation of the amides, asparagine and glutamine, from aspartic and glutamic acids. Glutamine then serves as an amine donor in general metabolism. The combined sample of asparagine-glutamine represented 11.6-13% of the total amino acid pool in P. ovis and P. cuniculi, respectively (Table 1).

Creatinine, an anhydride of creatine which is a possible metabolic product of arginine, glycine, and methionine, was found in large quantities in both species (Table 2). Tyrosine, which can be formed from phenylalanine, is very important in sclerotization in insects. This amino acid was present in both P. ovis and P. cuniculi.

The amino acids that occur in the proteins of mammals are also present in insects; our research indicates that, except for tryptophan, these amino acids are also found in the psoroptic mites. It is interesting to note that, even though tryptophan is an essential amino acid for mammals and was found to be essential for insects associated with animal parasitism (House 1958), it was absent in both the phytophagous (Rodriguez and Hampton 1966, Bocktor and Rasmy 1979) and parasitic mites (Zein-Edlin and Scott 1958, and the present study). Some other compounds that do not enter into protein molecules were also found in this study, i.e., ornithine, β -alanine, taurine, α -aminobutyric acid, and γ -aminobutyric acid.

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REARING OF TEXAS TABANIDAE (DIPTERA). III. TRAPPING, SURVIVORSHIP,
AND LIMITED REARING OF HYBOMITRA LASIOPHTHALMA (MACQUART) 1/

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ABSTRACT

From 29 March to 12 May 1978, 12,713 females of Hybomitra lasiophthalma (Macquart) were collected live on the Navasota River floodplain in south-central Texas and maintained in the laboratory for rearing purposes. Statistical analysis of these collection data showed that productivity of modified Manitoba Traps was related to trap site. Catches survived well in ventilated collection containers provided with sugar water. Samples of caged females collected early in the season lived 2 wks but survivorship dropped sharply in samples taken later in the season. Females refused beef blood from animals and from prophylactics (with ATP) but specimens which were partially engorged when collected did oviposit in the laboratory. Four lots of larvae, containing 4, 12, 16, and 19 specimens/pan of peat and sand, were reared together at 27°C until they matured in ca. 2 months, at which time they entered diapause. No evidence of cannibalism was observed. Although adult survivorship was encouraging, the short duration of wing life suggested that this species adapts to cage confinement very poorly, and therefore, will be difficult to colonize.

INTRODUCTION

Rearing attempts to establish several species of Texas Coastal Plain Tabanidae were begun in 1977 but initial work was impeded by the very small populations of most species available; however, Hybomitra lasiophthalma (Macquart) was an exception. Because this tabanid was locally available in large numbers and because it is abundant, widespread, and economically important in the U.S., studies were begun to establish it in the laboratory.

METHODS AND RESULTS

Hybomitra lasiophthalma is locally abundant in a floodplain forest of the Navasota River ca. 60 km east of Bryan at the intersection of the river with FM 2038, a bridge known locally as Channey Crossing. The floodplain at this point forms a 10 km-wide basin overlain with 3 natural vegetative cover types, described previously by (Thompson 1977).

Two types of traps were used in the study: the Gressitt Trap of Gressitt and Gressitt (1962), as modified by Thompson and Holmes (1977); and the Manitoba Horse Fly Trap of Thorsteinson et al. (1958), as modified by Thompson (1969) and Thompson and Gregg (1974). The solid ceilings of the collection containers from both traps were replaced with 0.3 cm-mesh hardware cloth in order to reduce heat within them and to permit trapped females to feed on gauze pads saturated with a 10% sugar solution which were resting on these screen ceilings.

1/ This paper reports the results of research only. Mention of a commercial or proprietary product does not constitute a recommendation for use by the U.S. Department of Agriculture.

One modified Gressitt Trap (GT) and 8 modified Manitoba Traps (MT) were operated along the path of an L-shaped woodland roadway between adjacent primary and secondary channels of the river. Traps were located 70-90 m apart along the edge of this trail; the trap line began at one end of the L with the GT, followed by 3 MTs along the remaining length of the 1st leg and the remainder along the length of the 2nd leg. The attractancy of each trap was augmented with CO₂ produced by a continuous source of dry ice. Traps were serviced daily from 14 March through 12 May.

Abundance and Seasonal Distribution of *H. lasiophthalma*. A total of 12,713 females and 4 males were taken in the 9 traps over the 6-wk period 29 March through 12 May. Twelve other tabanid species were taken during this period but *H. lasiophthalma* comprised 86.4% of the total catch. The only other abundant species was *Tabanus subsimilis subsimilis* Bellardi, a form representing 9.3% of the total catch.

Trap Selection and Site Location. Manitoba Traps collected 9.3-21.4%/trap of the total taken from all 9 traps (Table 1). Although the GT took

TABLE 1. Relative Effectiveness of Trap Type and Location on Collection of *H. lasiophthalma*, Navasota River Bottom, 29 March-12 May, 1978.

Trap No.	Manitoba traps								Gres. trap	Grand total
	1	2	3	4	5	6	7	8		
Total no. flies	1204	1184	1764	1331	1259	2722	1286	1453	510	12713 %
% of catch	9.5	9.3	13.9	10.5	9.9	21.4	10.1	11.4	4.0	100
Daily average	27	26	39	29	28	60	29	32	11	-

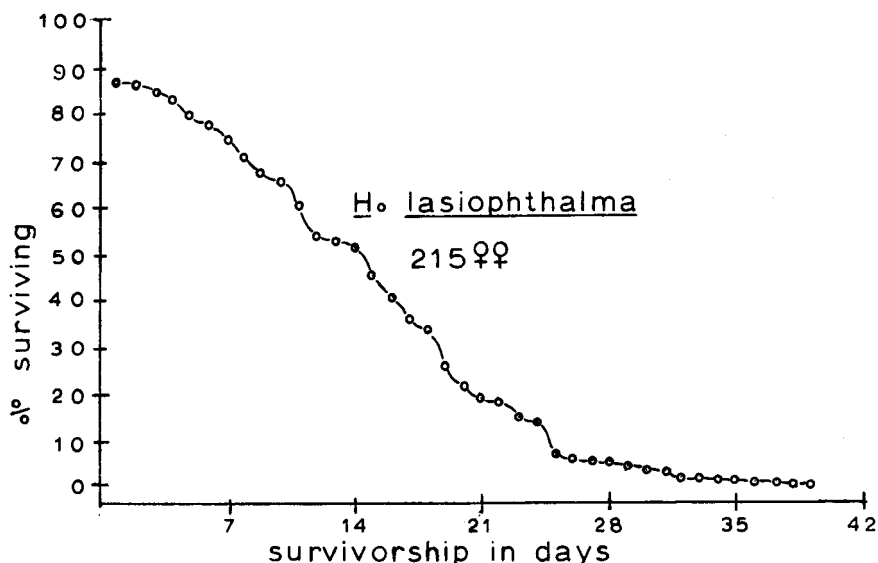
only 4% of this total - a figure including less than half of the lowest MT catch - it caught the only (4) males of this species taken. The data for trap sites were statistically analyzed and results indicated that trap no. 6 consistently collected more females throughout the season of activity ($P < .05$). In addition, traps 3 and 5 were consistently more productive than the remaining traps. Several other traps produced higher catches during certain parts of the season but their catches also showed larger variability. The high catches of some traps (nos. 6, 3, and 8) were associated with 2 recognizable factors - the extent of standing water in nearby woodland rainpools and the duration of that situation over the period of fly activity.

Handling of the Catch. Catches were transported to the laboratory in insulated picnic chests, chilled in a freezer for ca. 3 mins, and placed in the modified Animal Trap cages described previously (Thompson 1969; Thompson et al. 1979) where they remained until they died. These insects were held in ambient light (from 2 large windows) at ambient temperature ($23 \pm 2^\circ\text{C}$) and relative humidity (55-85%) in the laboratory. Supplemental moisture was provided by 10 cm X 10 cm cotton gauze pads placed atop the cages and saturated with distilled water. A similar arrangement provided 10% sucrose solution.

Longevity. The effect of age on consecutive samples of females can be observed in Table 2, where the 50% survivorship value drops as the collection date and age of the field population advance. Longevity for the April 6 cage is shown in Fig. 1.

TABLE 2. Survivorship of 7 Cage Populations of H. lasiophthalma.

	Collection date						May 1
	April						
	3	6	11	18	27	28	
Total no. flies	119	215	172	119	153	383	184
Day of 50% survivorship	17	14	13	10	3	4	5

FIG. 1 Survivorship of H. lasiophthalma cage population collected April 6, 1978.

Wing Wear. Observations of the wing life of H. lasiophthalma females suggest that this species settles down to cage life very poorly; only half the females retained 1 wing membrane intact after 3 days and after 1 wk, none did.

Feeding. Only 1 of 200 females fed on warmed and citrated whole beef blood during each of 2 exposures with the feeding stimulant, ATP, at 10^{-2} M and 10^{-3} M. The blood was contained in prophylactics derived from animal membranes (Fourex Natural Skins, Schmid Laboratories, Inc., Little Falls, NY 07424). In 1 exposure, 25 females refused warmed, citrated blood placed on cotton gauze. Also, 2 groups of 25 flies each refused blood from a wet, closely clipped cow on 2 different occasions.

Oviposition. Although no eggs were laid by any of the females exposed to blood in the laboratory, 44 egg masses were laid by trapped females that were partially engorged at the time of collection. (On 5 occasions, free blood was found on the cotton gauze sections which were saturated with sugar water and held on top of trap collection containers.) Most egg masses (38) were deposited on the wood frame of the cage while the remainder were laid on the stockinette sleeve.

Rearing. Eggs and larvae were maintained with the same methods described for Tabanus nigrovittatus (Thompson et al. 1979). Although H. lasiophthalma produced 44 egg masses, most failed to hatch. Fifty-one larvae in 4 lots reached maximum growth in ca. 2 months at 27°C and entered diapause at that time. No evidence of cannibalism was found in these 4 lots which included 4, 12, 16, and 19 larvae, respectively.

DISCUSSION AND CONCLUSIONS

The efficiency of the MT in collecting H. lasiophthalma is demonstrated by catches averaging 1525 specimens/trap in 1978. As with similar attempts by other workers, our efforts to induce feeding by this species were not encouraging. During transmission studies of equine infectious anemia in horses, McClain et al. (1975) reported that only 13 of the 348 H. lasiophthalma females tested penetrated the skin of host ponies; only 5 of these 13 specimens engorged. A short time later, Thomas and Gooding (1976) reported feeding 7 females on defibrinated beef blood through membranes during their studies of blood meal volume and digestive enzymes. These latter results, as well as those obtained by Thompson et al. (1979) with I. nigrovittatus, suggest that the right combination of blood source, membrane, and temperature could induce feeding by H. lasiophthalma. Once feeding was accomplished, oviposition and development of viable eggs in the laboratory is probable.

Hine (1906), Philip (1931), and Schwardt (1936) successfully reared adults of H. lasiophthalma from egg masses collected in the field. Schwardt's contribution was the most informative of these reports, but he failed to describe the nature of the substratum or to note whether larvae were held separately or en masse. Using crayfish as food, he reared larvae from one egg mass collected in early May and 31 adults emerged the following spring.

Our results show that the larvae of H. lasiophthalma can be reared to maturity en masse without the effect of cannibalism becoming restrictive. However, for these insects to complete development in 3 months or less, procedures must be developed to prevent diapause. At this point, the prospect of maintaining adults with the hope of mating is not encouraging, even after considering the degree of survivorship we observed in field-collected material. These problems should not be allowed to discourage research on rearing this important pest species.

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THE COURTSHIP BEHAVIOR OF THE HORN FLY^{1/}, ^{2/}

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ABSTRACT

Courtship behavior of the horn fly, Haematobia irritans irritans (L.), was studied by direct observation and photographic analysis. Subsequent to orientation, the male makes initial contact by tapping the female's abdomen with his prothoracic tarsi. The wings of the female become extended by the male's prothoracic legs as he initiates mounting activity. Male wing vibration also commences at this time. The male continues to move anteriorly on the female, reaches down with his prothoracic tarsi and encircles those of the female and proceeds to alternately lift her forelegs in a rapid manner. He then moves posteriorly on the female and attempts copulation following a short period of genital orientation and contact.

INTRODUCTION

Although numerous behavioral studies have been undertaken relative to courtship and mating habits of the house fly, Musca domestica L., (e.g., Colwell and Shorey 1975, 1976, 1977; Murvosh et al. 1964; Soliman et al. 1968; Tobin and Stoffolano 1973a); face fly, Musca autumnalis DeGeer, (e.g., Lodha et al. 1970, Teskey 1969, Tobin and Stoffolano 1973b); and stable fly, Stomoxys calcitrans (L.), (Anderson 1978), the courtship behavior of the horn fly, Haematobia irritans irritans (L.), has not been described. Horn flies have been observed in copula on the host (Bruce 1964, McLintock and Depner 1954), on vegetation (Bruce 1964) and in the laboratory (Harris et al. 1968, Schmidt 1972); however, these authors did not characterize the specific action patterns comprising the mating ritual of the horn fly. The purpose of this study was to examine and describe the elements of stereotyped horn fly courtship by direct observation and frame-by-frame analysis of 16-mm motion picture photography.

MATERIALS AND METHODS

Horn flies were reared in the laboratory according to procedures described by Bay and Harris (1978). Flies were permitted to emerge over a 12-h interval after which time they were immediately separated by sex on a chilling table at 2°C and held in cages for testing. Three- to 5-day-old virgin male and female individuals subsequently were paired in clear, cylindrical plastic chambers (70 x 100 mm) or rectangular arenas (25 x 25 x 15 mm) for observation and filming, respectively.

^{1/} Diptera: Muscidae

^{2/} Technical article No. 16412 from the Texas Agricultural Experiment Station. This research was conducted in cooperation with AR, SEA, USDA, College Station, TX 77841.

Photographic techniques were similar to those employed by Colwell and Shorey (1975) and Tobin and Stoffolano (1973a, 1973b). Filming was conducted with a 16-mm movie camera at 32 frames/second in an enclosed room at 27°C and 70-80% RH. Individual elements of courtship were determined by frame-by-frame analysis of filmed sequences and/or direct observation of mating behavior.

RESULTS AND DISCUSSION

Similar to other muscoid flies, normal male horn fly courtship behavior consists of a stereotyped continuum of action patterns directed towards the female; however, the individual elements of such behavior appear somewhat different among the various species studied. Although courtship *per se* commences when a male mounts a female and terminates as copulation is initiated, several elements prior to mounting as well as copulation are included in the following arbitrary segregation of events: orientation, abdominal tapping, wing extension, mounting, protarsal manipulation, backing, genital orientation, genital contact and copulation. The sequence and timing of the elements of horn fly courtship, with the exception of orientation, are provided in Table 1. Orientation was not timed because it could not be accurately differentiated from apparent random movement by the male.

TABLE 1. Time, in Seconds, of the Elements Comprising Horn Fly Courtship Behavior.

Element ^{a/}	Time (Seconds)
Abdominal Tapping	4/32 - 5
Wing Extension	} 11/32 - 17/32
Mounting	
Protarsal Manipulation	7/32 - 11/32
Backing	4/32 - 16/32
Genital Orientation	3/32 - 22/32
Genital Contact	2 - 4
Copulation	51 - 302

^{a/} Orientation was not timed because it could not be accurately differentiated from apparent random movement.

Orientation (Fig. 1A) of the male to the female is the initial step in horn fly courtship. As with the house fly and face fly, no definitive orientation posture was perceived in male horn flies. A resting male usually approached a female, at a distance of several centimeters, from the rear and positioned himself directly behind her and facing the same direction. Less frequently, in-flight males were observed striking other in-flight or resting individuals.

Initial contact is made by the male tapping the dorsum of the female's abdomen with his prothoracic tarsi (Fig. 1B). Although Soliman et al. (1968) stated that the male house fly may also tap the female's abdomen during orientation, no such activity was reported by Tobin and Stoffolano (1973a, 1973b) for either the house fly or face fly. Colwell and Shorey (1975), however, occasionally observed a male house fly touch the female, although not necessarily on her abdomen, prior to "jumping." Such species-specific behavioral responses may result from the spatial separation of male and female house flies and face flies thereby necessitating the more rapid and aggressive method of initial sexual contact described by Hewitt (1914) as a "strike" and Soliman et al. (1968) as a "jump." Due to the proximity of the sexes on the bovine host,

male horn fly intent may be conveyed by a more discerning manner of communication that will distinguish courting males from noncourting males or other females.

Subsequent to abdominal tapping, the wings of the female become extended ca. 30-45° and rotated so that the costal margins are dorsal (Fig. 1C). Female wing extension occurs concurrently with the initiation of mounting activity by the male and appears to be the result of action by the male's prothoracic legs. Although Tobin and Stoffolano (1973a) stated that it is difficult to ascertain whether the female house fly extends her wings or whether they are spread by the male's metathoracic legs, these authors concluded that the male appeared to push them apart as he moved forward along the body of the female. However, Colwell and Shorey (1975) observed that female house fly wing extension occurred voluntarily without pressure being exerted by the male's legs. Wing extension apparently is not an element of face fly courtship behavior (Tobin and Stoffolano 1973b). The wings of the female horn fly remain extended throughout the duration of courtship, but they usually revert to their normal resting position as copulation commences.

