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A PESTICIDE CALIBRATION AND MIXING PROGRAM FOR HAND-HELD CALCULATORS[1]

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Abstract: This paper reports on a software program written for a hand-held Hewlett-Packard HP-41 calculator that aids in pesticide mixing and sprayer calibration. Advantages offered by the calculator system over similar personal computer programs include reduced cost, field portability, and the ability to input spraying parameters in a combination of English and metric units. Furthermore, the calculator program is less rigid in input requirements and allows changes in individual sprayer parameters without reentry of unchanged parameters that personal computer programs require.

Key Words: Calculator, insecticide, mixing, software.

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The recent increase in personal computer usage by agricultural experimental station personnel and pesticide industry representatives has led to the development of software that aids in small-plot pesticide studies. These programs provide such things as plot randomization schemes, plot layout maps, and reports; and may be used to determine the amounts of chemicals to add to the spray tanks (PDMP 1986). Although the benefits of these mixing programs, i.e. improved accuracy and reproductibility, are obvious, these programs also represent some disadvantages. Mixing programs must be run on relatively expensive personal computers which are seldom taken into the field. Thus changes in sprayer parameters made in the field cannot be easily incorporated into the program. The mixing programs currently available also lack a routine for sprayer calibration. The program user is simply prompted to input the sprayer output. Following this, the user is required to enter additional sprayer parameters by following a rigid menu. If only slight differences in spraying parameters are needed, e.g. two different rates of the same material, the user must reenter all spraying parameters. A final problem with current mixing programs arises because pesticide mixing is commonly performed with a combination of English and metric units. Spray boom width, length of sprayer travel, and rate of chemical, i.e., lb. AI per A, are commonly measured in English units while volume of spray per nozzle, volumes of liquid formulations and weights of solid formulations are often measured in metric units. Personal computer software allows for metric or English calculations but not combinations.

The objective of this report is to describe a software program written for a hand-held Hewlett-Packard HP-41 calculator which aids in sprayer calibration and allows the input of a combination of English and metric units. The program also utilizes the user-definable keys on the HP-41 in order to eliminate reentry of unchanged sprayer parameters.

The HP-41 system components required are the HP-41 calculator, overlay (Fig. 1) and the program. Magnetic program cards and a card reader are beneficial but not required. Program lines are listed in Fig. 2. These lines may be entered

[1] Accepted for publication 30 October 1987.

through keystrokes or by use of the optional card reader. Once entered, the non-volatile HP-41 memory will retain the program, provided memory is not overwritten, after calculator power is turned off.

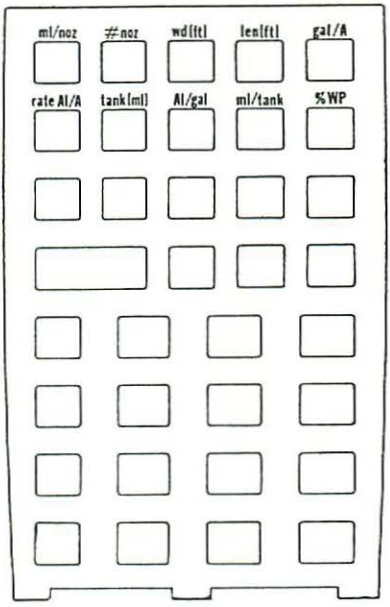

Fig. 1. Calculator overlay with spraying parameters.

The program is initialized and executed by pressing the following keys: EXQ, ALPHA, S, P, R, A, Y, E, R, and ALPHA. These keystrokes result in the user being prompted for sprayer parameters. The first four parameters (spray volume in ml per nozzle per time unit, e.g. 30 sec; number of nozzles; spray width in ft; and length in ft of sprayer travel per time unit, e.g. 30 sec) are required for sprayer calibration and are listed on the overlay above keys A, B, C and D, respectively (Fig. 1). Each parameter is entered and the corresponding key (A, B, C or D) is pressed to store it. For example, to calibrate a backpack sprayer with three nozzles, the user would measure the amount of spray delivered by one nozzle in 30 sec, e.g. 54 ml. This value (54) is entered and key A is pressed. The values for the number of nozzles (3) is entered and key B is pressed. The width of the spray pattern in ft is measured, the value, e.g. 6, is entered and key C is pressed. Finally, the distance in ft that the sprayer travels in 30 sec is measured, the corresponding value, e.g. 80, is entered and key D is pressed. The sprayer output (gal per A) is next determined and displayed by pressing key E. In this example, 'GAL:A = 3.88' is displayed when key E is pressed. Additional parameters of chemical rate (lb AI per A), tank capacity (ml) and liquid formulation (lb AI per gal) are entered with keys F, G and H, respectively (Fig. 1). For example, if carbaryl (4 lb AI per gal) is to be applied with the aforementioned 3-nozzle backpack sprayer at a rate of 1.5 lb AI per A and the amount of spray required or tank capacity is 2000 ml, the following keystrokes are needed. The rate 1.5 is

entered and key F is pressed. The value of the tank capacity (2000) is entered and key G is pressed. The formulation value (4) is entered and key H is pressed. The amount (ml) of liquid formulation to add to the tank is then calculated and displayed when key I is pressed. In this example the display is 'ML/TANK=193.1'. If a second carbaryl rate of 1 lb AI per A is needed, the rate value (1) is entered and key F is pressed. The amount of material to add to the tank is determined by simply pressing key I. For wettable powder (WP) formulations, when the percentage AI of the material is entered into key H, the amount of WP in g to be added to the spray tank is displayed.