Mounting (Fig. 1D) consists of a short "hop" onto the dorsum of the female. Initially, the forelegs, midlegs and hindlegs of the male contact the notum, costal margins of the wing bases, and anterior abdominal tergites of the female, respectively. Unlike house fly courtship behavior (Murvosh et al. 1964, Tobin and Stoffolano 1973a, Colwell and Shorey 1975), lifting of the female's metathoracic legs subsequent to mounting does not occur during horn fly mating. Leg lifting is also absent during face fly (Tobin and Stoffolano 1973b) and stable fly (Anderson 1978) courtship. As mounting is initiated, the male horn fly also commences to rapidly vibrate his wings. Wing vibration continues until just prior to genital orientation and contact.

As the male proceeds to move anteriorly on the dorsum of the female, the posterior end of his abdomen becomes elevated ca. 45° to the body of the female and his metathoracic legs partially encircle her abdomen. At this point, the head of the male is above and slightly anterior to that of the female. The male's prothoracic tarsi briefly contact the female's head and are then extended to contact and encircle the prothoracic tarsi of the female, after which he proceeds to alternately lift her forelegs at a rapid rate (Fig. 1E). Similar activity, termed "boxing," also has been described for the house fly (Tobin and Stoffolano 1973a, Colwell and Shorey 1975) and face fly (Tobin and Stoffolano 1973b).

Following manipulation of the female's prothoracic tarsi, the male horn fly begins moving to the rear (Fig. 1F) by simultaneously returning his prothoracic legs to the female's notum while alternately sliding his metathoracic legs posteriorly along the sides of her abdomen. The male's mesothoracic legs remain on or near the costal margins of the wing bases of the female. The male's claspers are usually extended during this movement as also reported by Tobin and Stoffolano (1973a) and Colwell and Shorey (1975) for the house fly and Tobin and Stoffolano (1973b) for the face fly. As the male continues his rearward and downward movements, his genitalia eventually become oriented below the posterior portion of the female's abdomen and counterposed with her genitalia (Fig. 1G). The male's wings cease vibration at this stage and he attempts to grasp the partially protruded ovipositor of a receptive female with his claspers.

Genital contact (Fig. 1H) is effected as the male grasps the female's ovipositor which he subsequently extends, presumably to clear a passage for spermatophore transfer (Murvosh et al. 1964), by elevating his abdomen. The male then lowers his abdomen and the copulatory position (Fig. 1I) is assumed. The extended wings of the female may return to their usual resting position over the abdomen during this stage. Similar to the house fly (Tobin and Stoffolano 1973a, Colwell and Shorey 1975) and face fly (Tobin and Stoffolano 1973b), the prothoracic legs of the male horn fly contact the female's thorax at or near the wing bases and the metathoracic legs are usually crossed under her abdomen.

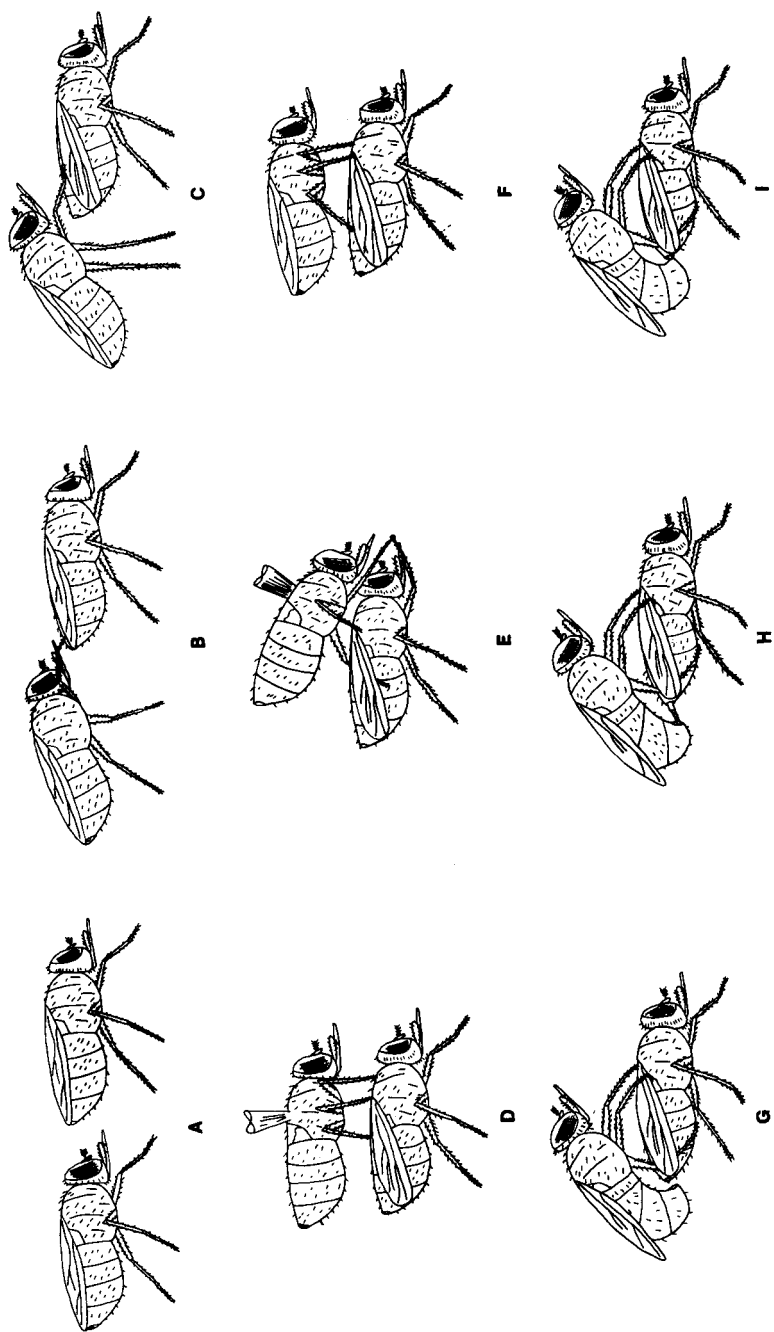


FIG. 1. Elements of horn fly courtship: A. orientation; B. abdominal tapping; C. wing extension; D. mounting; E. protarsal manipulation; F. backing; G. genital orientation; H. genital contact; I. copulation.

The male horn fly's mesothoracic tarsi partially encircle the costal margins of the female's wings. This finding is in agreement with studies by Murvosh et al. (1964) and Colwell and Shorey (1975) who also report that the mesothoracic legs of the male house fly are usually over the female's wings. Tobin and Stoffolano (1973a, 1973b), however, observed the male's mesothoracic legs to contact the wing bases of both the house fly and face fly female. Copulating flies normally remain quiescent unless disturbed. Movement by the female did not appear to result in premature termination of copulation by the male; however, males did occasionally become partially dislodged necessitating reorientation of the stereotyped copulation posture.

Although the male horn fly performs the active role in courtship, the acceptance of genital contact and subsequent successful copulation is contingent upon protrusion of the ovipositor by the female. As with the house fly and face fly (Tobin and Stoffolano 1973a, 1973b, Colwell and Shorey 1975), male horn flies are apparently unable to grasp the female's genitalia unless the ovipositor is at least partially extended. Other female rejection responses include kicking at and/or movement away from the male during prothoracic tapping, refusing wing extension prior to mounting, and lowering of the abdomen to the substrate during genital orientation. In addition, females were observed to attempt rejection at any stage during courtship by violent body shaking and leg kicking.

In summary, horn fly courtship consists of a continuous, and frequently overlapping, series of action patterns initiated primarily by the male. Such behavior is generally similar to that of the house fly, face fly and stable fly; major differences include tapping of the female's abdomen by the male's prothoracic legs immediately prior to mounting and the absence of female metathoracic leg lift following wing extension.

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FENVALERATE AND STIROFOS EAR TAGS FOR CONTROL
OF HORN FLIES ON RANGE CATTLE^{1/2/}

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ABSTRACT

Either 6 or 8% fenvalerate (cyano(3-phenoxyphenyl)methyl 4-chloro- α -(1-methylethyl)benzeneacetate) ear tags, when applied at rates of 1 or 2 tags/animal, controlled horn fly, *Haematobia irritans* (L.), infestations on cattle in central Texas for as long as 5 months. Stirofos (2-chloro-1-(2,4,5-trichlorophenyl)vinyl dimethyl phosphate) ear tags, when applied at the rate of 5 or 6 tags/10 animals, maintained populations below 200 flies/head for 6 wk. Loss rates for tags were very low even though animals were in heavy brush pastures.

INTRODUCTION

Ear tags impregnated with stirofos (Rabon[®], 2-chloro-1-(2,4,5-trichlorophenyl)vinyl dimethyl phosphate) protected cattle from infestations of Gulf Coast ticks, *Amblyomma maculatum* Koch, (Gladney 1976) and also controlled horn flies, *Haematobia irritans* (L.) (Ahrens 1977). Wilson et al. (1978) and Huston et al. (1979) reported that stirofos tags in each ear of range cattle reduced the horn fly population to < 100/head for 60-100 days, and 1 tag/cow or 1 tag/2 cows controlled the flies for less time. Ear tags with the synthetic pyrethroid fenvalerate (SD-43775, cyano(3-phenoxyphenyl)methyl 4-chloro- α -(1-methylethyl)benzeneacetate) gave > 95% control of horn flies for 12 wk (Gerhardt and Mullens 1978). Ahrens and Cocke (1979) reported that an 8% fenvalerate tag in each ear of the cow provided 100% control of horn flies for 20 wk.

Preliminary studies (J. A. Miller and S. E. Kunz, Kerrville, TX; unpublished information) indicate that when an animal is taken from a horn fly-infested herd, has the flies removed from it, and is then returned to the infested herd, it will quickly regain a population of flies comparable to the previous level. This suggests that horn flies move readily among animals within a herd and that populations of the pest might be suppressed without treating each animal in a herd. Since researchers had previously reported that either two 13.7% stirofos or two 8% fenvalerate tags/animal controlled horn flies, we conducted this study to determine the duration of horn fly control obtained from 1 or 2 tags/cow with 6% and 8% fenvalerate ear tags, and to determine the fly control obtained with various regimens of stirofos ear tag treatments.

1/ Diptera: Muscidae.

2/ This paper reports the results of research only. Mention of a pesticide does not constitute a recommendation for use by the U.S. Department of Agriculture, nor does it imply registration under FIFRA as amended. Also, mention of a proprietary product does not constitute an endorsement by the USDA.

METHODS AND MATERIALS

Gillespie County. Cooperator-owned Hereford or Angus cattle from Gillespie County, TX, were used for studies of the fenvalerate ear tags. The cows either had young calves or were calving, but the calves were not treated. The fenvalerate tags were 10-g Allflex®-type tags (Shell Development Company, Modesto, CA) containing either 6 or 8% AI; 20-g 13.7% stirofos tags were also used for additional comparisons of treatment effectiveness.

We determined control of horn flies by making estimates of flies on both sides of the cattle at ca. 2-wk intervals with 10 or more cattle checked for each observation. These checks were made at 1300-1500 h, which allowed time for killing of flies that emerged the previous evening and night.

On May 8, 1979, we put one 6% fenvalerate tag in each ear of 14 cows and 1 bull (Herd A). On May 15, 1979, we put one 8% fenvalerate tag in each ear of 14 cows (Herd B), one 8% fenvalerate tag on each of 30 cows (Herd C), and one 13.7% stirofos tag on each of 29 Brangus cows (Herd D). Cattle in herds B, C, and D were given either pour-on or spot-on treatments for grubs either 1 wk before or at tagging time. On May 21, we tagged 14 cows with one 8% fenvalerate tag/cow (Herd E) and 13 mixed-breed cows and heifers and 1 bull with one 6% fenvalerate tag/animal (Herd F).

Bexar County. On May 9, 2 herds (1 Hereford, 1 Angus) of 30 cows each were initially treated at the rate of one 13.7% stirofos tag/2 cows; no calves were treated. Plans called for application of tags to an additional 10% (3 animals) of the herd at 2-wk intervals until fly populations were essentially eliminated from the animals. Both herds were maintained in ca. 648 ha of heavy cedar and oak brush pastureland. Animals, in response to the sound of truck horns, were gathered once a wk at various sites throughout the respective pastures for observation. Fly counts were recorded on a per-animal basis.

In the 2nd test of this study, on August 15 the 2 herds were combined in 1 pasture, the old tags removed, and the cows retreated. Again, half of the Hereford and half of the Angus cows were treated with 1 ear tag each, but no additional tags were applied. The data resulting from this test and data from the 1st test when 60% of the animals had ear tags were compared for statistical significance with the t-test.

Kerr County. Three herds of Santa Gertrudis cows (50-75 each) were used in these studies. Half of each of herds A and B (50 cows each) were treated in June with 1 stirofos ear tag each (1 tag/2 cows). Herd C was treated at the same rate, but a 2nd group of 25 untreated animals was added to this herd a few days after treatment. Fly counts were obtained weekly following above procedures.

The control herds for all the tests contained various numbers of untreated cattle 2 miles or less from the test herds.

RESULTS AND DISCUSSION

Gillespie Study. During the 1st 8-wk posttreatment, the average number of horn flies/animal with all fenvalerate tags was 0-2; however, cattle with stirofos tags averaged 10-25 horn flies each. Untreated cattle nearby each carried from 100 to 1000 horn flies. The average numbers of horn flies/animal from days 56 to 169 posttreatment are shown in Table 1.

In Herd A, 6% fenvalerate tags (2/cow) gave excellent control for 18 wk. At 20 wk posttreatment, though it was mid-September and very dry, the horn flies had increased from 23 to 200/head; horn fly populations on untreated cattle in the area increased to > 1000/head.

Horn fly control was excellent in Herd B with two 8% fenvalerate tags/animal throughout the test (23 wk). In fact, horn flies were still being killed when we terminated the study due to cool weather and the decline of horn fly populations during the 4th wk of October.

TABLE 1. Average Number of Horn Flies/Cow After Treatment with Fenvalerate- or Stirofos-Impregnated Ear Tags, Gillespie County, TX, 1979.

Days	Number of tags and treatment							
	Fenvalerate						One stirofos	Untreated
	posttreatment	Two 6%	Two 8%	One 8%	One 6%			
	Herd	A	B	C	E	F	D	
0		600	--	--	> 300	> 500	> 300	
56		0	< 1	> 1			26	> 500
57			3	53				> 200
65					0	1		250
71			< 1	30			37	
73		0						
78					< 1 ^{a/}	< 1 ^{a/}		
84			> 1	80			145	1000
85		0			0 ^{a/}	0 ^{a/}		
91			0	50			150	
92					3 ^{a/}	3 ^{a/}		
98			0	300			300	
99		< 1						1000
106					25 ^{a/}	25 ^{a/}		
112			13	238			440	
114		32						750
120					43 ^{a/}	43 ^{a/}		
126			14	205			450	
127		23						> 700
141		200			43	100		1000
147			8	147				1000
159		150			53	50		
160			2	25				
169		100						> 400

a/ Herds were together 8/7-9/18.

Horn fly control was less with the use of one 8% fenvalerate tag/cow in Herd C than either in Herd E with one 8% tag or in Herd F with one 6% tag. However, several untreated cattle were put in the pasture with Herd C for purpose of herd management, and on the adjoining ranch upwind, cattle had 1000 horn flies/head. Also due to the cooperator's grazing management practices, Herds E and F were in the same pasture from August 7 to September 18, but control of horn flies due to the combined treatment of these 2 herds resulted in fewer than 50 flies/head. The single stirofos tag treatment gave good control (< 200 flies/head) for ca. 13 wk. Retention of the tags was very good. Of the 116 fenvalerate tags used, 2 were lost because they separated from the base (poor application) and 1 was torn from the ear.

The test was terminated after 169 days due to cold weather which resulted in reduction of horn fly populations; therefore, actual residual effectiveness was not established. One tag/animal also provided good control with generally < 50 flies/head for this study period. Thus, these data indicate that a single tag could provide adequate season long control of horn flies in central Texas.

Bexar and Kerr County Trials. These trials were not intended to determine the effectiveness of stirofos ear tags per se but to determine the minimum number of animals in a herd that would require treatment to achieve acceptable levels (200 flies/head) of fly control. Fly counts resulting from treatment of cattle in Bexar County are shown in Table 2. Control was significantly greater, according to the t-test, in the herd with 60% of the animals tagged than in the herd with only 50% of the animals tagged; however, even with 50% treated animals, populations generally remained < 200 flies/head. Pretreatment fly levels of 700-800

TABLE 2. Average Number of Horn Flies/Cow After Treatment with 1 Stirofos Ear Tag/2 Cows, Bexar County, TX, 1979.

Wk posttreatment	Herd no. 1	% reduction	Herd no. 2	% reduction	Control herd
<u>Test no. 1</u>					
0	800	--	700	--	1000
1	200	80	200	80	1000
2 ^{a/}	100	90	100	90	1000
3	30	96	20	98	750
4	40	95	30	97	875
5	5	99	20	97	650
6	10	98	5	99	575
7	15	98	15	98	650
8	45	92	25	96	575
9	80	--	45	--	--
10	75	80	45	87	350
11	150	58	100	72	350
<u>Test no. 2</u>					
0	200	--			300
1	165	66			475
2	165	78			750
3	200	72			700
4	190	70			625
5	150	77			650
6	100	82			550
7	150	77			650
8	300	15			350
9	200	20			250

a/ Three additional cows (10%) were tagged after this observation only.

flies/head were quickly reduced to 200/head after the initial treatment. Following the added treatment of 3 animals/herd, counts dropped to < 50/head and consistently remained there for 55-59 days. Counts remained at 200 flies/head or less for 12 wk, but untreated infestations also declined. Following treatment for the 2nd test when the herds were combined, acceptable levels of control were maintained for as long as 47 days. The reduced control level for this test may have been a result of infestation pressure normally arising during this period (late August and September), as shown by the numbers of flies on the control herd (Table 2). This fluctuation in population, along with the decline noted above, occurs naturally (Kunz and Cunningham 1977).

Pretreatment populations on the Kerr County herds averaged 200 flies/head. (These animals had been exposed to dust bag treatments until 3 wk prior to the ear tag treatments.) Fly populations differed somewhat from herd to herd, but remained at 100 flies/head or less for as long as 7 wk in Herds A and B (Table 3) following treatment. The animals in Herds A and B were compatible, but the 2nd group of 25 added to Herd C did not mix with the treated group. These 25 animals remained essentially untreated and carried 400-600 flies/head (Table 3).

The studies in Bexar and Kerr Counties further demonstrated that, because of the significant movement of flies between animals in common enclosures, treatment of all animals will not be necessary to reduce horn fly populations below generally accepted economic levels (200/head for southwestern U.S.A.). With the newer synthetic pyrethroids, it may be possible to limit the treatment level to 30 or 40% of the herd, thereby reducing the treatment expense, yet maintaining adequate control. Properly defined economic threshold levels of horn flies for the different cattle production areas will ultimately determine the number of animals requiring treatment.

TABLE 3. Average Number of Horn Flies/Cow After Treatment with 1 Stirofos Tag/2 Cows, Kerr County, TX 1979.^{a/}

Date	Wk posttreatment	Herds			b/c/
		A	B	C	
6/22	0 + 3 days	200	200	200	
	1	75	25	25	
	2	--	--	--	
7/2	2	25	25	25	
7/13	3	75	25		
7/20	4	100	25	25 (400)	
7/27	5	75	25	25	
7/30				200 (400)	
8/1	6			75 (300)	
8/6		250	25	50 (400)	
8/10		200	50	200 (600)	
8/21	9	500	700	End (1000+)	
9/14	12	750	1000		

a/ All cattle were subjected to dust bag treatments until 3 wk prior to start of ear tag study.

b/ Herd C with 25 treated and 50 untreated. Second 25 head untreated herd remained totally isolated from remainder of animals. The fly population on these animals was essentially untreated (in parenthesis).

c/ Counts on untreated control animals ranged from 400 to 700 flies/head during test.

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CONTROL OF FACE FLY^{1/} LARVAL DEVELOPMENT WITH
THE IVERMECTIN, MK-933^{2/}

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ABSTRACT

MK-933 injected into cattle at 200 µg/kg body weight resulted in 100% corrected mortality of face fly, *Musca autumnalis* DeGeer, larvae developing in the feces for 9 days. Larvae emerging from feces sampled 10-15 days post-treatment developed into malformed pupae, with ca. 90% failing to undergo eclosion. Effectiveness of MK-933 decreased after 15 days posttreatment.

INTRODUCTION

MK-933 (ivermectin) represents a synthetic derivative of the antiparasitic avermectins and exhibits activity against a wide variety of metazoan pests. Burg et al. (1979) and Egerton et al. (1979) have reported on the success of the avermectin complex as an anthelmintic. Egerton et al. (1980) and Wilkins et al. (1980) demonstrated significant systemic potential of MK-933 against various ectoparasitic arthropods. The documented systemic activity of MK-933 prompted an investigation into its potential for controlling larval development of the face fly, *Musca autumnalis* DeGeer. The face fly has become one of the foremost livestock pests in North America, primarily because no adequate control measure has been developed.

This study was conducted to evaluate the degree and duration of activity of an injectable formulation of MK-933 against the face fly.

MATERIALS AND METHODS

At the Merck Research Farm, Springdale, AR, Holstein bull calves, weighing 115-158 kg each, were assigned to 3 treatment groups of 3 animals each. The treatment groups consisted of MK-933 plus solvent (glycerol formal and propylene glycol, 40:60 v/v), solvent alone, and an untreated control. MK-933 and the solvent were injected subcutaneously at 200 µg/kg and 0.022 ml/kg body weight (BW), respectively.

Feces were taken rectally from each animal daily for 24 days. Each sample was subdivided 3 times and bioassayed with 10, 1st-instar face fly larvae obtained from a laboratory culture. The samples were maintained under room conditions until after pupation occurred, after which time the pupae were counted. Abbott's formula was used to determine the % corrected mortality (CM) of face fly larvae for each treatment, based on the average of the 3 subsamples.

^{1/}Diptera: Muscidae.

^{2/}Published with the approval of the Arkansas Agricultural Experiment Station.

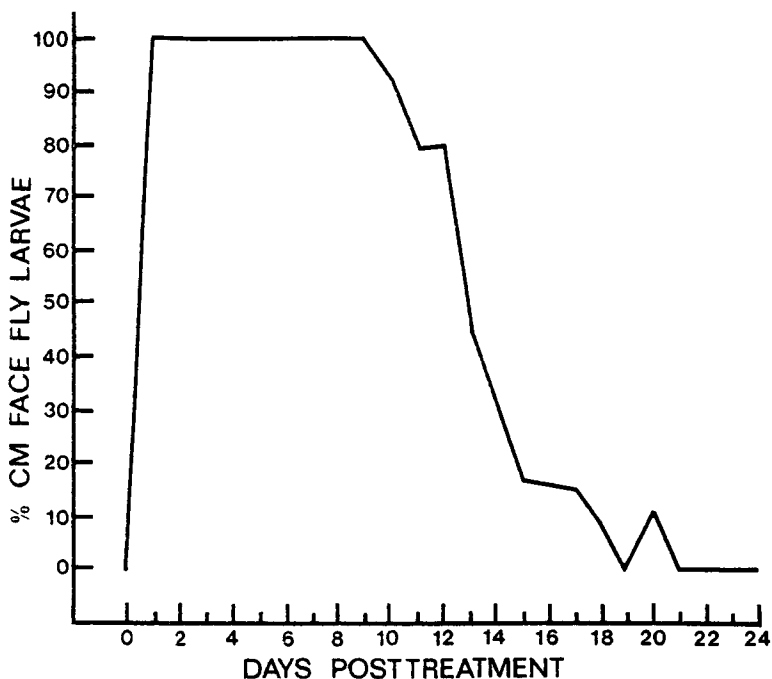


FIG. 1. Percent corrected mortality (CM) of face fly larvae occurring in feces from cattle injected with MK-933 (average of 3 replicates).

RESULTS AND DISCUSSION

MK-933 gave total control of face fly larvae for 9 days posttreatment, as shown in Fig. 1. Increasing numbers of larvae pupated successfully from feces sampled 10-14 days posttreatment, however the resulting pupae were malformed (Fig. 2) and only ca. 10% developed to the adult stage. After day 14, the numbers of normal pupae tended to increase progressively. There was no mortality of face fly larvae as a result of the solvent treatment. The occurrence of a dry, compact fecal sample was probably responsible for the insignificant mortality in the day 20 sample (Fig. 1).

MK-933 gave essentially 14 days of 100% control of face fly larvae. This sustained activity would make MK-933 very beneficial in suppressing early spring development of populations of the face fly. Many integrated livestock pest management programs could be enhanced through the effectiveness and ease of treatment offered by MK-933, if either dosage or formulation could be modified to increase the duration of the activity.

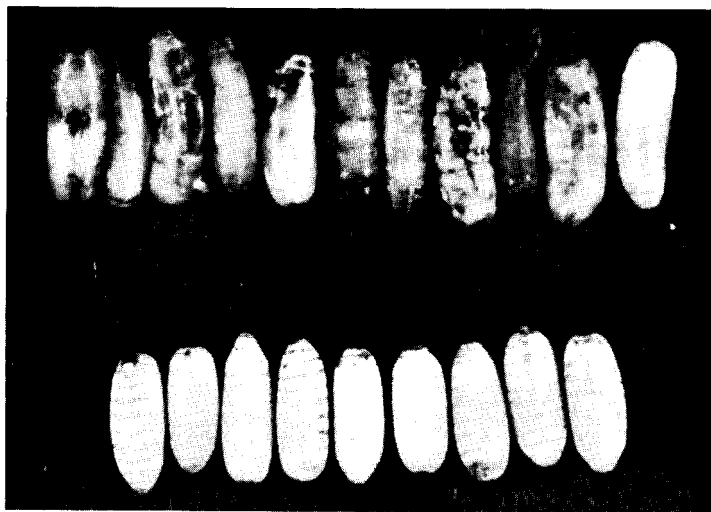


FIG. 2. Examples of malformed face fly pupae that developed in cattle feces samples 10-14 days posttreatment with MK-933 (top row-treated, bottom row-control).

ACKNOWLEDGMENT

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EXHAUST FUME ANESTHETIZER FOR USE WITH WIND-ORIENTED SCREWORM TRAPS^{1/}F. H. Tannahill^{3/}, J. Wendell Snow^{4/}, and J. R. Coppedge^{3/}Screwworm Research, AR, SEA, USDA, P. O. Box 986
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ABSTRACT

A new method of anesthetizing screwworm flies, Cochliomyia hominivorax (Coquerel), captured in wind-oriented traps was developed that uses exhaust fumes from a vehicle. Results of tests using this procedure showed that the mean times for anesthetization at wind speeds of 30.5, 91.4, and 152.4 m/min were 14.2, 22.1, and 46.3 sec, respectively. Anesthetization with this system is rapid, and the condition of the tested flies is such that they can be easily identified and the females dissected.

RESUMEN

Se desarrolló un método nuevo para anestesiar moscas del gusano barrenador del ganado capturadas en trampas orientadas por el viento. Este método utiliza el humo del escape de un vehículo. Pruebas utilizando este procedimiento mostraron que el tiempo promedio para anestesiar las moscas en vientos de 30.5, 91.4, y 152.4 metros por minuto fueron 14.2, 22.1, y 46.3 segundos, respectivamente. Este sistema es rápido y la condición de las moscas después de anestesiadas es tal que pueden ser identificadas y las hembras disectadas.

INTRODUCTION

Swormlure-2 is currently the standard attractant for screwworm Cochliomyia hominivorax (Coquerel) adults (Coppedge et al. 1977). This chemical mixture has proven to be an effective attractant for the screwworm (Coppedge et al. 1977), especially when used in conjunction with wind-oriented (WO) traps (Broce et al. 1977). Currently, WO traps baited with swormlure-2 are widely used in the Southwestern United States and Mexico for detection of adult screwworms in the eradication area. Because of the extensive use of swormlure-2 baited traps, a need has arisen for an efficient method of anesthetizing the captured flies so they can be easily removed from the trap and transported to the laboratory. Ideally, anesthetization procedure should give a rapid knockdown of captured flies (to save servicing time at each trap), should not leave a toxic residue in the trap (so that captured flies would remain alive in the trap as long as possible to prevent dessication), and should not be hazardous to human health.

In the past, benzene and pyrethrin insecticides (aerosol spray 0.1% AI) have been used to kill flies captured in the traps. However, these methods of killing flies have certain problems: benzene may be hazardous to human health, and a residue of pyrethrins that is toxic, and perhaps also repellent to screwworm flies, accumulates in the traps after several applications.

^{1/} Diptera: Calliphoridae.

^{2/} Mention of a commercial or a proprietary product in this paper does not constitute an endorsement of this product by the USDA.

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In our field research activities, we found that when a WO trap was placed directly under the exhaust pipe of a vehicle, live flies captured in the trap were rapidly anesthetized. Research was therefore initiated to investigate the effectiveness of a system using vehicle exhaust fumes. The results of this investigation are reported herein.

METHODS AND MATERIALS

The exhaust fume anesthetizer (EFA) system is constructed from a 7.6-L galvanized bucket (Fig. 1). The extension tube from the bucket to the exhaust pipe is a 17.8-cm length section of 20 cm X 7.6-cm-diam galvanized air conditioning duct that is formed and soldered to an 8.9 X 6.4-cm hole in the bottom portion of the bucket.

The tests were conducted at the Screwworm Eradication Program facility at Moore Air Base in Mission, TX. Packaged sterile flies were obtained from the fly packaging facilities, and taken to our laboratory where they were immobilized by chilling. Approximately 100 flies/replicate were removed from the boxes and placed in a WO trap where they were allowed to recover. Next, the WO trap bait holder and counterbalance weight were removed, and then the bucket and cone were placed in the EFA and carried to the vehicle. The flies were exposed to the vehicle exhaust until they were immobilized; recovery data were obtained by observing the numbers that were alive in the traps at intervals of 15 min, 1 h, 4 h, and 24 h posttreatment.

Each treatment was replicated 10 times, and 1 control treatment was conducted for each series of 10 replicates. Ambient temperature during testing ranged from 21.1^o to 26.7^o C. Wind speed was measured with an air velocity indicator and recorded for each replication (an average wind speed was determined at the end of each series of tests).

RESULTS AND DISCUSSION

The time required to immobilize flies varied proportionately with wind speed (Table 1). The vehicle in the 3rd test was not allowed to heat to the normal operating temperature before exposure of the flies; therefore, a longer anesthetization time was required. Percentage of recovery was low in all 3 tests.

TABLE 1. Mean Anesthetization and Recovery Time for Adult Screwworm Flies Exposed in the EFA.

Treatment	No. of reps.	\bar{x} Wind speed (m/min)	\bar{x} Anesth time (sec)	\bar{x} No. flies/trap	\bar{x} Recovery at indicated time postexposure			
					15 min	1 h	4 h	24 h
Control 1	10	30.5	14.2	99	100	100	100	98
1	10			92	18.2	21.3	25.4	34.0
Control 2	10	91.4	22.1	100	100	100	100	100
2	10			97	24.8	27.0	30.5	42.8
Control 3	10	152.4	46.3 ^{a/}	112	100	100	100	100
3	10			97	21.6	26.2	30.8	38.2

^{a/}Test conducted before vehicle engine had heated to normal operating temperature.

Heat emitted from the exhaust does not appear to be a problem with this system because of the short length of time required to anesthetize the flies. Even after 46 sec., the bucket can still be hand-held without discomfort to the user. The design of this system is such that the exhaust fumes can be directed away from the user by holding the bucket away from the face, allowing wind movement to carry off the exhaust fumes.

Discoloration of the flies was not notable and all external identification characteristics could be seen after exposure to the exhaust fumes. Ovaries of

dissected female screwworms were still easily identifiable, and distinction could be made between native and released flies when the system was used in the field.

Results from this test showed that the EFA system was effective for quick anesthetization of screwworm flies captured in WO traps. However, the fumes are fairly toxic (maximum recovery of only 42.8%) and other means such as anesthetization with CO_2 or chilling will be required for taking samples of live flies.

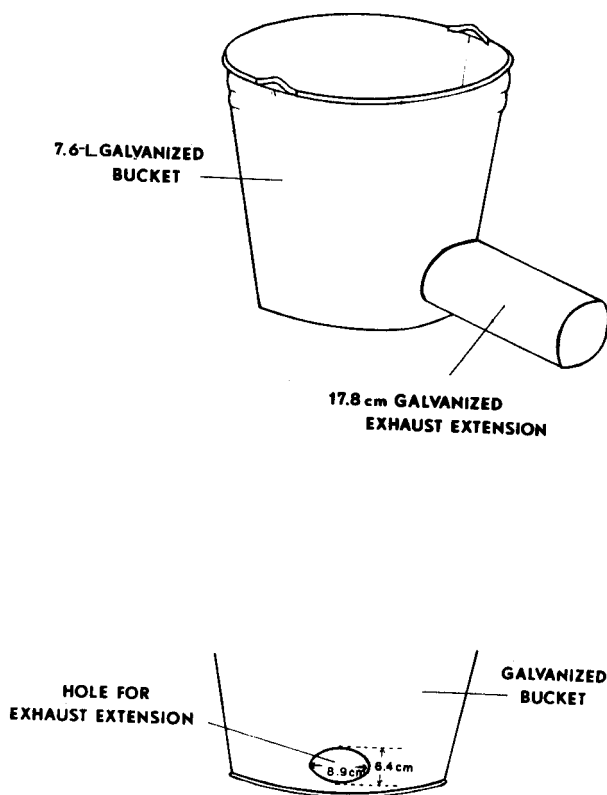


FIG. 1. Schematic drawing of exhaust fume anesthetizer (EFA) system.

ACKNOWLEDGMENT

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TICKS COLLECTED FROM SMALL AND MEDIUM-SIZED
WILDLIFE HOSTS IN LEFLORE COUNTY, OKLAHOMAH. G. Koch and J. E. Dunn^{1/}

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ABSTRACT

A total of 32 of 67 species of wild birds and mammals sampled from April to September in 1978 and 1979 in LeFlore County, OK, were positive for ticks. About 75% of the ticks collected were the lone star tick, Amblyomma americanum (L.). All stages of this tick were common on coyotes. Ground-feeding birds had more immature lone star ticks than other bird species.

The remaining species, collected in decreasing order of abundance, were Haemaphysalis leporispalustris (Packard), Dermacentor variabilis Say, Ixodes scapularis Say, I. texanus Banks, I. cookei Packard, I. woodi Bishopp, I. dentatus Marx, Ornithodoros hermsi Wheeler, Herms, and Meyer, and Rhipicephalus sanguineus (Latreille). Ixodes woodi and O. hermsi have not previously been reported in Oklahoma.