The sprayer calibration and mixing program may appear complicated, however it offers a quick and functional method for sprayer calibration and mixing. Total time for program initialization, sprayer calibration and mixing determinations for a small plot study with 10 insecticide treatments, is generally less than two min. Furthermore, the calculator system is portable, less expensive and, under most field conditions, offers a practical alternative to the personal computer program.

Program Line	Comment
01 LBL 'SPRAYER'	labels program
02 SF 27	places calculator in user mode
03 'PARAMETERS'	prompts program user for
04 PROMPT	sprayer parameters
05 LBL A	stores ml per nozzle in
06 STO 01	register 01
07 RTN	
08 LBL B	stores number of nozzles
09 STO 02	in register 02
10 RTN	
11 LBL C	stores spray pattern width
12 STO 03	in register 03
13 RTN	
14 LBL D	stores distance of sprayer
15 STO 04	travel in register 04
16 RTN	
17 LBL E	calculates sprayer output (GAL/A)
18 RCL 01	total GAL caught = (ml per nozzle *
19 RCL 02	number nozzles)/3785
20 *	
21 3785	
22 /	
23 RCL 03	area sprayed = (spray pattern width *
24 RCL 04	distance of sprayer travel)/43560
25 *	
26 43560	
27 /	
28 /	GPA = total GAL caught/area sprayed
29 STO 05	
30 'GAL:A='	
31 ARCL 05	
32 AVIEW	displays GPA
33 RTN	
34 LBL F	stores pesticide rate in
35 STO 06	register 06
36 RTN	
37 LBL G	stores sprayer tank capacity in
38 STO 07	register 07

Fig. 2. Sprayer calibration and mixing program lines.

Program Line	Comment
39 RTN	
40 LBL H	stores liquid formulation in
41 STO 08	register 08
42 RTN	
43 LBL I	calculates amount of liquid formulation
44 RCL 06	to add to spray tank
45 RCL 07	ml = (lb AI per A * ml per tank)/
46 *	(GPA * lb AI per GAL)
47 RCL 05	
48 RCL 08	
49 *	
50 /	
51 'ML/TANK='	
52. ARCL X	
53 AVIEW	displays ml per tank
54. RTN	
55. LBL J	calculates amount of WP
56. STO 09	to add to tank
57. 100	g = (100 * lb AI per A * ml per tank
58. RCL 06	* 454) / (WP% * GPA * 3785)
59. *	
60. RCL 07	
61. *	
62. 454	
63. *	
64. RCL 09	
65. RCL 05	
66. *	
67. 3785	
68. *	
69. /	
70. 'G/TANK='	
71. ARCL X	
72. AVIEW	displays grams per tank
73. .END.	

Fig. 2. Continued.

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FUMIGANT ACTION OF VARIOUS INSECTICIDES ON THE EGG AND FIRST LARVAL STAGE OF *HELIOTHIS ZEA*[1]

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Abstract: The fumigant effect of chlorpyrifos, methyl parathion, monocrotophos, methomyl, amitraz, chlordimeform, and fenvalerate was studied in the laboratory on *Heliothis virescens* (F.) eggs and young larvae. Chlordimeform and methyl parathion caused ca. 30% egg mortality while the other compounds caused ca. 20% mortality. Vapors of both chlorpyrifos and methyl parathion induced high mortality of first instar larvae, while mortality due to the vapor of the remaining compounds did not differ from the water control. Larval mortality was greatest when larvae were exposed to insecticide vapor during emergence.

Key Words: Fumigant effect, tobacco budworm, cotton, chlorpyrifos, methyl parathion, chlordimeform.

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The tobacco budworm (TBW), *Heliothis virescens* (F.), is a major pest of cotton in the United States. This pest and the cotton bollworm, *H. zea* (Boddie), are the primary causes of the loss of cotton production throughout the cotton belt (Head 1985; King et al. 1986). To reduce crop loss due to these *Heliothis* species, numerous pesticide treatments are applied to commercial cotton fields, primarily by aerial application. The main disadvantages of this application method are incomplete coverage and poor penetration into the cotton canopy (Uk and Courshee 1982). However, if insecticides are used which have an active vapor phase, control might be achieved through a fumigant effect.

In a previous paper we reported on the ovicidal activity of various insecticides on TBW (Horowitz et al. 1987). The purpose of this paper is to report on the fumigant effect on these insecticides on the egg and early larval stages of TBW.