INTRODUCTION

Ticks comprise a relatively small group of 3 families and only about 800 species are distributed worldwide (Hoogstraal 1978). Most of the 81 species that occur in the USA are uncommon (Strickland et al. 1976). The lone star tick, Amblyomma americanum (L.), is the predominant tick species in the Ozark region (Tugwell and Lancaster 1962, Clymer et al. 1970, Cooney and Burgdorfer 1974) and throughout much of southeastern and southcentral USA (Cooley and Kohls 1944, Bishopp and Trembly 1945, Sonenshine et al. 1965). The role of specific wildlife in supporting populations of this species is difficult to evaluate due to its wide host range and relative abundance in relation to habitat type (Lancaster 1957, Sementner et al. 1971, Sonenshine and Levy 1971).

The purpose of the present study was to determine the distribution of different species of ticks, particularly lone star ticks, present on small and medium-sized birds and mammals in 2 areas in the Ozark region of Oklahoma as a basis for studying lone star tick population dynamics. Seasonal populations of the free-living stages of the lone star tick were monitored with Dry Ice® sampling and drags to determine which ticks were available to hosts each month during the studies.

MATERIALS AND METHODS

Collections of animals were made from April to October during 1978 and 1979 primarily from 2 areas in LeFlore County, OK. One area, the Wister Public Hunting Area, encompasses several thousand unimproved acres of mixed hardwoods and meadows along the Fourche Maline River. The other area, the Robert S. Kerr Ranch, encompasses several thousand acres but is mostly improved pastureland along the Poteau River. According to preliminary surveys with dragging and dry ice sampling methods for free-living ticks, these areas

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had moderate tick populations (Gladney 1978) for wooded oak-hickory habitats as compared to the heavy densities obtained by Semtner and Hair (1973a). Additional host collections were made throughout LeFlore County when animals were available.

Birds and mammals were collected with live traps, steel traps, mist nets, and shotgun; road kills were also collected. Steel traps were checked daily to prevent host mortality. Live catches were either anesthetized and examined for ticks or held over trays for collection of detached ticks. Dead hosts were placed in plastic bags and examined for ticks as soon as possible. All ticks were stored in 70% isopropyl alcohol for future identification.

The seasonal activity of the lone star tick was monitored in both study areas with dry ice and cotton flannel cloth drag sampling methods at semi-monthly intervals throughout the seasons. The dry ice trap consisted of a 227-g cube of dry ice placed centrally on a 0.7 x 0.9-m white cloth and left for 1 h. Ticks were then counted and returned to the area. Drags were made by pulling a white flannel cloth (0.9 x 1.4 m) across low vegetation for 50 m and counting ticks clinging to the drag. A total of 49 dry ice samples and 29 drag samples were monitored regularly for ticks over permanent sites.

RESULTS AND DISCUSSION

A total of 7,145 ticks were collected from 32 of 67 bird and mammal species^{2/} samples. All mammalian species sampled except the Armadillo (2 collected) and Eastern Mole (1 collected) had ticks. No ticks were found on the following bird species (numbers in parentheses are the number of each species collected): American Goldfinch (1), Barn Swallow (2), Black-and-white Warbler (1), Blue-gray Gnatcatcher (4), Blue Grosbeak (1), Brewer's Blackbird (1), Brown Thrasher (1), Carolina Chickadee (1), Common Crow (3), Downy Woodpecker (3), Eastern Kingbird (4), Eastern Wood Pewee (1), Field Sparrow (7), Great Blue Heron (1), Great Crested Flycatcher (2), Green Heron (2), Lark Sparrow (1), Mourning Dove (4), Nashville Warbler (1), Orchard Oriole (2), Philadelphia Vireo (1), Red-bellied Woodpecker (1), Summer Tanager (2), Swainson's Thrush (1), Trail Flycatcher (1), Tufted Titmouse (4), White-crowned Sparrow (3), Wilson's Warbler (1), Woodthrush (1), Yellow-breasted Chat (2), and Yellow-throat (1).

Lone Star Tick. The lone star tick (Table 1) was the most prevalent species, comprising 74.5% of the collections. Host collections were divided at June 19 to June 20 in Table 1 due to the absence of larvae sampled early in the season (Table 2). This seasonal bias may help explain why some hosts collected only in the 1st part of the season had no larvae.

Of the hosts surveyed, the coyote was the most relatively important host of adult ticks, followed by the domestic dog and raccoon, according to the number of ticks per host. Because they are not considered wildlife, few dogs were examined, although many roam freely and are often heavily parasitized by ticks. The red fox and gray fox were the most relatively important hosts of adults in studies in Arkansas (Tugwell and Lancaster 1962) and Virginia (Sonenshine and Stout 1971), but in eastern Oklahoma (Clymer et al. 1970) and in Tennessee (Cooney and Burgdorfer 1974), deer were very important. No attempts were made to sample these hosts in this study. No adult ticks were found on any of the birds sampled.

2/ Common names of birds adopted by the American Ornithologists' Union (Check list of North American Birds, 5th ed. Baltimore, MD; American Ornithologists' Union, 1957) and common names of mammals adopted by Miller, G. S., Jr. and R. Kellogg (List of North American recent mammals, U.S. Nat. Mus. Bull. 205, 1955) were used in this study.

TABLE 1. Host List of *Amblyomma americanum* (L.) Collected April Through September 1978 and 1979 in LeFlore County, OK.

Host	No. hosts examined a/		No. hosts with ticks	No. of			
	4/1-6/19	6/20-9/30		Larvae	Nymphs	Females	Males
Coyote	0	6	4	108	115	72	110
Raccoon	2	15	17	2648	249	2	3
Striped Skunk	1	3	1	1	51	---	---
Domestic Dog	1	1	2	----	---	8	3
Woodchuck	0	2	2	21	9	---	---
Fox Squirrel	11	13	16	896	133	---	---
Cottontail Rabbit	25	29	34	364	326	---	---
Gray Squirrel	4	0	4	----	18	---	---
Opossum	26	14	2	3	3	1	---
Swamp Rabbit	0	2	1	1	---	---	---
Domestic Cat	1	0	1	----	7	---	---
Eastern Woodrat	50	11	5	6	2	---	---
Deer Mouse	57	17	1	1	---	---	---
Wild Turkey	4	0	1	----	60	---	---
Roadrunner	1	2	3	43	23	---	---
Eastern Meadowlark	1	9	3	10	2	---	---
Bobwhite	4	3	3	4	2	---	---
Blue Jay	2	1	2	1	1	---	---
No. Cardinal	4	3	3	1	3	---	---
White-throated Sparrow	9	0	3	1	2	---	---
Vesper Sparrow	0	1	1	----	4	---	---
Turkey Vulture	1	1	1	----	1	---	---
Brown-headed Cowbird	6	1	1	----	1	---	---
Catbird	0	2	1	----	1	---	---
Totals	210	136	112	4109	1013	83	11

a/ Larvae did not appear in significant numbers in the field until mid-June; thus, hosts collected early in the season did not have larvae attached.

The raccoon, fox squirrel, cottontail rabbit, and coyote were the hosts parasitized most by immatures. Foxes, raccoons, woodchucks, and skunks were important hosts of immatures, as reported by Tugwell and Lancaster (1962), Clymer et al. (1970), Sonenshine and Stout (1971), and Cooney and Burgdorfer (1974). Patrick and Hair (1977) reported that white-tailed deer were also important hosts of immatures. Probably because of a greater probability of an encounter, ground-feeding birds generally had more ticks than did tree-dwelling species. Our data relative to the incidence of lone star tick immatures on birds agrees with the results reported by Lancaster (1973) for Arkansas, but not with those of Sonenshine and Stout (1970) for Virginia and North Carolina. The latter authors found only 1 of 10,700 birds parasitized by this tick species.

Infestations of ticks on hosts (Table 1) correspond to off-host sampling (Table 2). No larvae were found on hosts before June 19 except for a single larva from a White-throated Sparrow examined on May 5, 1979. Free-living populations of nymphs and adults were present throughout most of the period over which the host survey was conducted and were attracted readily to CO₂. Those ticks ascending vegetation and available for drags and hosts moving through an area were fewer early in the season than later. Fewer nymphs and adults were sampled in 1979 than in 1978, presumably due to a drought affecting larval survival from July to September 1978.

TABLE 2. Numbers of Amblyomma americanum (L.) Sampled With Dry Ice and Dragging During 1978 and 1979 in LeFlore County, OK.

Month	Sample ^{b/}	Average no. of ticks per sample ^{a/}							
		Males		Females		Nymphs		Larvae	
		dry ice ^{c/}	Drag ^{d/}	dry ice	Drag	dry ice	Drag	dry ice	Drag
April	1	2.88	0.03	3.08	0.00	1.21	0.00	0.00	0.00
	2	3.96	0.03	5.59	0.00	5.78	0.07	0.00	0.00
May	1	4.63	0.48	5.63	0.24	7.39	0.03	0.00	0.00
	2	3.92	0.72	5.41	0.45	7.78	0.24	0.00	0.00
June	1	3.33	0.41	4.88	0.24	7.43	0.14	0.00	0.00
	2	2.49	0.28	4.27	0.24	9.04	0.03	3.54	2.55
July	1	1.41	0.28	2.06	0.38	5.73	0.24	0.00	0.00
	2	1.08	0.14	1.76	0.34	4.67	0.62	0.00	8.62
Aug.	1	0.59	0.07	0.59	0.03	3.41	0.62	3.04	3.62
	2	0.16	0.00	0.29	0.00	2.47	0.21	6.04	21.66
Sept.	1	0.04	0.00	0.08	0.00	0.25	0.10	6.46	5.48
	2	0.00	0.00	0.04	0.00	0.00	0.00	3.58	3.28

a/ Results for years are averaged.

b/ 1 sample/half month.

c/ Means of 49 1-h samples.

d/ Means of 29 flannel drags.

Rabbit Tick. The rabbit tick, Haemaphysalis leporispalustris (Packard), comprised 18.8% of the ticks collected but was taken only from rabbits and birds (Table 3). Adult ticks were found only on cottontail rabbits. Other authors (Peters 1936, Eddy 1942, Green et al. 1943, Tugwell and Lancaster 1962) indicated a wide host range for immatures, including small mammals and many birds. Sonenshine and Stout (1970) found rabbits and ground-feeding birds the most significant hosts of immatures in Virginia, but rabbits were the only significant hosts for adults.

TABLE 3. Host List of Haemaphysalis leporispalustris (Packard) Collected April Through September 1978 and 1979 in LeFlore County, OK.

Host	No. hosts examined	No. hosts with ticks	No. of			
			Larvae	Nymphs	Males	Females
Cottontail Rabbit	54	44	505	268	280	99
Swamp Rabbit	2	1	26	12	---	--
Bobwhite	7	5	85	10	---	--
Roadrunner	3	2	24	3	---	--
Lincoln's Sparrow	4	2	4	1	---	--
No. Cardinal	7	1	3	2	---	--
White-throated Sparrow	9	2	2	---	---	--
Savannah Sparrow	4	2	2	---	---	--
Blue Jay	3	1	---	2	---	--
House Wren	2	1	1	2	---	--
Grasshopper Sparrow	2	1	1	---	---	--
Indigo Bunting	6	1	2	---	---	--
Brown-headed Cowbird	7	2	1	3	---	--
Yellow-billed Cuckoo	10	1	0	3	---	--
Totals	120	66	656	306	280	99

American Dog Tick. The American dog tick, Dermacentor variabilis Say, comprised 3.4% of the ticks collected and were recovered only from mammals (Table 4). Adult ticks were found primarily on coyotes, opossums, and raccoons. Immatures were collected primarily from rabbits and rodents. These findings agree with feeding preference studies of Sonenshine and Atwood (1967) and field data reported by Clymer et al. (1970) and Lancaster (1973).

TABLE 4. Host List of Dermacentor variabilis Say Collected April Through September 1978 and 1979 in LeFlore County, OK.

Host	No. hosts examined	No. hosts with ticks	No. of			
			Larvae	Nymphs	Males	Females
Coyote	6	4	--	--	38	12
Opossum	40	27	--	--	54	29
Raccoon	17	11	--	4	18	16
Fox Squirrel	26	5	1	--	4	3
Striped Skunk	4	1	--	1	1	--
Woodchuck	2	1	--	--	1	1
Eastern Woodrat	61	12	9	10	--	--
Cottontail Rabbit	54	5	15	4	--	--
Hispid Cotton Rat	1	1	--	2	--	--
Eastern Harvest Mouse	1	1	1	--	--	--
Domestic Dog	2	1	--	--	--	1
Deer Mouse	74	12	14	6	--	--
Totals	288	81	40	27	116	62

Black-legged Tick. The black-legged tick, Ixodes scapularis Say, was found on 5 mammalian species (Table 5). Few adults were found, since this stage normally attaches to hosts in the fall and winter (Cooley 1945, Harris 1959). These ticks are very common on deer (Clymer et al. 1970).

TABLE 5. Host List of Ixodes scapularis Say Collected April Through September 1978 and 1979 in LeFlore County, OK.

Host	No. hosts examined	No. hosts with ticks	No. of			
			Larvae	Nymphs	Males	Females
Cottontail Rabbit	54	9	25	14	1	--
Eastern Woodrat	61	11	89	22	--	--
Fox Squirrel	26	2	--	3	--	--
Raccoon	17	1	--	--	--	1
Deer Mouse	74	2	--	2	--	--
Totals	232	25	114	41	1	1

Uncommon Ticks. Six additional species of ticks were collected from mammals but were relatively uncommon on the hosts collected (Table 6). The brown dog tick, Rhipicephalus sanguineus (Laterille), is very common on domestic dogs in LeFlore County, and infestations of several hundred ticks of all stages/host are not rare.

TABLE 6. Host List of 6 Species of Ticks (Ixodoidea) Collected April Through September 1978 and 1979 in LeFlore County, OK.

Tick Species	Host Species	No. hosts with ticks		No. of			
		No. hosts examined		Larvae	Nymphs	Male	Female
<u>R. sanguineus</u> (Laterille) Coyote		1/	6	--	2	-	--
<u>I. cookei</u> Packard	Eastern Woodrat	1/	61	--	1	-	--
	Raccoon	2/	17	--	7	2	1
<u>I. dentatus</u> Marx	Eastern Woodrat	1/	61	--	1	-	--
	Cottontail Rabbit	1/	54	1	3	1	--
<u>I. texanus</u> Banks	Raccoon	7/	17	10	24	1	10
<u>I. woodi</u> Bishopp	Eastern Woodrat	2/	61	8	3	-	--
<u>O. hermsi</u> - Wheeler, Herms, Meyer	Woodchuck	1/	2	--	6	-	--

Ixodes cookei Packard, I. dentatus Marx, and I. texanus Banks were reported from Oklahoma previously (Eddy 1938^{3/}, but I. woodi Bishopp and the soft tick (Argasidae) Ornithodoros hermsi Wheeler, Herms, and Meyer are apparently new state records (Pers. Comm. - Don Arnold, State Survey Entomologist, Stillwater, OK).

Some species, such as the Gulf Coast tick, Amblyomma maculatum Koch, are relatively common in some parts of Oklahoma (Semtner and Hair 1973b) but were not found in our studies. Additional collecting, particularly from larger hosts, would probably uncover additional tick species. This study indicated that at least 4 species of ticks of medical and/or veterinary importance are commonly found on wildlife hosts in southeastern Oklahoma.

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COMPARATIVE EFFICACY OF ACARICIDES FOR CONTROL
OF PSOROPTES OVIS (HERING) AND P. CUNICULI (DELAFOND)^{1/2/}

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ABSTRACT

Thirty-seven compounds with acaricidal properties were evaluated to compare their efficacy against an ear canker mite [Psoroptes cuniculi (Delafond)] of rabbits and the common scabies mite of cattle [sheep scab mite, P. ovis (Hering)]. Four types of acaricides were tested: chlorinated hydrocarbons, organophosphates, carbamates and miscellaneous compounds, and tertiary amines. Compounds were rated I to V according to their activity against mites. Two compounds were rated as Class V (dieldrin and endrin) and 11 as Class IV against P. ovis. Five compounds were rated as Class V (dieldrin, endrin, isobenzan, isodrin, and toxaphene) and 7 as Class IV against P. cuniculi. Thirty-two compounds (86.5%) were equally effective against both species of mites or were more effective against P. ovis than P. cuniculi. In 13.5% of the tests, compounds exhibited greater activity against P. cuniculi. Therefore, P. cuniculi is confirmed as an acceptable substitute for P. ovis in the evaluation of candidate acaricides.

INTRODUCTION

One of the primary goals of the research program in the Scabies and Mange Research Unit at the U. S. Livestock Insects Laboratory, Kerrville, TX, is to find new control agents for use in the eradication of psoroptic scabies of cattle in the USA. Psoroptic scabies in cattle, caused by Psoroptes ovis (Hering), has increased since the early 1970's in the USA but the disease has been eradicated from domestic sheep (Graham and Hourrigan 1977). In attempts to find acaricides to replace those presently used, a large number of established and candidate acaricides and insecticides have been evaluated (Wright and Riner 1979b). During the summer months the natural population of P. ovis mites undergoes a latent period and limited numbers of mites are available for research. However, ear canker mites [Psoroptes cuniculi (Delafond)] of rabbits are generally available throughout the year in adequate numbers for testing. The ear canker mite is very similar morphologically to P. ovis, so similar in fact that the only means of distinguishing the 2 species is in the length of the outer opisthosomal setae of the adult males (Sweetman 1958). For this reason we have used P. cuniculi as a substitute for P. ovis in the evaluation of a variety of compounds as acaricides (Wright and Riner 1979a, Wright et al. 1979, Wright and Riner 1980). In this paper, we report the results of the evaluation of 37 acaricides against both P. ovis and P. cuniculi.

^{1/}Acarina:Psoroptidae.^{2/}This paper reports the results of research only. Mention of a pesticide in this paper does not constitute a recommendation for use by the USDA nor does it imply registration under FIFRA as amended.

METHODS AND MATERIALS

The psoroptic mites used in this study were obtained from infested cattle (P. ovis) and rabbits (P. cuniculi) maintained at this laboratory. The compounds evaluated were selected from those previously tested against P. cuniculi mites. Some that were selected had demonstrated good activity and others poor activity in our 24-h evaluation (Wright and Riner 1979b). A total of 37 compounds, representing chlorinated hydrocarbons, carbamates, organophosphates, tertiary amines, and miscellaneous compounds were evaluated. Most of the acaricides were tested as company formulations [emulsifiable concentrate (EC) or wettable powder (WP)] or as technical materials formulated as EC's in xylene:Triton X-100 (65:10) to contain 10, 20, or 25% active ingredient (AI). The tertiary amines, synthesized by the Insect Physiology Laboratory, AR, SEA, USDA, Beltsville, MD, were formulated as 10% EC's in ethanol:Triton X-100 (85:5). Three bags made of heat-sealable rice paper each containing 20 to 25 P. cuniculi or P. ovis mites were dipped in each concentration of insecticide tested (Wright and Riner 1979b). The strongest concentration tested was 0.1% AI. Increasing dilutions were tested if necessary. The following classification system was developed to rate the activity of the compounds tested 24 h after dipping.

- Class I - <100 kill at 0.1%
- Class II - 100% kill at 0.1%
- Class III - 100% kill at 0.01%
- Class IV - 100% kill at 0.001%
- Class V - 100% kill at 0.0001%

RESULTS AND DISCUSSION

The results of evaluating 11 chlorinated hydrocarbons against P. ovis and P. cuniculi are given in Table 1. Five of the compounds (dieldrin, endrin, isobenzan, isodrin, and toxaphene) had excellent activity (Class V) against P. cuniculi and 2 (dieldrin and endrin) also had Class V activity against P. ovis. Seven of the compounds yielded identical results against both species, 3 (isobenzan, isodrin and toxaphene) were more effective against P. cuniculi and 1 (lindane) was more effective against P. ovis.

TABLE 1. Mortality Ratings of Selected Chlorinated Hydrocarbons Evaluated by Identical Test Conditions Against P. ovis and P. cuniculi.

Compound	Formulation	Rating ^{a/} of compounds against	
		<u>P. ovis</u>	<u>P. cuniculi</u>
aldrin	25% EC	IV	IV
chlordane	25% EC	IV	IV
chlorobenzilate	45.5% EC ^{b/}	IV	IV
dieldrin	25% EC	V	V
endrin	20% EC	V	V
heptachlor	25% EC	IV	IV
isobenzan	25% EC	IV	V
isodrin	25% EC	IV	V
lindane	25% WP ^{b/}	IV	III
methoxychlor	25% EC	I	I
toxaphene	61% EC ^{b/}	IV	V

^{a/}See text for rating system.

^{b/}Company formulation.

Eight of the 10 organophosphates evaluated gave identical results against both species of mites (Table 2). Only 1 compound, phosmet (WP), was rated as Class IV and then only against P. ovis. Phosmet (EC) was more effective against P. cuniculi than P. ovis.

TABLE 2. Mortality Ratings of Selected Organophosphates Evaluated by Identical Test Conditions Against P. ovis and P. cuniculi.

Compound	Formulation	Rating ^{a/} of compounds against	
		<u>P. ovis</u>	<u>P. cuniculi</u>
azinthosmethyl	10% EC	III	III
bromophos-ethyl	25% EC	I	I
coumaphos	25% WP ^{b/}	I	I
crotoxyphos	25% EC	III	III
fensulfothion	25% EC	III	III
fenthion	25% EC	III	III
iodofenphos	25% EC	III	III
phosmet	11.6% EC ^{b/}	II	III
phosmet	50% WP ^{b/}	IV	III
trichlorfon	25% EC	I	I

^{a/} See text for rating system.

^{b/} Company formulation.

All 7 of the carbamate and miscellaneous compounds gave identical results against both species of mites (Table 3). Two of the compounds, dinocap and methiocarb, were Class IV, while the rest were less active.

TABLE 3. Mortality Ratings of Selected Carbamates and Miscellaneous Compounds Evaluated by Identical Test Conditions Against P. ovis and P. cuniculi.

Compound	Formulation	Rating ^{a/} of compounds against	
		<u>P. ovis</u>	<u>P. cuniculi</u>
carbaryl	10% EC	I	I
cyhexatin	50% WP ^{b/}	III	III
dimethrin	25% EC	III	III
dinocap	25% EC	IV	IV
methiocarb	25% EC	IV	IV
mexacarbate	25% EC	II	II
propargite	25% EC	I	I

^{a/} See text for rating system.

^{b/} Company formulation.

Eight of 9 tertiary amine compounds were equally active (Class III) against both species (Table 4). The other compound, N,N-dimethylhexadecanamine, was more effective against P. cuniculi than P. ovis.

TABLE 4. Mortality Ratings of Selected Tertiary Amines Evaluated by Identical Test Conditions Against P. ovis and P. cuniculi.

Chemical Name	Rating ^{a/} of compounds against	
	<u>P. ovis</u>	<u>P. cuniculi</u>
<u>N,N</u> - dimethylpentadecanamine	III	III
<u>N</u> - methyl - <u>N</u> - ethylhexadecanamine	III	III
<u>N,N</u> - dimethylhexadecanamine	III	IV
<u>N,N</u> - dimethylheptadecanamine	III	III
<u>N,N</u> - dimethyl-3-methyltridecanamine	III	III
<u>N,N</u> - dimethyl-4-methyltetradecanamine	III	III
<u>N,N</u> - dimethyl-5-methylpentadecanamine	III	III
<u>N</u> - methyl- <u>N</u> -propyldodecanamine	III	III
<u>N</u> - methyl- <u>N</u> -propylundecanamine	III	III

^{a/} See text for rating system.

Against P. ovis, 2 compounds were rated Class V, 11 as Class IV, 16 as Class III, 2 as Class II, and 6 as Class I, against P. cuniculi 5 were rated as Class V, 7 as Class IV, 18 as Class III, 1 as Class II, and 6 as Class I.

Of the 37 compounds evaluated, 32 (86.5%) either were as effective against both species or were more effective against P. ovis. Five compounds were more effective against P. cuniculi (13.5%), but there was no case in which there was more than a 1 log difference in effective concentrations against the 2 species of mites. We therefore conclude that the use of P. cuniculi as a substitute for P. ovis in the evaluation of compounds for acaricidal activity is reliable.

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THE EFFECTIVENESS OF VARIOUS SECONDARY AND TERTIARY AMINES
FOR THE CONTROL OF PSOROPTES^{1/2/}F. C. Wright^{3/}, M. J. Thompson^{4/}, J. C. Riner^{3/}, and W. E. Robbins^{4/}

ABSTRACT

Forty-five C₈- to C₁₆-alkyl straight and branched chain amines were evaluated against Psoroptes cuniculi (Delafond) by the tea-bag dipping test, and 8 of the compounds were also tested against P. ovis (Hering). The most active amines tested, 1-dodecylpiperidine and N-dodecyl-N-methylcyclohexanamine, killed 100% of test mites at a concentration of 0.001%. Eighteen other amines were effective at a concentration of 0.005%. Tertiary amines were generally more active than secondary amines, and diamines were relatively inactive. Methyl branches on the alkyl chains did not seem to alter their effectiveness.

INTRODUCTION

Cattle producers and feeders are becoming increasingly concerned over the problem of psoroptic scabies in their herds. Infestations by the sheep scab mite, Psoroptes ovis (Hering), caused severe economic losses in feedlot and range cattle (Tobin 1962, Meleney and Roberts 1979), and the problem has been compounded by the dramatic increase in the incidence of the disease in recent years (Meleney and Christy 1978). At present, hot lime-sulfur, coumaphos, phosmet, and toxaphene are registered as dips to control psoroptic infestations in cattle in the USA. However, hot lime-sulfur is impractical for field use because of the need to keep the temperature of the dip solution at 35° to 40°C, so it is normally used only on lactating dairy cows. Coumaphos and phosmet, though approved by the Animal and Plant Health Inspection Service (APHIS), are not accepted as treatments by all state Animal Health Commissions. Toxaphene, on the other hand, has recently become controversial and has been labeled a carcinogen by the National Cancer Institute (Anonymous 1979). Finally, the method approved by APHIS for applying these materials, the dipping of infested or exposed cattle, is laborious and expensive.

A major objective of the Scabies and Mange Research Unit at Kerrville, TX, is to find new acaricides that are inexpensive, effective against psoroptic mites, and have low mammalian toxicity. Certain straight chain dimethyl amines have shown good effectiveness against Psoroptes cuniculi (Delafond) (Wright et al. 1979, Fisher et al. 1979), and we, therefore, decided to evaluate a number of structurally diverse secondary, tertiary, and cyclic amines against psoroptic mites. Our findings are the subject of this report.

^{1/}Acarina:Psoroptidae.

^{2/}Mention of a proprietary product does not constitute an endorsement or a recommendation of this product by the USDA.

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^{4/}Insect Physiology Laboratory, Plant Protection Institute, AR, SEA, USDA, Beltsville, MD 20705.

MATERIALS AND METHODS

Psoroptes cuniculi, the ear canker mite of rabbits, was the primary organism used to evaluate the acaricidal activity of the chemicals tested; P. ovis, the sheep scab mite, from cattle, was also used when available. These 2 species of mites are similar morphologically, and we have shown in preliminary tests that the mites react similarly to a variety of chemicals (Wright 1980). A total of 45 compounds prepared by the Insect Physiology Laboratory, Beltsville, MD, were tested including straight and branched chain amines ranging from the simple N,N-dimethyl substituted compounds to the more complex cyclic amines. The 45 amines evaluated consisted of 4 structural groups: 6 straight chain alkyl amines (1-6), 5 branched chain alkyl amines (7-11), 34 amines (12-45) containing cyclic groups, and diamines (Tables 1-4). The test method (Wright and Riner 1979) involved dipping 3 tea bags containing 20 to 25 adult or nymphal mites for 30 seconds in various concentrations of the test compounds, allowing them to air dry, and then opening the bags after 24 h to determine % mortality, which was corrected for control mortality by the method of Abbott (1925).

All of the compounds were emulsified as 10% active ingredients by the addition of a mixture (85:5, V/V) of ethyl alcohol (95%) and Triton X-100. Appropriate dilutions (50 ml volumes) were prepared with water. The highest concentration of the test chemicals was 0.1%. If 100% kill was obtained, then lower concentrations were tested (0.05, 0.01, 0.005, 0.001, 0.0005%) until 100% kill was not obtained. Control mites were dipped in dilute solvent plus emulsifier at concentrations equivalent to those used in the test solutions. Toxaphene was used as the standard.

RESULTS AND DISCUSSION

The results shown in Table 1 indicate that the secondary amine, N-(1-methyl=ethyl)-1-dodecanamine (2) was the least effective of this group against P. cuniculi. However, the tertiary amines 3, 5 and 6 were more active than the corresponding dimethylamines (Wright et al. 1979), which suggested that substituting a propyl or butyl group for 1 of the N-methyl groups increases the effectiveness of compounds with a 11- or 12-carbon chain length. On the other hand the ethyl= methylamine (1) which has a 16 carbon chain was less active than the corresponding C₁₆-dimethylamine. Even an 0.001% concentration of the latter compound caused 100% mortality of P. cuniculi and this material was the most active of the amines previously tested (Wright et al. 1979).

TABLE 1. Minimum Concentrations of Straight Chain Alkyl Amines Required to Kill^{a/} 100% of Exposed Psoroptes cuniculi and P. ovis^{b/} After 24 H. ^{c/}

Compound	Chemical name	Concentration (%)
1	<u>N</u> -ethyl- <u>N</u> -methylhexadecanamine	0.005 (0.005)
2	<u>N</u> -(1-methylethyl)-1-dodecanamine	0.05
3	<u>N</u> -methyl- <u>N</u> -(1-methylethyl)dodecanamine	0.005
4	<u>N</u> -butyl- <u>N</u> -methyldecaneamine	0.01
5	<u>N</u> -butyl- <u>N</u> -methylundecanamine	0.005 (0.005)
6	<u>N</u> -propyl- <u>N</u> -methyldecaneamine	0.005 (0.005)

^{a/} % mortality corrected by Abbott's method.

^{b/} The figures in parentheses are the minimum concentrations of amines required to kill 100% of exposed P. ovis.

^{c/} Three bags of 20-25 adult or nymphal mites were tested at each concentration.

The acaricidal activity of the branched chain alkyl dimethylamines listed in Table 2 indicates that the amine 7 was the least active, probably because of its shorter (C_8) chain length. Although N,N,3,7,11-pentamethyldodecanamine (8) was more active than compound 7, it was not as active as compounds 9, 10, and 11, all of which have longer carbon chains and only 1 methyl substituent. All 3 were active at a concentration of 0.005% against both P. cuniculi and P. ovis.

TABLE 2. Minimum Concentrations of Branched Chain Alkyl Amines Required to Kill^{a/} 100% of Exposed Psoroptes cuniculi and P. ovis^{b/} After 24 H. ^{c/}

Compound	Chemical name	Concentration (%)
7	<u>N,N,3,7-tetramethyloctanamine</u>	> 0.1
8	<u>N,N,3,7,11-pentamethyldodecanamine</u>	0.01
9	<u>N,N,3-trimethyltridecanamine</u>	0.005 (0.005)
10	<u>N,N,4-trimethyltetradecanamine</u>	0.005 (0.005)
11	<u>N,N,5-trimethylpentadecanamine</u>	0.005 (0.005)

^{a/}% mortality corrected by Abbott's method.

^{b/}The figures in parentheses are the minimum concentrations of amines required to kill 100% of exposed P. ovis.

^{c/}Three bags of 20-25 adult or nymphal mites were tested at each concentration.

The C_9 - to C_{15} -substituted cyclic amines had varying degrees of acaricidal activity (Table 3). As in the case of straight chain alkyl amines, cyclic amines substituted with alkyl chains of less than 10 carbons were considerably less active than the longer chain cyclic amines. For example, a concentration of 0.01% of 1-decylpiperidine (16) was required to kill 100% of the mites dipped, whereas 1-dodecylpiperidine (18) was completely effective at a concentration of 0.001%. A chain length of 12 carbons appears to be the optimum for maximum activity for the substituted piperidines.

Similar results were obtained with the pyrrolidines; those with chain lengths of C_{11} to C_{15} (23 to 27) were equally active and killed 100% of the test mites at a concentration of 0.005%. The presence of 3 methyl groups on the chain of the C_{12} compound (28) did not alter its activity. The C_9 - and C_{10} -substituted pyrrolidines, compounds 21 and 22, respectively, were also less active than the longer chain compounds.

Interestingly, the substitution of an oxygen atom for carbon at the C-4 position of piperidine caused the resulting morpholines to be far less active than the corresponding piperidines (Table 3). The 2 furanmethanamines (35 and 36) were lethal to mites at a concentration of 0.005%. The secondary amine, cyclohexanamine (14), was also active at this concentration and converting it to the tertiary methylcyclohexanamine (15) further enhanced its effectiveness. The diamines and the amine oxide (38) listed in Table 4 were considerably less active than the other groups of amines.

Our results in this study and in the earlier study (Wright et al. 1979) indicate that the tertiary amines are generally more effective miticides than secondary amines and that amines with an alkyl chain length of more than 10 carbons were also more effective than amines with shorter chain length. Methyl branches on the alkyl chain do not appreciably change the activity of the amines. The replacement of the dimethylamino group with cyclohexylamine, piperidine, pyrrolidine, or furanmethanamine yielded a number of highly active compounds that compared favorably in activity with the corresponding alkyl dimethyl amines (Wright et al. 1979, Fisher et al. 1979).

TABLE 3. Minimum Concentrations of Amines containing Cyclic Groups Required to Kill^{a/} 100% of Exposed Psoroptes cuniculi^{b/} and P. ovis After 24 H. ^{c/}

Compound	Chemical name	Concentration (%)
12	N-dodecyl-4-ethylbenzenamine	> 0.1
13	N-dodecyl-4-ethyl-N-methylbenzenamine	> 0.1
14	N-dodecylcyclohexanamine	0.005 (0.01)
15	N-dodecyl-N-methylcyclohexanamine	0.001 (0.005)
16	1-decylpiperidine	0.01
17	1-undecylpiperidine	0.01
18	1-dodecylpiperidine	0.001
19	1-tridecylpiperidine	0.005
20	1-tetradecylpiperidine	0.005
21	1-nonylpyrrolidine	0.05
22	1-decylpyrrolidine	0.01
23	1-undecylpyrrolidine	0.005
24	1-dodecylpyrrolidine	0.005
25	1-tridecylpyrrolidine	0.005
26	1-tetradecylpyrrolidine	0.005
27	1-pentadecylpyrrolidine	0.005
28	1-(3,7,11-trimethyldodecyl)pyrrolidine	0.005
29	4-decylmorpholine	> 0.1
30	4-undecylmorpholine	0.05
31	4-dodecylmorpholine	0.05
32	4-tridecylmorpholine	0.05
33	4-tetradecylmorpholine	0.05
34	N-decyltetrahydro-2-furanmethanamine	0.05
35	N-decyl-2-furanmethanamine	0.005
36	N-dodecyl-2-furanmethanamine	0.005

^{a/} % mortality corrected by Abbott's method.