MATERIALS AND METHODS

A colony of TBW was maintained for this study with the original material obtained from a laboratory colony at the ARS-USDA Western Cotton Research Lab, Phoenix, Arizona. Larvae were reared on a pinto bean diet at room temperature (21 to 24°C). Moths were allowed to mate in a cylindrical carton lined with paper towel which served as a substrate for oviposition.

Seven insecticides and a water control were tested in this study: monocrotophos (Azodrin 5M, Shell Corp.), chlorpyrifos (Lorsban 4E, Dow Chemical Corp.), methyl parathion (Methyl Parathion 5M, FMC Corp.), methomyl (Lannate 1.8W, DuPont Corp.), amitraz (Baam 1.5EC, Nor-Am Chemical Corp.), fenvalerate (Pydrin 2.4 EC, DuPont Corp.) and chlordimeform (Galecron 4E, Ciba-Geigy). In each case, an aqueous solution of the formulated material was used. The concentrations treated were extrapolated from field rates (see Table 1).

[1] LEPIDOPTERA: Noctuidae. Accepted for publication 30 October 1987.

The technique used for testing fumigant effect of each material was modified from Sun and Johnson (1963). Each test unit consisted of a plastic petri dish (10 cm diameter). The insecticide being tested was incorporated into a 4 cm^2 piece of filter paper; the filter paper was dipped into an aqueous solution of the formulated material for 3 s, then allowed to air dry for 1 h under a fume hood. In each test, a control using only water was also prepared. The paper was then placed onto a petri dish lid and covered with a 9-cm-diameter filter paper (Whatman #1) to provide a barrier against direct contact of insecticide by the insect.

Eggs used in each test were no more than 24 h old. A piece of paper towel containing ova was placed in the bottom of the petri dish and the dish closed with the lid containing the insecticide. Each dish was then sealed with masking tape. The units were maintained at $27 \pm 0.5°C$, $60 \pm 5\%$ RH, and continuous light. Under these conditions, the majority of larval emergence occurred after 43 to 48 h. For each test, one set of ova (15 to 25 eggs) was exposed to insecticide for 24 ha second for 48 h. Eggs were then transferred into clean dishes and egg mortality recorded after 72 h.

A possible delayed fumigant effect of each insecticide on ova was evaluated by examining the mortality of larvae hatching from these eggs. Tests were conducted as previously described, with eggs exposed for a 42 h period after which ova were transferred to clean petri dishes containing artificial diet. In tests of chlorpyrifos and methyl parathion, larval mortality was determined after exposure periods of 24, 32, 42, and 72 h. In all tests, mortality of 1st stage larvae was determined at 32 h after larval emergence. Larvae were considered dead if they did not respond to a probe test.

A second test was conducted with chlorpyrifos and methyl parathion to determine whether death of newly emerged larvae resulted from fumigant penetration of the ova and delayed effect on the embryo (Smith and Salkeld 1965) or from larval exposure to insecticide absorbed by the substrate. Following the technique described previously, ova were exposed to the insecticide for 32 h, after which each ovum was taken from the paper and transferred to a clean dish containing a small amount of bean medium.

A third test was conducted to determine the relative insecticide absorption of cotton leaf and filter paper substrates and the resulting larval mortality. Circular sections (4.0 cm diameter) were exercised from cotton leaves and placed on a thin layer of agar (for maintaining leaf turgidity) in a small petri dish lid (4.0 cm diameter). Pieces of cotton and filter paper (of approximately the same dimensions) were exposed to the vapors of either chlorpyrifos or methyl parathion for 42 h. A small section of paper towel containing ova was then placed on each substrate.

Each test was replicated on at least three different days with ca. 20 eggs per replicate. Data were transformed using an arc-sin transformation and analyzed by two-way analysis of variance and Duncan's Multiple Range Test (Duncan 1955).

RESULTS AND DISCUSSION

All materials tested affected TBW eggs through their vapor phase compared with the water control (Table 1). Chlordimeform and methyl parathion caused ca. 30% oval mortality; the other compounds caused ca. 20% mortality. No significant difference was found between the two exposure periods (24 and 48 h) tested. In

both exposure periods, the embryo continued to develop. Thus, the egg's mortality was determined if the larvae did not hatch.

Table 1. Fumigant effect of various insecticides on tobacco budworm eggs.

Treatment	Concentration (mg AI/ml)	n*	% Mortality†
Chlordimeform	0.8	175	33.9a
Methyl parathion	2.5	179	31.6ab
Chlorpyrifos	3.1	281	23.0bc
Amitraz	5.0	169	22.4bc
Methomyl	2.8	239	22.3bc
Monocrotophos	3.1	176	22.0bc
Fenvalerate	0.6	207	18.9c
Water Control		160	5.4d

* The data from two exposure periods (24 to 48 h) were pooled since no significant difference was found ($P > 0.05$).

† Means followed by the same letter are not significantly different ($P = 0.05$; Duncan's [1955] multiple range test).

Eggs exposed to vapors of both chlorpyrifos and methyl parathion caused high mortality of 1st instar larvae, while mortality due to the remaining compounds did not differ significantly from the water control (Table 2).