^{b/} The figures in parentheses are the minimum concentrations required to kill 100% of exposed P. ovis.

^{c/} Three bags of 20-25 adult or nymphal mites were tested at each concentration.

In the evaluation of chemicals as acaricides, we are interested in compounds that produce 100% mortality at a concentration of 0.001% or less. In our first report on the acaricidal activity of amines (Wright et al. 1979), we noted that N,N-dimethylhexanamine was active at this level. In this study, compounds 15 and 18 were active at this concentration and another 18 amines were active at a concentration of 0.005%. Although considerably less active than toxaphene, the amines, because of their ready availability, their low cost, and their low mammalian toxicity, offer considerable promise as controls for Psoroptes spp.

TABLE 4. Minimum Concentrations of Diamines and an Amine Oxide Required to Kill^{a/} 100% of Exposed Psoroptes cuniculi.^{b/}

Compound	Chemical name	Concentration (%)
37	N,N,N',N'-tetramethyl-1,12-dodecanediamine	> 0.1
38	N,N-dimethyl-1-dodecanamine N-oxide	> 0.1
39	N'-decyl N,N-dimethyl-1,3-propanediamine	> 0.1
40	N'-decyl N,N,N'-trimethyl-1,3-propanediamine	0.1
41	N'-dodecyl N,N-dimethyl-1,3-propanediamine	> 0.1
42	N-dodecyl N,N',N'-trimethyl-1,3-propanediamine	0.01
43	N-decyl-4-morpholinepropanamine dihydrochloride	> 0.1
44	N-decyl-N-methyl-4-morpholinepropanamine dihydrochloride	> 0.1
45	N-dodecyl-N-methyl-4-morpholinepropanamine dihydrochloride	> 0.1

^{a/} % mortality corrected by Abbott's method.

^{b/} Three bags of 20-25 adult or nymphal mites were tested at each concentration.

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ACTIVITY OF CERTAIN O-ETHYL S-PROPYL PHOSPHOROTHIOATES
AND PHOSPHORODITHIOATES AND OXIME CARBAMATES AGAINST ORGANOPHOSPHORUS
RESISTANT AND SUSCEPTIBLE STRAINS OF THE TOBACCO BUDWORM^{1,2}

J. B. Nosky^{3/}, J. A. Harding^{4/}, and D. A. Wolfenbarger^{3/}

ABSTRACT

The LD₅₀'s of an organophosphorus insecticide-resistant strain (R) and a susceptible strain (S) of the tobacco budworm, Heliothis virescens (F.), did not differ when larvae were topically treated with profenofos. Also, the LD₅₀'s for sulprofos and for all but one of several O-ethyl S-propyl phosphorothioate or phosphorodithioate compounds did not differ by more than 4X. In both laboratory and field tests, profenofos was more effective than sulprofos against the tobacco budworm and the bollworm, H. zea (Boddie). Thiodicarb (dimethyl ester of N,N'-[thiobis[(methylimino)carbonyloxy]]bisethanimidothioic acid) was more effective than methomyl against Heliothis spp. in field trials but was less toxic than methomyl when applied topically to R-strain tobacco budworm larvae in the laboratory.

INTRODUCTION

Wolfenbarger and Harding (1977) showed that the tobacco budworm, Heliothis virescens (F.), was not resistant to sulprofos in 1975. However, the budworm does tend to develop resistance to insecticides very rapidly. For example, Wolfenbarger et al. (1973) found as early as 1967 that strains of the budworm were resistant to 2 organophosphorus compounds and that other strains were resistant to methyl parathion and to several other O-ethyl S-propyl phosphorothioate or phosphorodithioate compounds. Therefore, in 1978, we tested compounds with similar structure including sulprofos and profenofos against a field-collected strain of the tobacco budworm that is resistant (R) to methyl parathion and a laboratory-reared strain that is susceptible (S) to methyl parathion and compared the results with those Wolfenbarger and Harding (1977) obtained earlier. [Sulprofos is registered for use on cotton for control of the tobacco budworm and the bollworm, H. zea (Boddie); profenofos has been reported to be more effective than sulprofos against populations of Heliothis spp. in cotton (Pfrimmer 1979).] In addition, we tested the same strains to compare the toxicity of methomyl and a new carbamate insecticide, thiodicarb (Larvin®, UC-51762; dimethyl ester of N,N'-[thiobis[(methylimino)carbonyloxy]]-bisethanimidothioic acid). McGarr (1972) and Cowan and Davis (1972) found that methomyl was effective in the field against the Heliothis spp., and Wolfenbarger (1973) reported that a methyl parathion R strain of the tobacco budworm was not resistant to methomyl in laboratory tests in 1970. Thiodicarb has shown promise for control of Heliothis spp. in field trials (Pfrimmer 1979).

^{1/} Lepidoptera: Noctuidae.

^{2/} In cooperation with the Texas Agricultural Experiment Station, Texas A&M University, College Station, TX 77843. Mention of a commercial or proprietary product does not constitute an endorsement by the USDA.

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MATERIAL AND METHODS

Laboratory Tests. The R-strain tobacco budworms used in the laboratory experiments were progeny of insects collected from cotton near Brownsville during the summers of 1976 and 1978. The S strain had been reared in the laboratory for ca. 8 yr without exposure to insecticides.

The test compounds were applied topically (Wolfenbarger 1973) to 3rd-stage tobacco budworm larvae (25 mg each) with a micrometer-driven syringe. A graded series of 8 concentrations ranging from 0.078 to 10 $\mu\text{g AI}/\mu\text{l}$ of acetone was applied to 30-150 larvae in each of 3 replications. The LD_{50} values ($\mu\text{g}/\text{larva}$ at 48 h posttreatment) were calculated from log-dosage mortality curves by using the method of Daum and Killcreas (1966).

Field Tests. Tests were conducted on fruiting cotton in the Weslaco-Elsa area of Texas; cultivars were standard commercial varieties planted specifically for these tests. For the 1976 field tests (Wolfenbarger and Harding 1977), profenofos and sulprofos were formulated as emulsifiable concentrates at rates of 400 g AI/L and 720 g AI/L, respectively. For the 1977-78 field tests, thiodicarb was formulated as a 75% wettable powder, and methomyl was formulated as an emulsifiable concentrate at a rate of 216 g AI/L.

The experimental procedures used for all field tests were similar to those described by Wolfenbarger and Harding (1980). Plots 2 m wide (=2 rows) and 30 m long were arranged in a randomized complete block design (3 replicates including the untreated check), and were sprayed with the insecticide solutions at a volume rate of 21 L/ha. Sprays in all 3 yr were applied through 3 fan nozzles to only 1 row/plot; the adjacent row served as a buffer for spray drift from adjacent plots. The nozzles were 0.5 m apart on the boom and were held about 0.3 m above the plants.

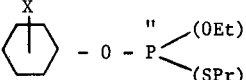
Percentage of fruit damaged by larvae of the *Heliothis* spp. was determined by examining 25 squares or bolls/plot/sampling date (3-5 dates) in each of the 3 yr. An average was then determined as percentage damage for all sampling dates. Also, in 1976, undamaged squares were counted along 3 m of row/plot/sampling date; and in 1977 and 1978, undamaged squares and bolls were counted on 2 m of row/plot/sampling date. Significant differences between means were separated by Duncan's new multiple range test at the 5% level of probability.

RESULTS AND DISCUSSION

Laboratory Tests. Differences in the LD_{50} values of the R strain and the S strain treated with compounds containing the *Q*-ethyl *S*-propyl esters did not exceed 6X and the values for compounds I, IV, V, and profenofos were about equal (Table 1). Compound III was the most toxic to the S strain and the 2nd most toxic to the R strain. Profenofos was the most toxic to the R strain, and sulprofos was the least toxic to the R strain. In general compounds with halogen atoms on the phenyl ring were more toxic than compounds with no halogen atoms.

The results with the carbamate insecticides indicated that the LD_{50} values determined for thiodicarb and compound XI on R insects were 18X and 9X greater, respectively, than those determined for the S strain (Table 2). The LD_{50} for methomyl on R insects was only 2X greater. Similar tests in 1970 by Wolfenbarger (1973) indicated that methomyl was about 4X more toxic to the S laboratory strain than to a strain collected in the field from the same general geographical area. Our results suggest that there has been no development of increased tolerance to methomyl among field populations of the pest in the Lower Rio Grande Valley of Texas.

TABLE 1. LD₅₀ Values Obtained From Topical Applications of O-ethyl S-propyl Phosphorodithioate or Phosphorothioate Compounds to a Resistant (R)^{a/} and a Susceptible (S) Strain of the Tobacco Budworm in the Laboratory.

Number	Compound		LD ₅₀ (μg/larva)		Ratio of LD ₅₀ 's (R/S)
		Y	S	R	
		X	Y		
I		O	0.7	0.8	1
II	2,4,6-tri-Cl	O (trifenofos)	0.3	0.6	2
III	4-Cl	O	0.07	0.4	6
IV	4-CH ₃	O	2.1	1.9	1
V	4-CO ₂ CH ₃	O	1.5	0.9	0.6
VI	4-NO ₂	O	0.4	0.6	2
VII	2-Cl,4-Br	O (profenofos)	0.3 ^{a/}	0.3	1
VIII	4-CH ₃ S	S (sulprofos)	1.3 ^{a/}	5.7 ^{a/}	4
IX	2,4-Di-Cl	S	0.6	2.2	4

^{a/} Taken from Wolfenbarger and Harding (1977). Larvae used in all other tests were collected in 1978 from cotton.

TABLE 2. LD₅₀ Values Obtained From Topical Applications of Carbamate Insecticides to a Resistant Strain (R)^{a/} and a Susceptible Strain (S) of the Tobacco Budworm in the Laboratory.

Compound		Formula		LD ₅₀ (μg/larva)		Ratio of LD ₅₀ 's (R/S)
Number		$\text{CH}_3-\text{C}(\text{SCH}_3)=\text{N}-\text{O}-\text{C}(\text{O})-\text{N}(\text{CH}_3)-\text{R}$	S	R		
X (thiodicarb)	R=	$-\text{S}-\text{N}(\text{CH}_3)\text{C}(\text{O})\text{ON}=\text{C}(\text{SCH}_3)\text{CH}_3$	5.0	91.8	18	
XI	R=	$-\text{S}-\text{S}-\text{C}(\text{CH}_3)_3$	2.3	21.1	9	
XII	R=	$-\text{S}-\text{N}-\text{H}$	21.3	37.3	2	
XIII	R=	$-\text{S}-\text{N}(\text{C}_2\text{H}_5)-\text{P}(\text{S})-(\text{OC}_2\text{H}_5)_2$	27.7	70.0	3	
XIV (methomyl)	R=	$-\text{H}$	8.0	18.4	2	

^{a/} Larvae used were collected in 1978 from cotton.

Field Tests. In the field tests, at least 90% of the *Heliothis* spp. populations consisted of tobacco budworms. In 1976 and 1977, all application rates of profenofos and all but 1 rate of sulprofos significantly reduced *Heliothis* damage compared with the check; the single exception was the low dose (1.12 kg/ha) of sulprofos in 1977 (Table 3). In both years, applications of profenofos (1.12 kg/ha) were significantly more active than like rates of sulprofos; profenofos reduced damage to squares and bolls by 66 and 60%, respectively, compared with the check. Thus, the effectiveness of profenofos observed in laboratory tests was apparently confirmed in field tests.

Thiodicarb significantly reduced the percentage of damaged squares compared with damage in the check (88% and 80% in 1977 and 1978, respectively) (Table 4). Also, thiodicarb at 0.56 kg/ha was more effective than methomyl at 0.25 kg/ha in 1978.

TABLE 3. Effectiveness of Profenofos and Sulprofos Against Heliothis Spp. on Cotton in Field Trials at Weslaco, TX, 1976-77.^{a/}

Compound	Application Rate (kg AI/ha)	1976 ^{b/}		Undamaged Squares/ha	1977 ^{c/}		Undamaged Fruit/ha
		% damaged Squares	% damaged Bolls		% damaged Squares	% damaged Bolls	
Profenofos	0.56	65 b	67 c	21,185 c			372,740 a
	0.84				14 a	7 a	372,840 a
	1.12	32 a	38 a	128,740 a	34 ab	15 a	318,518 a
Sulprofos	1.12	51 b	50 b	81,481 b	45 bc	15 a	135,802 b
	1.68				17 a	5 a	288,888 a
Check		94 c	95 d	0 d	59 c	38 b	244,444 ab

a/ Means within a column not followed by the same letter are significantly different as determined by Duncan's new multiple range test at 5% level of probability.

b/ Applied July 20, August 2 and 5; squares counted August 4, 6, 9, and 10; and bolls counted August 9 and 11.

c/ Applied July 22 and 28, August 2 and 9; squares counted July 29, August 4 and 11; boll and undamaged fruit counted August 4, 11, and 16.

TABLE 4. Effectiveness of Thiodicarb and Methomyl Against Heliothis Spp. on Cotton in Field Trials at Weslaco and Elsa, TX, 1977-78.^{a/}

Compound	Rate (kg/ha)	1977 ^{b/}		1978 ^{c/}		No. undamaged fruit/ha on August 18
		% damaged Squares	% damaged Bolls	% damaged squares on August 10	% damaged squares on August 16	
Thiodicarb	0.56	7 a	8 a	6 a	9 a	985,926 a
Methomyl	0.25			10 a	37 a	70,864 b
Untreated		62 b	34 b	31 b	50 b	203,161 b
						3,585 b

a/ Means in a column followed by the same letter are not significantly different from each other at 5% level of probability as determined by Duncan's new multiple range test.

b/ Applications made on July 22, 28, and August 9; squares counted July 29, August 4, and 11; and bolls counted August 4, 11, and 16.

c/ Applications made on July 31 and August 4, 8, and 11.

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EFFECTS OF GAMMA RADIATION ON MATING, REPRODUCTION, AND LONGEVITY OF LABORATORY-REARED PINK BOLLWORMS AND THEIR F₁ PROGENY CROSSED WITH MOTHS OF A LABORATORY-REARED OR NATIVE ST. CROIX STRAINT. J. Henneberry^{1/}, T. Clayton^{1/}, and D. F. Keaveny^{2/}

ABSTRACT

The number of F₁ progeny produced by irradiated (10, 15, or 20 krad) laboratory-reared pink bollworm, Pectinophora gossypiella (Saunders), males paired with untreated laboratory-reared or native St. Croix females were reduced 52-97% as compared to those produced by untreated pairs. Reduced oviposition (40-79%) and egg hatch (19-73%) occurred when surviving F₁ progeny were paired with untreated insects. Reduced oviposition (48-98%) and egg hatch (0-37%) also occurred when irradiated (5, 10 or 20 krad) laboratory-reared females were paired with untreated native St. Croix males. No F₁ progeny were produced by 20-krad irradiated females. Fertility of F₁ progeny of 5, 10, or 15-krad irradiated females was only slightly reduced. Oviposition of paired irradiated (10 krad) laboratory-reared males and females was reduced 54% and egg hatch 85%; F₁ progeny were infertile. Laboratory-reared male pink bollworms mated less with native St. Croix females than with laboratory-reared females. However, laboratory-reared females mated readily with native St. Croix males or laboratory-reared males.

INTRODUCTION

Graham et al. (1972) found that newly emerged pink bollworm, Pectinophora gossypiella (Saunders), male adults exposed to doses of gamma radiation within the range of 10 to 40 krad were highly sterile, as were F₁ progeny obtained. Cheng and North (1972) and LaChance et al. (1973) reported similar results. Graham et al. (1972) also reported that irradiated female parents (10 and 15 krad) mated to untreated males, although 77 to 89% sterile, produce a few progeny of ca. the same fertility level as untreated pairs. The more recent data of Bartlett and Butler (1979) indicate that the few F₁ female progeny from irradiated (15 to 20 krad) female parents mated to untreated males were more than 90% sterile. Using computer simulation modeling techniques, the authors showed that sterile male only, sterile female only (10, 15, or 20 krad in each case), or 10-krad mixed sex releases should reduce pink bollworm populations 82.6 to 99.9% over 2 generations. Thus, the sterile release method for suppression of established pink bollworm infestations appears to be feasible. Moreover, several field cage studies have demonstrated successful suppression of pink bollworm populations by sterile moth releases (Richmond and Graham 1970, Richmond and Graham 1971, Flint et al. 1974, Flint et al. 1975). Although these reports indicate that the method is promising, sterile moth release studies to suppress established infestations under field conditions have not been successful (Bariola et al. 1973, Graham 1978). The latter authors suggested that migrating moth populations prevented valid evaluation of the effects of the sterile releases.

Graham and Cantelo (1973) reported that there were low level populations of pink bollworm in Sea Island cotton, Gossypium spp., on the relatively isolated island of St. Croix, U.S. Virgin Islands. The island was selected in 1979 as an experimental site on which to conduct research to determine the impact of sterile pink bollworm moth releases on suppression of an established infestation. Since no information was available regarding the compatibility of irradiated laboratory-

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reared moths and moths of the native St. Croix strain, we conducted studies in 1979 and 1980 to determine the effects of gamma radiation on reproduction of Western Cotton Research Laboratory reared pink bollworm moths and their F_1 progeny crossed with native St. Croix insects. The present paper is a report of these studies.

METHODS AND MATERIALS

The laboratory-reared pink bollworms used have been cultured since 1970 on the modified wheat germ diet of Ouye (1962). The St. Croix insects were obtained from infested cotton bolls and okra pods picked on the island and air-shipped to Phoenix, AZ. Bolls and pods were cut open, larvae removed and placed in 10-cm diameter petri dishes with moist paper toweling pupation substrates. Pupae of each strain were sexed and held separately to adult emergence. Adults were irradiated when 1-2 days old in a Cs^{137} source of gamma radiation at the rate of 2243 R(Roentgen)/min.

Experiment 1 was conducted to determine the effect of gamma radiation on mating and longevity of laboratory-reared males and their F_1 progeny when paired with untreated females. Laboratory-reared males were exposed to 0, 5, 10, or 20 krad. They were then paired in groups of 5 with 5 untreated female moths of the laboratory or the St. Croix strain. Pairs were held in the mating-oviposition containers as described by Henneberry and Leal (1979). All moths were fed with food of 10% sucrose solution and each cage contained fresh cotton squares as oviposition substrates since native St. Croix moths preferred them to artificial substrates (Henneberry, unpublished data).

Eggs laid on squares were counted daily, and the squares with eggs were placed in 946-ml waxed-paper rearing containers with artificial diet (Ouye 1962). The numbers of eggs hatched were recorded ca. 1 wk later. Numbers of mature larvae and pupae were also recorded, and pupae were sexed (Butt and Cantu 1962) and held separately to obtain F_1 adults. Male and female progeny were paired, as described, with moths of the opposite sex of the laboratory-reared or the St. Croix strain.

Experiments 2, 3, and 4 were conducted in a similar manner, but in Experiment 2 laboratory-reared females were exposed to single doses of 0, 5, 10, and 20 krad and subsequently crossed to untreated St. Croix male moths. In Experiments 3 and 4, males and females, respectively, were exposed to 0, 10, or 15 krad before pairing with untreated native St. Croix moths. Experiment 5 was conducted to determine the effect of exposing both sexes of laboratory-reared moths to gamma radiation before pairing. F_1 progeny were reared on artificial diet as described in Experiment 1.

Treatments in all experiments were replicated 3-7 times, and the studies were conducted in temperature-controlled cabinets at 26°C and 14:10 h light-dark photoperiods.

RESULTS

Experiment 1. In most cases, irradiated and untreated laboratory-reared males mated less frequently with St. Croix females than with laboratory-reared females (Table 1, Experiment 1). Oviposition of females of the laboratory-reared strain mated to irradiated males (10 and 20 krad) was reduced. These results are similar to those of LaChance et al. (1975, 1978). The authors reported that lack of oviposition response of untreated females mated to irradiated males probably was a result of lack of transfer of eupyrene sperm and/or accessory gland fluid. Egg hatch was also reduced 21 and 54%, respectively. The number of progeny from untreated females of either strain mated to 10 or 20 krad treated males was reduced more than 83%.

Treatment did not affect longevity of treated males except for males exposed to 5 krad. Untreated St. Croix males were shorter lived than untreated laboratory-reared males.

F_1 male progeny of laboratory-reared or native St. Croix females mated to 20 krad or 10 krad laboratory-reared males, respectively, were over 70% sterile (Table 2); the fertility of F_1 female progeny was affected less. Mating of F_1 progeny was not affected by radiation treatment of the parents. However, longevity of F_1 progeny was reduced.

TABLE 1. Effects of Gamma Radiation on Mating, Oviposition, Reproduction, and Longevity of Pink Bollworms.^{a/}

Cross ^{b/}		Treatment	Spermato-	%	No.	%	F ₁	Longevity ^{c/}
♀	♂	(krad)	phores/♀	mated	eggs/♀	hatch	adults ♀	(days)
Experiment 1 - L Male Parents Irradiated								
L	L	0	2.4ab	92a	121a	67a	21.8	18.3a
SC	L	0	0.8c	50b	32cd	71a	6.4	17.7a
SC	SC	0	1.1c	66ab	20cd	72a	1.5	7.3b
L	TL	5	2.4ab	75ab	81ab	65a	10.5	9.7b
		10	2.9ab	95a	46b-d	53ab	3.7	14.1ab
		20	3.3a	100a	55bc	31b	0.8	12.5ab
SC	TL	5	1.1c	55b	23cd	68a	0.5	11.7ab
		10	0.7c	65ab	9d	48ab	0.6	13.7ab
		20	1.8bc	55b	9d	48ab	0.1	14.5ab
Experiment 2 - L Female Parents Irradiated								
L	L	0	2.3a	90a	12ab	67a	23.0	17.8a
L	SC	0	2.0a	83a	44b	50a	6.4	18.0a
SC	SC	0	1.1a	66a	22b	73a	0.5	12.3a
TL	SC	5	1.5a	80a	66b	69a	4.1	14.5a
		10	2.1a	80a	29b	48a	1.4	15.0a
		20	2.2a	84a	3c	42a	0	20.7a

a/Means of 5 replications, 3-5 moth pairs/replication. Means within a column and experiment not followed by the same letter are significantly different. Duncan's multiple range test (P=0.05).

b/Parental designations used in all tables are: L=untreated laboratory-reared, TL=gamma-irradiated laboratory-reared pink bollworms, SC=untreated native pink bollworm from St. Croix.

c/Longevity of treated males in Experiment 1, irradiated females in Experiment 2.

Experiment 2. Treated or untreated laboratory-reared females mated as frequently with untreated St. Croix males as with untreated laboratory-reared males (Table 1, Experiment 2). Oviposition of treated or untreated laboratory-reared females mated to St. Croix males was reduced as compared to oviposition of laboratory-reared females mated to laboratory-reared males.

No F₁ adults were produced by 20-krad treated females mated to St. Croix males, and numbers produced by 10 krad laboratory-reared females were reduced ca. 78%. Irradiation did not affect female longevity.

F₁ progeny of 5 or 10 krad irradiated laboratory-reared females mated to untreated St. Croix males were approximately as fertile as progeny from untreated female parents (Table 2). However, no F₁ progeny were produced by 20-krad treated laboratory females mated to untreated St. Croix males. Mating of the F₁ progeny from irradiated females mated to St. Croix males was not affected by parental treatment but was significantly less than mating of the F₁ progeny of untreated laboratory-reared moth pairs.

Experiments 3 and 4. The numbers of F₁ progeny per female produced by untreated St. Croix females paired with 0, 10, or 15 krad laboratory-reared males were 9, 1.9, and 0.4, respectively. Untreated native St. Croix females paired with F₁ male progeny of 10 or 15 krad treated laboratory-reared males produced few fertile eggs

TABLE 2. Effects of Gamma Radiation on F_1 Progeny of TL Pink Bollworm Males or Females Crossed with Untreated L or SC Insects.^{a/}

F ₁ Cross		Parental treatment ^{b/}		Spermato-	No.	%	Longevity	
♀	♂	♀	♂	phores/♀	eggs/♀	hatch	♂	♀
Experiment 1 - L Male Parents Irradiated								
L	F ₁	L	L (0)	3.3ab	111a-c	69a-c	16.1a-c	15.9ab
		L	TL (5)	2.3bc	55cd	60a-d	12.3cd	12.9b
		L	TL (10)	1.7c	67b-d	45d	18.5ab	14.1b
		L	TL (20)	2.6bc	38d	18e	10.3d	18.2a
F ₁	L	L	L (0)	3.6ab	159a	70a-c	17.9ab	16.3ab
		L	TL (5)	2.2bc	57cd	57cd	12.1cd	13.1b
		L	TL (10)	2.7bc	34d	18e	13.2b-d	16.4ab
		L	TL (20)	2.2bc	34d	18e	13.2b-d	16.4ab
F ₁ SC	F ₁ F ₁	L	L (0)	4.2a	119ab	58b-d	16.9a-c	18.5a
		SC	L (0)	2.2bc	85b-d	73a-c	17.0a-c	13.9b
		SC	TL (5) ^{b/}	3.0	89	48	15.5	14.8
		SC	TL (10) ^{b/}	2.1	62	20	9.9	15.8
F ₁	SC	SC	L (0)	2.7bc	153a	76a	15.1a-d	18.7a
		SC	TL (5) ^{b/}	3.3	52	59	16.8	12.5
		SC	TL (10) ^{b/}	2.0	71	54	16.3	13.3
		SC	L (0)	3.1a-c	155a	75ab	19.3a	16.6ab
F ₁	F ₁	SC	L (0)	3.1a-c	155a	75ab	19.3a	16.6ab
Experiment 2 - L Female Parents Irradiated								
L	F ₁	L (0)	SC	2.9bc	87bc	72a	14.0a-c	13.7d
		TL (5)	SC	1.8c	49cd	51a	16.1a-c	14.6cd
		TL (10)	SC	1.9c	40d	50a	12.3c	16.3b-d
F ₁	L	L (0)	SC	2.1c	89bc	72a	15.5a-c	15.3cd
		TL (5)	SC	1.7c	79b-d	75a	12.9bc	17.4a-c
		TL (10)	SC ^{b/}	2.0	19	54	11.9	16.4
L	F ₁	L (0)	L	3.4ab	110b	68a	16.1a-c	15.9b-d
F ₁	L	L (0)	L	3.5ab	160a	72a	17.9a	16.3b-d
F ₁	F ₁	L (0)	L	4.1a	118ab	59a	16.9ab	18.5ab

^{a/}Means of 5 replications, 3-5 moth pairs/replication. Means in a column not followed by the same letter are significantly different. Duncan's multiple range test ($P=0.05$).

^{b/}Few F_1 progeny produced, 1-2 replications, data not included in statistical analysis.

(Table 3, Experiment 3). F_1 female progeny paired with untreated St. Croix males produced as many fertile eggs as F_1 female progeny of untreated parents. Longevity of F_1 progeny was variable, and there were no significant differences related to irradiation of the parent moths.

The numbers of F_1 progeny per female produced by 0, 10 or 15 krad treated laboratory-reared females paired with untreated St. Croix males were 7, 1.1, and 0.2, respectively. F_1 progeny of irradiated females (10 or 15 krad) produced nearly as many fertile eggs as the F_1 progeny of untreated female parents (Table 3, Experiment 4).

Experiment 5. When males and females were exposed to 5 or 10 krad before pairing, oviposition and egg hatch were significantly reduced (Table 4). Mating was not affected by the treatment. Longevity of males exposed to 5 or 10 krad was

TABLE 3. Effects of Gamma Radiation on F₁ Progeny of TL Males or Females Crossed with Untreated SC Pink Bollworm Moths.^{a/}

F ₁ Cross		Krad/parental treatment		Spermato-	%	No.	%	Longevity	
♀	♂	♀	♂	phores/♀	mated	eggs/♀	hatch	♂	♀
Experiment 3 - L Male Parents Irradiated									
SC	F ₁	SC	L (0)	0.5bc	53a-c	56a	83a	10.8a	11.6a
		SC	TL (10)	0.6bc	46bc	14b	28cd	12.1a	10.5a
		SC	TL (15)	0.3c	26c	2b	3d	10.3a	14.2a
F ₁	SC	SC	L (0)	1.3a	80ab	64a	62ab	13.9a	13.8a
		SC	TL (10)	1.1ab	86a	62a	46bc	15.2a	16.4a
		SC	TL (15) ^{b/}	1.7	100	36	58	25.0a	25.3a
F ₁	F ₁	SC	L (0)	1.0ab	80ab	77a	76a	13.1a	13.6a
		SC	TL (10)	-	-	-	-	-	-
		SC	TL (15)	1.0ab	88a	86	21d	13.6a	16.9a
Experiment 4 - L Female Parents Irradiated									
SC	F ₁	L (0)	SC	0.2c	26b	18b	60a	18.8a	9.2b
		TL (10)	SC	0.1c	13b	17b	40a	10.6b	7.8b
		TL (15)	SC ^{b/}	1.7	66	50	81a	19.0	17.8
F ₁	SC	L (0)	SC	1.0b	73a	37b	59a	12.1ab	20.5a
		TL (10)	SC	0.3c	33b	25b	45a	8.3b	18.2a
		TL (15)	SC ^{b/}	2.0	100	31	35	7.5	14.2
F ₁	F ₁	L (0)	SC	2.3a	100a	109a	75a	18.3a	18.9a

^{a/}Means of 5 replications, 3-5 moth pairs/replication. Means in a column and experiment not followed by the same letter are significantly different. Duncan's multiple range test ($P=0.05$).

^{b/}Few F₁ progeny produced, 1-2 replications, data not included in statistical analysis.

TABLE 4. Effects of Gamma Radiation on Mating, Oviposition, and Longevity of TL Male and Female Pink Bollworm Moths.^{a/}

Cross		Krad parental treatment (both sexes)	Spermato-	%	No.	%	F ₁ adults/♀	Longevity	
♀	♂		phores/♀	mated	eggs/♀	hatch		♂	♀
TL	TL	0	3.9a	100a	147ab	91a	27.9	27a	20ab
TL	TL	2.5	3.9a	100a	173a	77b	17.7	24ab	17b
TL	TL	5.0	3.2a	100a	81b	51c	7.5	17c	18ab
TL	TL	10.0	3.6a	100a	68b	14d	1.4	20bc	22a

^{a/}Means of 5 replications, 7 moth pairs/replication. Means in the same column not followed by the same letter are significantly different. Duncan's multiple range test ($P=0.05$).

reduced but females did not appear affected except at the 2.5 krad treatment. F_1 progeny of 10 krad treated pairs produced no fertile eggs (Table 5).

DISCUSSION

Laboratory-reared males mated more frequently with laboratory-reared females than with St. Croix females. However, St. Croix pairs generally mated less than laboratory-reared pairs and laboratory-reared females mated as frequently with males of either strain. LaChance et al. (1975) reported similar results with a native and a laboratory-reared pink bollworm strain and suggested that laboratory-reared females had been selected for higher mating frequency. This may be a partial explanation for these results; however, the native St. Croix strain in the present studies may also have reacted to the artificial environment of mating-oviposition cages and/or other environmental laboratory conditions. Native pink bollworm females hand-collected in cotton fields on St. Croix have been found to contain as many as 6 spermatophores (D. F. Keaveny, unpublished data). Thus the mating interactions of laboratory-reared insects with native St. Croix insects should be studied under field conditions. However, Van Steenwyk et al. (1979) reported a similar lack of male competitiveness between a laboratory-reared untreated or irradiated pink bollworm strain and a native strain under field cage conditions. Irradiated (10, 15, or 20 krad) laboratory-reared male pink bollworm moths mated with untreated laboratory-reared or native St. Croix females produced a few F_1 male progeny which were highly sterile; a lesser degree of inherited sterility occurred in the F_1 female progeny. Although irradiated females (5, 10 krad) paired with untreated males produced fewer eggs and fewer F_1 progeny than untreated pairs, the fertility level of these progeny was nearly as high as that of untreated parents. Therefore, it appears that the radiation dose applied to the insects in a mixed-sex release system should be 15 krad or higher to minimize the effect of recovered fertility in F_1 females produced.

TABLE 5. Effects of Gamma Radiation on F_1 Progeny of TL Pink Bollworm Parents Crossed with Untreated SC Moths.^{a/}

F_1 Cross		Krad Parental treatment		Spermato-phores/♀	♂	%	mated	No. eggs/♀	%	hatch	Longevity	
		♀	♂								♂	♀
SC	F_1	L (0)	L (0)	1.1ab	80ab	3a	6c	17a	12a			
		L (2.5)	L (2.5)	0.9ab	73a-c	17a	58a	16a	14a			
		L (5.0)	L (5.0)	0.7b	59bc	14a	28bc	13a	12a			
		L (10.0)	L (10.0)	0.5b	46c	2a	0c	14a	13a			
F_1	SC	L (0)	L (0)	1.2ab	93a	28a	43ab	11a	17a			
		L (2.5)	L (2.5)	1.8a	80ab	6a	27bc	9a	16a			
		L (5.0)	L (5.0)	0.8b	66a-c	14a	18bc	12a	15a			
		L (10.0)	L (10.0) ^{b/}	1.0	83	1a	0c	9	13			
F_1	F_1	L (0)	L (0)	1.8a	86ab	36a	68a	14a	12a			
		L (2.5)	L (2.5)	1.9a	83ab	23a	26bc	13a	15a			
		L (5.0)	L (5.0)	1.8a	93a	9a	18bc	115a	13a			
		L (10.0)	L (10.0) ^{b/}	-	-	-	-	-	-			

^{a/}Means of 5 replications, 3 moth pairs/replication. Means in the same column not followed by the same letter are significantly different ($P=0.05$).

^{b/}Few or no F_1 progeny, data not included in statistical analysis.

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DEVELOPMENT OF THE SOUTHWESTERN CORN BORER^{1/} ON THREE ARTIFICIAL DIETS^{2/3/}Tina A. Whittle^{4/} and Robert L. BurtonWheat and Other Cereal Crops Research Unit, AR, SEA, USDA
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ABSTRACT

Southwestern corn borer, *Diatraea grandiosella* (Dyar), development was compared on wheat germ, bean, and CSM diets by determining larval stage duration, pupal weights, fecundity, % egg hatch, female moth longevity, and number of spermatophores/female moth. Pupal weights, fecundity, and % egg hatch were significantly higher on bean and CSM diets than on wheat germ diet. Other parameter measurements were nearly the same for all diets.