Table 2. Effect of exposure of tobacco budworm eggs to insecticide vapors for 42 hours on 1st instar larvae.

Treatment	n	% Mortality*
Chlorpyrifos	107	96.4a
Methyl parathion	105	84.7a
Monocrotophos	92	5.4b
Fenvalerate	64	4.5b
Amitraz	56	1.0b
Chlordimeform	61	1.0b
Methomyl	75	1.0b
Water Control	155	0.5b

* Means followed by the same letter are not significantly different ($P = 0.05$; Duncan's [1955] multiple range test).

The removal of eggs from the poisoned substrate was found to substantially reduce subsequent larval mortality, particularly regarding exposure to methyl parathion (Table 3). The effect of the period of exposure to the substrate on larval mortality is shown in Table 4. Larval mortality was greatest when larvae were exposed to insecticide vapors during emergence (72-h exposure) with almost total mortality resulting. The level of mortality was generally correlated with the period of exposure.

Larval exposure to both substrates (cotton and filter paper) treated with methyl parathion or chlorpyrifos vapor showed no significant difference in larval mortality (Table 5). The larval mortality in this test was lower (especially with methyl parathion) than found when pieces of paper containing the eggs were directly exposed to insecticide vapor.

Table 3. Mortality in tobacco budworm larvae following transfer of eggs exposed to insecticides for 32 hours to an untreated substrate.

Treatment	Type Transfer*	n	% Mortality†
Chlorpyrifos	NT	192	61.3a
Methyl parathion	NT	171	59.0a
Chlorpyrifos	TS	89	17.7b
Methyl parathion	TS	60	2.7c
Water	NT	151	0.1c
Water	TS	38	0.0c

* TS, each egg was transferred to a clean dish after exposure for 32 h; NT, non-transfer.

† Means followed by the same letter are not significantly different (P = 0.05; Duncan's [1955] multiple range test).

Table 4. Ovicidal/larvicidal effect after different exposure time of eggs to various insecticidal vapors.

Treatment	Period (h)	n	% Mortality*
Chlorpyrifos	72	109	97.5a
Chlorpyrifos	42	107	96.3ab
Methyl parathion	72	82	91.0ab
Methyl parathion	42	105	84.7b
Chlorpyrifos	32	192	61.3c
Methyl parathion	32	171	59.0c
Methyl parathion	24	67	55.8cd
Chlorpyrifos	24	80	33.9d
Water	24	96	0.5e
"	32	217	0.3e
"	42	155	0.1e
"	42	155	0.1e
"	72	63	0.0e

* Means followed by the same letter are not significantly different (P = 0.05; Duncan's [1955] multiple range test).

Table 5. Ovicidal/larvicidal effect by leaf discs or filter paper after having been exposed to the vapor phase of chlorpyrifos or methyl parathion.

Treatment	Substrate	n	% Mortality*
Chlorpyrifos	cotton leaf	217	81.9a
	filter paper	170	78.1a
Methyl parathion	cotton leaf	174	14.4b
	filter paper	253	5.4b
Water	cotton leaf	163	0.0c
	filter paper	96	0.0c

* Means followed by the same letter are not significantly different (P = 0.05; Duncan's [1955] multiple range test).

Previous studies have shown that the ovicide chlordimeform (CDF) affected eggs of various insects (including TBW) and spider mites through the vapor phase (Dittrich 1966, 1967; Phillips 1971; Streibert and Dittrich 1977). We also observed ca. 30% mortality of TBW eggs produced by the fumigant action of CDF. Methyl parathion demonstrated the same level of vapor activity (Table 1). The other two ovicides, methomyl and fenvalerate, that were found effective against TBW eggs (Horowitz et al. 1987) caused only ca. 20% egg mortality through their vapor phase. However, in a study by Chalfant et al. (1979), no fumigant effect was found for CDF, methomyl, or fenvalerate on eggs of the cabbage looper.

Streibert and Dittrich (1977) found that exposure of both young eggs (0 to 24 h old) and old eggs (24 to 28 h old) of four noctuids and one coccinellid to a saturated atmosphere of CDF induced a similar level of mortality. Similarly, our results with CDF, as well as other insecticides, show that short egg exposure (24 h) to the vapors caused the same level of mortality as longer exposures (48 h). Smith and Salkeld (1966) pointed out that early ovicidal treatment with organophosphates allowed continued development of the embryo to the stage when cholinesterase and acetyl-choline are found with mortality occurring at this stage. Apparently, poisoning of young TBW eggs occurred by a similar process.

Both chlorpyrifos and methyl parathion demonstrated high fumigant effect on young TBW larvae. Chlorpyrifos, which exhibited low ovicide activity (Horowitz et al. 1987), was found very effective against TBW young larvae with high larval mortality occurring soon after hatching (Table 2). Vapor of CDF showed a very low level of larvicidal activity on TBW. Streibert and Dittrich (1977) also reported TBW larvae to be less sensitive to vaporized CDF than the eggs.