INTRODUCTION

The study of the southwestern corn borer, *Diatraea grandiosella* (Dyar), a major pest of corn in the southern United States (Henderson and Davis 1969), is often facilitated by laboratory rearing; artificial diets have been developed for this purpose. Keaster and Harrendorf (1965) modified the wheat germ diets of Adkisson et al. (1960) and Vanderzant et al. (1962) to a medium suitable for rearing southwestern corn borers. Since then, other workers (Bailey and Chada 1968, Jacob and Chippendale 1971, Davis 1976) have also used different modifications of the wheat germ diet in southwestern corn borer rearing programs, and a premixed wheat germ diet for these insects is commercially available from Bio-Serv, Inc., Frenchtown, NJ.

In the present study, southwestern corn borer development on a modification of the wheat germ diet used by Jacob and Chippendale (1971) was compared with that on a lima bean diet (Burton 1969, Shorey and Hale 1965) and CSM (corn, soy flour, milk solid blend) diet (Burton 1970), which have both been used extensively in rearing other lepidopterous insects. As the latter 2 diets require fewer ingredients, are more easily formulated, and are more economical than the wheat germ diet, their successful utilization by the southwestern corn borer could be an asset in rearing these insects.

METHODS AND MATERIALS

Ingredients of all 3 diets are listed in Table 1. The wheat germ and CSM diets were prepared by 1st mixing the agar with cold water and then bringing the solution to a boil; this solution was then combined with the other ingredients in a 1-L blender and mixed thoroughly with the blender set at high speed. In formulating the lima bean diet, the beans were covered with water, brought to a boil, and simmered until softened (about 10 min). Unabsorbed cooking water was

1/ Lepidoptera: Pyralidae.

2/ Mention of a proprietary product in this paper does not constitute an endorsement of this product by the USDA.

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TABLE 1. The Composition of the 3 Artificial Diets Studied.

Ingredient	Amounts/L in indicated diet		
	Wheat Germ	Bean	CSM
Casein	44 g	-	-
Sucrose	23 g	-	-
Wheat germ	28 g	50 g	-
Salt mix ^{a/}	9 g	-	-
Ascorbic acid	5 g	3 g	4 g
Alphacel	5 g	-	-
Na alginate	5 g	-	-
B-sitosterol	2 g	-	-
Locust bean gum	3 g	-	-
Sorbic acid	1.5 g	1.5 g	1.5 g
Methyl paraben	2 g	2 g	2 g
Vitamins ^{a/}	25 ml	-	-
Choline chloride (10%)	19 ml	-	-
KOH (4 M)	5 ml	-	-
Wheat germ oil	3 ml	-	-
Formaldehyde (10%)	4 ml	4 ml	4 ml
Water	500 ml	400 ml	500 ml
Agar	20 g	20 g	20 g
Water for agar	500 ml	400 ml	400 ml
Lima beans	-	105 g	-
Torula yeast	-	32 g	15 g
CSM	-	-	188 g

^{a/} Jacob and Chippendale (1971).

retained and included in the total amount needed for the diet; the softened beans and water were blended at high speed until homogenous in texture. From this point, preparation of the bean diet proceeded as described for the other 2 diets.

Immediately following preparation, ca. 12 ml of each diet were dispensed into 30-ml plastic cups that were set into 5 cardboard trays (25 cups/tray) under a laminar flow clean work bench that had been wiped with 10% Clorox[®]. The diet was allowed to cool and solidify, then the diet in each cup was pierced with forceps to provide entry holes for the larvae. The trays of diet were stored in plastic bags at 4°C for no more than 5 days before use.

One 1st-instar southwestern corn borer larva was transferred to each cup of diet with a small artist paint brush cleaned in 10% Clorox, and infested cups were fitted with paperboard caps. Fifty cups were infested/day for 5 days/wk throughout the study.

The southwestern corn borer colony was maintained in a small room held at approximately 25°C, with a 16-h photoperiod to prevent diapause (Chippendale and Reddy 1973). Relative humidity was uncontrolled, but the trays of larvae were held in plastic bags for the 1st wk, and moisture from the diet kept the humidity quite high. During the 2nd wk, the trays were transferred to a plastic-lined, enclosed shelf, which provided moderate humidity. The trays were then moved to an open shelf, where the larvae completed their development under less humid conditions. This procedure prevented desiccation of diet and larvae, and also deterred the development of microbial contamination that sometimes became a problem during extended periods of high humidity.

After the larvae had pupated, the sexes were separated and placed in 4-L ice cream cartons until their emergence as adults. At that time, the newly emerged male and female moths used in the study were paired and placed in 500-ml paper cups. The cups were lined with waxed paper as a substrate for oviposition and the tops were covered with cheesecloth held in place by a rubber band. The remaining moths were released into round wire mesh cages wrapped with waxed paper.

The moths rested on the mesh and oviposited through it onto the waxed paper. Egg-covered strips of paper were then placed in large desiccators lined with damp paper towels.

During all stages of their life cycle, the southwestern corn borers on each diet were segregated from those on the other diets. In this way, the eggs from 1 generation provided the basis for a 2nd generation reared on the same respective diet; the data in this study were collected for 2 generations.

Southwestern corn borer development was compared on wheat germ, bean, and CSM diets by determining larval stage duration, pupal weights, fecundity, % egg hatch, female moth longevity, and number of spermatophores/female moth. Larval stage duration was determined by dating cups of larvae when 1st infested and checking larvae daily until pupation occurred. All larvae that pupated were included in this portion of the study. The remainder of the data collection incorporated equal numbers of insects from each diet, and quantities mentioned hereafter in this section refer to the number of insects evaluated/diet during each of the 2 generations covered by the study.

Seventy-five pupae of each sex were weighed 4 days after pupation. (This was done because the younger pupae are soft and easily damaged by handling.) Following adult emergence and mating, the fecundity of 40 female moths was assessed by counting the eggs laid by each mated moth during its lifespan. The longevity of these moths was also recorded, and after death, the number of spermatophores present was determined by dissection. Egg hatch was determined by observing groups of 100 eggs cut from egg-covered waxed paper. One group was placed in each of seven 30-ml plastic cups fitted with paperboard caps and stored in a desiccator containing damp paper towels.

RESULTS AND DISCUSSION

The average values of all parameters determined for southwestern corn borers on each of the 3 diets are shown in Table 2. The larval stage of insects on CSM diet was extended by 1 day compared to those on the wheat germ diet and the bean diet. Both male and female pupae reared on bean diet or CSM diet weighed more

TABLE 2. Different Parameters Measured for Southwestern Corn Borers on 3 Artificial Diets (mean values \pm SD).^{a/}

	Diet		
	Wheat Germ	Bean	CSM
Duration of larval state (days)	23.9 \pm 1.9 _a	24.1 \pm 1.1 _a	25.2 \pm 1.4 _a
Weight of male pupae (mg)	112.0 \pm 20.0 _a	132.0 \pm 26.0 _b	130.4 \pm 2.5 _b
Weight of female pupae (mg)	145.9 \pm 30.0 _a	184.4 \pm 36.0 _b	179.5 \pm 33.0 _b
Number of eggs laid/female	213.1 \pm 64.6 _a	290.0 \pm 63.5 _b	290.8 \pm 51.2 _b
Eggs hatched (%)	93.0 \pm 4.6 _a	95.8 \pm 2.0 _b	95.4 \pm 2.1 _b
Lifespan of females (days)	3.7 \pm 0.5 _a	4.0 \pm 0.4 _a	4.0 \pm 0.2 _a
Spermatophores/female	1.2 \pm 0.4 _a	1.1 \pm 0.4 _a	1.1 \pm 0.3 _a

a/ Values in the same row followed by different subscripts are significantly different at the 0.01 level (except for % eggs hatched: 0.05 level) according to Duncan's new multiple range test.

than those grown on wheat germ diet; this difference was highly significant ($P = 0.01$). Significantly more eggs ($P = 0.01$) were laid by moths reared on CSM diet and bean diet than by moths reared on wheat germ diet, and eggs laid by moths from the former 2 diets had a significantly higher ($P = 0.05$) hatch. On the other hand, longevity of the adult females and the number of spermatophores present in the female moths differed very little among the 3 diets. Larval mortality on all 3 diets was rare and, when it occurred, was usually caused by secondary microbial contamination. Also, the incidence of adults with deformed wings was negligible.

There was no apparent decline in the quality of southwestern corn borers reared on any diet over the 2 generations during which the study was conducted or through 5 successive generations following the study. The information collected, particularly pupal weights, indicates that all 3 diets met the nutritional requirements of this insect, including the needs established by Reddy and Chippendale (1972) for protein, B-vitamins, choline chloride, and inorganic salts, and by Chippendale and Reddy (1972) for linoleic acid and B-sitosterol. While those nutrients were provided in the wheat germ diet by specific supplements, no such additions were necessary for the bean and CSM diets.

The data obtained from this study strongly suggest that southwestern corn borers can be reared successfully on bean and CSM diets as well as on wheat germ diet. However, certain aspects of development on bean and CSM diets were significantly superior. The larger pupae, more fecund moths, and higher % egg hatch resulting from rearing on bean diet and CSM diet, in addition to the ease of preparation and economy of these diets, suggests that they have good potential for use in southwestern corn borer rearing programs.

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EFFECT OF COTTON PLANT SIZE, HOST EGG LOCATION, AND LOCATION
OF PARASITE RELEASE ON PARASITISM BY TRICHOGRAMMA
PRETIOSUM^{1/2/}

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ABSTRACT

Greenhouse studies indicated that mean % parasitism of tobacco budworm, Heliothis virescens (F.), eggs by the parasite Trichogramma pretiosum Riley after release in a greenhouse was significantly affected by the size of the cotton plant, the vertical location of the host eggs within the plant canopy, and the location of parasite release. Overall mean % parasitism of eggs on small plants (32.8 ± 1.5 cm) was greater than that on larger plants (61.5 ± 3.8 cm) regardless of parasite release location. Significantly greater parasitism was observed for eggs located in the lower and middle parts of the plant than for those in the upper part of the plant when parasites were released below canopy level but was greater for eggs in the upper canopy when parasites were released above the canopy. Factorial analysis indicated that both plant size and vertical location of host eggs were significant factors and that interaction between these factors was present.

INTRODUCTION

The use of Trichogramma spp. to suppress lepidopteran populations on field crops has received considerable attention in recent years (Ridgway et al. in press). However, the degree of successful use of these parasites has been relatively unpredictable; thus, the need for additional research on factors affecting parasite efficacy has been suggested (Ables et al. 1979a, Ridgway et al. in press). Ridgway et al. (in press) recently reviewed the factors that appear to affect the field performance of Trichogramma, and among the important factors they listed was parasite area of search. In their theoretical appraisal of Trichogramma, Knipling and McGuire (1968) suggested that as host plants grew in size, an increasing number of parasites would be required for pest suppression. Need and Burbutis (1979) tested and substantiated the inverse relationship between growing corn plants and the searching ability of Trichogramma nubilale Ertle and Davis, which attacks European corn borer, Ostrinia nubilalis (Hübner).

We report here results of a study conducted to determine the influence of cotton plant size on parasitism of tobacco budworm, Heliothis virescens (F.), eggs by T. pretiosum Riley. We also report the effect of host egg location and location of parasite release within the plant on the activity of this parasite.

METHODS AND MATERIALS

Potted cotton plants ('Stoneville 213' variety) used in the test were designated as small or large according to average stem height (32.8 ± 1.5 and

^{1/}Hymenoptera: Trichogrammatidae.

^{2/}In cooperation with the Texas Agricultural Experiment Station, Texas A&M University, College Station, TX 77843.

61.5 ± 3.8 cm, respectively). The plants were randomly mixed in groups of 10 plants on concrete benches in a 180-m² greenhouse. Tobacco budworm eggs (< 24 h old) from a laboratory culture were transferred with a moistened brush to cotton plants. Single eggs were placed on 2 leaves at each of 3 levels of the plant canopy for a total of 6 eggs/plant. The 3 levels approximated the upper 1/3, middle 1/3, and lower 1/3 of the plants.

Parasites were obtained from a laboratory culture that was reared on the Angoumois grain moth, *Sitotroga cerealella* (Olivier). We released the parasites by 2 different methods (equivalent rate of 247,000/ha), either by placing cups of emerging adults below canopy level on the benches between the plants or by suspending the cups ca. 1 cm above the canopy. Host eggs were collected 30 h after parasite release and held at 27°C for 10 days at which time percentage parasitism was recorded. Ambient temperature in the greenhouse during the test ranged from 15 to 35°C. The test was replicated 3 times with 5 plants/size group/replicate for each parasite release method. The data were analyzed with a factorial analysis of variance (Freese 1967).

RESULTS AND DISCUSSION

Percentage parasitism of host eggs on the 2 plant size groups and at the 3 different canopy levels following parasite release from 2 locations is shown in Table 1. Release of parasites from either below or above the plant canopy resulted in significantly ($P < 0.05$) greater parasitism of eggs on the smaller plants than on the larger plants. Thus, the searching efficiency of *T. pretiosum*, like that of *T. nubilale* (Need and Burbutis 1979), appears to decrease as the size of the host plant increases.

TABLE 1. Mean Percentage Parasitism of *Heliothis virescens* Eggs at 3 Canopy Levels on Small and Large Cotton Plants in a Greenhouse Following the Release of *Trichogramma pretiosum* Below and Above the Plant Canopy.^{a/}

Egg location on plant	$\bar{X} \pm SD$ % egg parasitism		
	Small plants (32.8 ± 1.5 cm)	Large plants (61.5 ± 3.8 cm)	Overall \bar{X}
<u>Release Below Plant Canopy</u>			
Upper 1/3	83.1 ± 5.4 A,ab/	48.1 ± 6.7 A,b	65.8 ± 18.4 A
Middle 1/3	90.7 ± 4.5 B,a	88.7 ± 5.6 B,a	89.7 ± 5.3 B
Lower 1/3	95.3 ± 3.3 C,a	88.7 ± 7.2 B,b	91.9 ± 6.5 C
Overall X	89.7 ± 5.03 a	75.3 ± 19.0 b	
<u>Release Above Plant Canopy</u>			
Upper 1/3	91.9 ± 3.4 A,a	76.2 ± 6.2 A,b	84.1 ± 7.8 A
Middle 1/3	63.3 ± 5.5 B,a	24.4 ± 7.5 B,b	43.8 ± 19.4 B
Lower 1/3	79.6 ± 5.8 B,a	35.5 ± 7.9 B,b	57.6 ± 22.1 B
Overall X	78.3 ± 11.8 a	45.4 ± 22.3 b	

^{a/}Means of 3 replicates of 5 plants with 2 eggs at each plant stratum. Parasites were released at the equivalent rate of 247,000/ha.

^{b/}Means within columns followed by the same capital letter and means across columns followed by the same lower case letter are not significantly different at the 0.05 probability level based on the *t*-test for comparisons among means in a split-plot design (Freese 1967).

Parasitism of host eggs after release of the parasites below the plant canopy level differed significantly at each of the 3 canopy levels on small plants and decreased from lower to the upper regions. On the large plants, parasitism of eggs did not differ significantly between the lower and middle regions but was significantly less in the upper region of the plants. Conversely, when the parasites were released above the plant canopy, parasitism of host eggs on small and large plants was significantly greater for eggs located in the upper than in the middle and lower levels of the plants. Regardless of parasite release location, F tests for size, level, and size:level interaction were all significant ($P < 0.001$), which indicated that both plant size and the canopy level affected the rate of parasitism and that there was interaction between the 2 factors. Location of parasite release was also a significant factor because the parasites appeared to search most effectively in the level of the plant canopy in which they were released.

Burbutis et al. (1977) reported that T. nubilale discovered more corn borer egg masses located in the lower and middle portions of corn plants, which is where eggs of that host are most frequently deposited. Thus, this parasite species appeared to be well adapted to finding its host.

Gonzalez et al. (1970) reported that the release of T. pretiosum onto field-caged cotton resulted in greater parasitism of eggs which were placed in the upper region of the plants where the majority of Heliothis eggs are normally deposited. Our results do not consistently agree with those of Gonzalez et al. (1970). In their test, the latter authors used paper cards to which ca. 100 host eggs were attached and the cards were secured to the plants. Thus, host eggs were concentrated compared to their normal distribution when laid individually by the female moth. Perhaps this factitious egg distribution affected parasite search behavior somewhat differently than in our studies which involved much lower densities of individually placed eggs. More importantly, the location in which the parasites are released appears to account for the location of parasite activity on the host plant. Burbutis et al. (1977) reported that the performance of T. nubilale was influenced by release sites within corn plants. In the present study when T. pretiosum was released below the plant canopy, parasite searching on the smaller plants apparently was not strongly influenced by this release site because the majority of eggs at all 3 canopy levels were parasitized. However, parasite searching on large plants was apparently restricted to the lower two-thirds of the plants. Release of the parasites above the plant canopy resulted in the greatest parasitism of eggs located in the upper 1/3 of the plant, regardless of plant size.

Field releases of T. pretiosum onto cotton would not normally be made until plants were as large or larger than the 61-cm plants used in our study. Therefore, our data suggest that placement of parasites in the lower plant regions would not result in an effective distribution of the parasites and that, as suggested by Ables et al. (1979b), precise placement of the parasites within the host plant habitat may critically affect parasite performance. Furthermore, approaches such as those of Ables et al. (1979a) and Jones et al. (1979), which attempt to place T. pretiosum in the upper part of the cotton plant, are probably appropriate. However, it should be noted that the greenhouse conditions during our tests would likely differ from actual field conditions and thus, parasite behavior may also differ. Nevertheless, T. pretiosum, if released in or near the upper portion of the cotton plant where Heliothis spp. eggs are normally found, seems behaviorally well suited for release against these pests in cotton.

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