Smith and Salkeld (1965) reported no oval mortality in the large milkweek bug from exposure to parathion vapor, but nymphs from treated eggs died soon after hatching. We found no such delay effect in our study, as egg treatment did not produce larval mortality (Table 3). We suggest that mortality resulted from larval contact with the contaminated substrate, cotton leaf or filter paper, which absorbed the insecticide vapors. In the case of chlorpyrifos, the small amount of vapor that apparently attached to the egg shell caused ca. 18% mortality of the young larvae.

Our findings demonstrate potential fumigant action of CDF, chlorpyrifos, and methyl parathion on TBW eggs and young larvae. This potential may be of importance in practical application, since the incomplete coverage and penetration associated with aerial applications could be accomplished by the fumigant activity of these insecticides.

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SPATIAL AND SEASONAL DISTRIBUTION OF DAMAGE TO APPLES BY *ARGYROTAENIA CITRANA* (FERNALD) AND *PANDEMIS PYRUSANA* KEARFOTT[1]

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Abstract: Within-tree distribution and seasonal occurrence of fruit damage caused by *Argyrotaenia citrana* (Fernald) and *Pandemis pyrusana* Kearfott was studied in Watsonville, CA apple orchards in 1983 and 1985. There was no significant difference ($P > 0.05$) in the distribution of damage between 10 regions of the tree at bloom, but the proportion of fruit damaged at bloom which remained on the tree declined as the season progressed. Total number of fruit damaged by both species increased as the season progressed, and stabilized by mid to late August. There was a significant difference ($P < 0.05$) in within-tree distribution of damage by both species in both years. The proportion of total damage located in the lower regions of the tree was 0.32 in 1983 and 0.38 in 1985. The proportion of total damage in the upper regions of the tree was 0.68 in 1983 and 0.62 in 1985.

Key Words: *Argyrotaenia citrana*, *Pandemis pyrusana*, apples, distribution, sampling.

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California has five principal apple growing regions (Tyler et al. 1983): the central coast, the northern coast, the central valley, southern California, and the Sierra Nevada Mountain foothills. Pest complexes are different in each locality, but the principal insect pest in all localities is the codling moth, *Cydia pomonella* (L.). Two other tortricids, the orange tortrix, *Argyrotaenia citrana* (Fernald) and the apple pandemis, *Pandemis pyrusana* Kearfott also cause significant fruit damage in coastal apple orchards, especially those orchards not receiving insecticide treatment. *Argyrotaenia citrana* and *P. pyrusana* which feed on fruit are known as 'apple skinworms' in this region because of their surface feeding habit which causes fruit scarring. Knowledge of the biology of these species on apple is limited, although Essig (1958) reported that both *A. citrana* and *P. pyrusana* feed on the leaves and fruit of apple.

Argyrotaenia citrana, the more damaging of these two species on apples in California, has a wide host range including orange, walnut, willow, Monterey pine, and numerous weeds including curley dock, filaree, mallow, mustard and various grasses (Lange 1936; Coquillet 1984). This insect has been a major pest of grapes in coastal regions since 1968, possibly due to changing orchard floor management from winter discing to using herbicides or cover crops (Kido et al. 1981). *Argyrotaenia citrana* is found throughout coastal California including the interior valleys of the coast ranges. The warmer and drier climate inland limits its distribution (Basinger 1938). Pheromone lures are commercially available for *A. citrana*. However, their usefulness is limited because no numerical relationship has been established between the number of moths caught and the level of infestation of any crop (Kido et al. 1981). There are three generations per year in California.

[1] LEPIDOPTERA: Tortricidae. Accepted for publication 30 October 1987.
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Here we describe damage to apples caused by *A. citrana* and *P. pyrusana* at bloom and after petal fall, including seasonal and within-tree distribution of damage. We also suggest how this knowledge may be applied in a monitoring program.

MATERIALS AND METHODS

The same ten 'Yellow Newtown Pippin' apple trees were examined for fruit damage every 2 to 3 weeks, beginning 8 weeks after petal fall in 1983, and 4 weeks after petal fall in 1985, until harvest in unsprayed Watsonville, CA orchards. The orchard used in 1983 was 15 years old with an average tree height of 4 m. The orchard used in 1985 was 10 years old with an average tree height of 3 m. Both orchards were densely planted in hedgerows, and were in an area of other apple orchards. Twenty randomly selected fruit were examined from the center and each exterior quadrant delimitated by compass direction in both the upper and lower halves of the tree. Therefore, ten locations per tree canopy were sampled in this manner. Fruits were not removed. A total of 200 fruits were examined per tree on each date. The presence and cause of damage was identified in both years. In 1985, an effort was made to determine whether the damage was caused by *A. citrana* or *P. pyrusana*. Multivariate analysis was conducted from bloom damage, damage by each species, and total damage to determine distribution of damage in the tree and seasonal damage trends. Results of this analysis were used to develop sampling strategies. Binomial data were transformed using an arc sine conversion prior to analysis.

RESULTS AND DISCUSSION

Damage by both species which occurs during bloom later appears as bronze colored, roughened scars on the fruit surface. Damage after petal fall results in shallow, irregular scars on the surface. Fruit packers in the Watsonville area have established quality thresholds for total fruit damaged in this manner: (7% for fresh market and 10% for processing market). Loads with proportion of damaged fruit exceeding these levels are not marketable.

Bloom damage

There were no significant differences ($P > 0.05$) in the levels of damage that occurred at bloom among the 10 locations sampled on the trees in 1983 ($F = 1.62$; $n = 10$) and 1985 ($F = 0.693$; $n = 10$), nor were there any interactions between location within the tree and the date on which the sample was taken in 1983 ($F = 1.32$; $n = 50$) and 1985 ($F = 1.02$; $n = 70$). The proportion of bloom damage located in the upper half of the tree was 0.53 in 1983 and 0.45 in 1985.

In both years the proportion of fruit that remained on the tree after being damaged at bloom declined as the season progressed. In 1983, 0.074 of the fruit on the tree on the initial sampling date (23 June) had been damaged at the time of bloom. Some fruit damaged during bloom remained on the tree through harvest, but there was no significant difference ($F = 1.42$; $P > 0.05$; $n = 5$) between the sampling dates for these fruits. Fruit damage at bloom was lower in 1985, and only 0.001 of these fruits were present on the trees on the 25 June sampling date. No fruit damaged at bloom remained on the tree on the 12 July sampling date. There was a significant difference ($F = 6.316$; $P < 0.01$; $n = 7$) in fruit damaged at bloom that remained on the trees for the sampling dates in 1985 (Table 1).

Table 1. Mean and standard deviation proportion of fruit (n = 200 per tree) damaged at bloom remaining on each tree (n = 10) on each sampling date in 1985.

Date	$\bar{x} \pm SD$
5/24	0.14 ± 0.49a*
6/7	0.15 ± 0.43a
6/25	0.09 ± 0.32a
7/12	0.01 ± 0.10b
7/30	0.00 ± 0.00b
8/14	0.00 ± 0.00b
8/20	0.00 ± 0.00b

* Means followed by different letters are significantly different by Duncan's (1951) multiple range test.

Total damage

Total damage resulting from the feeding of *A. citrana* and *P. pyrusana* increased significantly ($P < 0.05$) as the season progressed in both 1983 ($F = 6.32$; $n = 5$) and 1985 ($F = 6.27$; $n = 7$) (Table 2). In both years, the greatest increase in damage occurred after the late June sampling date. Total damage did not increase significantly after mid to late August.

Table 2. Mean and standard deviation number of total fruit damaged (200 fruit per tree from 10 trees) by *A. citrana* and *P. pyrusana* on each sampling date in 1983 and 1985.

1983		1985	
Date	$\bar{x} \pm SD$	Date	$\bar{x} \pm SD$
—	—	5/24	0.2 ± 1.4a*
—	—	6/7	0.5 ± 2.2a
6/23	7.4 ± 7.0a*	6/25	0.9 ± 2.9ab
7/12	12.4 ± 11.2b	7/12	2.5 ± 5.6bc
7/28	13.2 ± 11.0b	7/30	3.9 ± 7.9c
8/16	10.5 ± 10.0b	8/14	6.3 ± 10.2d
9/13	13.2 ± 13.2b	8/20	5.7 ± 9.4d

* Means followed by different letters in each year are significantly different by Duncan's (1951) multiple range test.

No effort was made to distinguish the damage of the two species in 1983. In 1985, we distinguished between damage of each species on each sampling date. There were significant differences ($P < 0.01$) in damage increments between sampling dates for both *A. citrana* ($F = 19.51$; $n = 7$) and *P. pyrusana* ($F = 5.88$; $n = 7$). Some new damage by *A. citrana* was observed on all but the last sampling date. New damage by *P. pyrusana* was only found for the three sampling dates from mid July to mid August. Overall, most of the damage was caused by *A. citrana* (Fig. 1).

Distribution of damage

There was a significant difference ($P < 0.05$) in the distribution (Table 3) of total fruit damage that was recorded for the 10 locations sampled on the trees in

both 1983 (F = 12.21; n = 10) and 1985 (F = 1.899; n = 10). The most consistent difference was between damage in the lower five locations sampled and the upper five locations sampled. The upper region of the tree had the greater proportion of damaged fruit on every sampling date in both years. The average proportion of total damage located in the upper regions of the tree was 0.68 in 1983 and 0.62 in 1985.

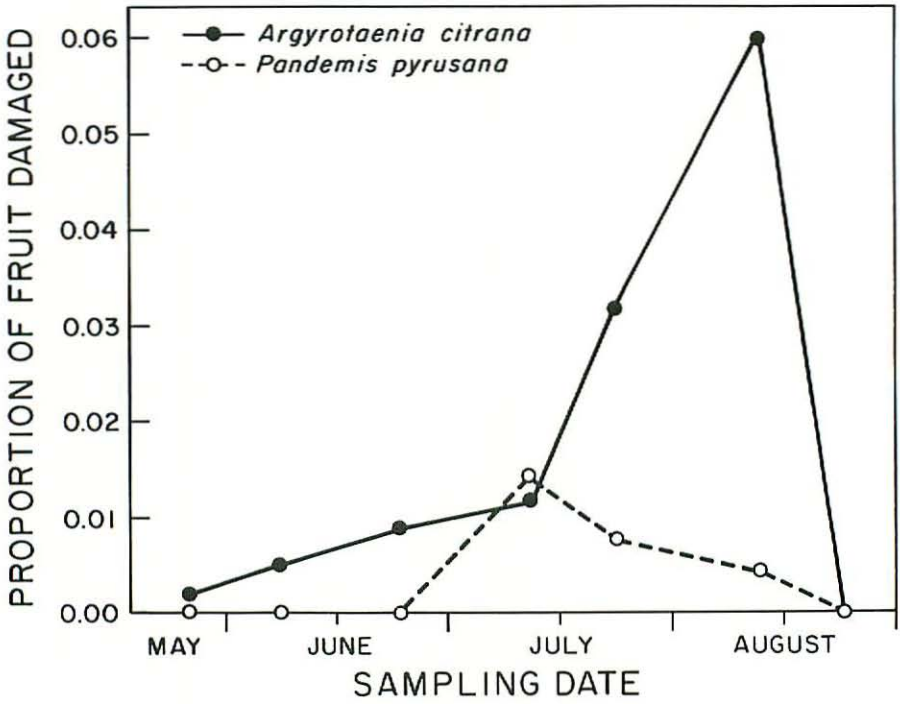

Fig. 1. Proportion of all fruit sampled that had been damaged since the previous sample by *A. citrana* and *P. pyrusana*. On each sampling date in 1985, 200 fruits were examined on each of 10 trees.

Monitoring

Management of *A. citrana* and *P. pyrusana* can be improved by assessing the severity of fruit damage at regular intervals, then making a control action decision based on observation of total damage and increase in damage from the previous sampling date.

As there is no significant difference in the distribution of damaged fruit between the center of the tree and the four tree quadrants, samples of equal size taken from both the lower and upper regions of the tree could be used to represent accurately the amount of damage present on the tree. However, if an individual samples fruit exclusively from the lower region of the tree, as would be most convenient in a monitoring program, the damage estimate must be weighted to reflect the differences in distribution of damage by the two species between the

lower and upper tree regions. Weighting can be achieved by increasing the damage estimate of the sample by a factor of 1.41 (the ratio of vertical distribution of damage on the tree if damage in both tree regions were equivalent [0.500] to the average proportion of actual damage from the lower portion of the tree [0.354]). A control action should be initiated during the season if the estimated amount of damage (actual damage in lower portion of tree × 1.41) approaches the level of damage at which packers or processors would reject the fruit. Similarly, the grower should sort damaged fruit from undamaged fruit in the harvesting process if the estimated amount of damage in a pre-harvest sample exceeded the level of damage acceptable to the packer or processor at harvest.

Table 3. Mean, standard deviation, and proportion fruit damage (20 fruit per location) for the 10 tree locations sampled.

Location on tree	Fruit Damage	
	$\bar{x} \pm SD$	Proportion
Lower/North	1.14 ± 1.23a*	5.7
Lower/South	1.36 ± 1.66a	6.8
Lower/East	1.70 ± 1.73a	8.5
Lower/West	1.28 ± 1.23a	6.4
Lower/Center	1.58 ± 1.95a	7.9
Upper/North	3.10 ± 2.42bc	15.5
Upper/South	3.14 ± 2.04bc	15.7
Upper/East	3.24 ± 2.84bc	16.2
Upper/West	2.46 ± 2.04b	12.3
Upper/Center	3.64 ± 2.69c	18.2

* Means followed by different letters are significantly different by Duncan's (1951) multiple range test.

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RESISTANCE TO THE FALL ARMYWORM[1] IN SORGHUM SEEDLINGS FROM ETHIOPIA AND YEMEN

B. R. Wiseman[2] and G. R. Lovell[3]

Abstract: New collections of sorghum, *Sorghum bicolor* (L.) Moench, from Ethiopia (5200 entries) and Yemen (3350 entries) were screened as seedlings for leaf-feeding resistance to larvae of fall armyworm, *Spodoptera frugiperda* (J. E. Smith). Approximately 100 of the entries were rated as having resistance that was better than the resistant check. Under a second evaluation in a replicated test, only 17 and 13 of the entries rated significantly better than the resistant check on the first and second ratings, respectively.

Key Words: *Spodoptera frugiperda*, *Sorghum bicolor*, leaf-feeding damage.

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The search for resistance in plants to insect pests should be a continual process. However, most current plant resistance programs have identified sufficient genetic diversity in their respective crops and, thus, the 'continual seaching' part of their program is minor or non-existent. As new collections of germplasm become available, we as plant resistance researchers must examine them against as many pest organisms as possible to determine their possible utility. Once a sufficient level of resistance is found, a breeding program should utilize the germplasm to develop it into usable forms so that the resistance can eventually be used by commercial industry and/or the grower.

This paper reports seedling resistance data in sorghum from Ethiopia and Yemen which are housed at USDA-ARS Plant Introduction Station at Experiment, GA.

MATERIALS AND METHODS

Seed from both sorghum collections were increased at the Puerto Rico Station at Mayaguez in groups of ca. 3,000 each year. Then they were packaged at Experiment, GA, in lots of 30 seeds/package for screening at Tifton, GA. The collections were evaluated for seedling resistance to the fall armyworm (FAW), *Spodoptera frugiperda* (J. E. Smith), during the winter months of 1985-1987.

Galvanized metal trays (187.5 cm × 90 cm × 10 cm deep) were constructed and filled with ca. 7.5 cm of fine washed river sand for each planting. Seeds of ca. 250 sorghum collection entries were evaluated in single rows per tray in rows of ca. 15 cm in length with ca. 2.5 cm spaces between entry ranges. Seeds were planted in moistened sand in rows 15-cm long, 2.5 cm apart, and 1.25 cm deep. Outside rows were bordered with a commercial hybrid. A resistant (1821 [CIMMYT] cm) and a susceptible (Huerin Inta) check were planted in each range of 50 single row entries. Infestations of ca. four FAW neonates per seedling were made using the modified 'bazooka' (Wiseman et al. 1980a; Wiseman and Gourley 1982). Precalibrations

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of the 'bazooka' were made in the laboratory prior to infestations on each seedling test. Fall armyworm larvae were reared on a pinto bean diet (Perkins 1979). All screenings were made using the Insect Biology and Population Management Research Laboratory's original culture. The larvae were dispensed in corncob grits directly onto the sand adjacent to treatment rows 2 d after the seedlings had emerged. The screenings (20-25 plants/single row plots) were accomplished in a greenhouse maintained at ca. 27-30°C. The tests were watered daily by gently applying the water between the ranges or between the treatment rows.

All entries were visually rated for damage (plot ratings) after Huerin Inta, the susceptible check, approached a maximum damage rating of 9 [ca. 4-5 d after infestation (DAI)] (Wiseman et al. 1980b). The visual rating scale used was that of 0-9 devised by Wiseman et al. (1966) where 0 = no damage; i = small amount of pinhole-type injury; 2 = several pinholes, 3 = small amount of shot-hole type injury with 1 or 2 lesions; 4 = several shot-hole type injuries and a few lesions; 5 = several lesions; 6 = several lesions, shot hole injury and portions eaten away; 7 = several lesions and portions eaten away and areas dying; 8 = several portions of the whorl eaten away and areas dying; and 9 = the whorl completely eaten away and more areas dying or plant dead. Additional ratings were recorded on successive days to determine if any entries would sustain less damage that the resistant, 1821 cm, check.

When the screening for seedling resistance of both the Ethiopia and Yemen sorghums was completed in the winter of 1987, the best 100 performing entries (< 4.5 rating) were retested in the metal trays in a randomized complete block design with 8 replications using the methods described above. Both resistant and susceptible checks were provided. Fall armyworm larvae used in this test were less than 6 months from the feral population.

RESULTS AND DISCUSSION

Based on the leaf-feeding damage for ratings 1 (4 DAI) and 2 (6 DAI), Tables 1 and 2 list the distribution of the Ethiopia and Yemen sorghum entries initially screened for FAW seedling leaf-feeding resistance. Approximately 100 sustained < 4.5 rating whereas Huerin Inta, the susceptible check, rated an average of 9.0 (plants dying or dead), and 1821 cm, the resistant check, rated an average of 6.0+. There appeared to be a higher frequency of Ethiopia entries resistance to leaf-feeding by the FAW than occurred in the Yemen entries.

Table 3 lists the FAW damage ratings for the best performing of the Ethiopian and Yemen sorghum entries in the second evaluation test. All entries rated approximately 4.0 (4 DAI) when Huerin Inta approached a leaf-feeding damage rating of 9.0. Therefore, FAW larvae were permitted to feed 2 d longer and all entries were rated again. Seventeen of the 100 entries rated significantly better than the resistant check, which had a mean rating of 6.25 on the first rating. Only 13 of the 100 performed significantly better than the resistant 1821 cm check for rating 2.

The entries with the highest resistance to FAW found in these Ethiopia and Yemen collections should now be used in a breeding program, but preferably after they are converted to day-sensitive entries. It is extremely difficult for these types of material to be used before they are transformed through the sorghum conversion program. Therefore, it is suggested that any new efforts of searching for pest

resistance in the sorghum collection should involve materials converted to day-length-sensitive and subtropical usable germplasm.

Table 1. Ratings distribution of Ethiopian sorghum plant introductions to seedling damage by larvae of the FAW.*.

Damage Ratings class	Damage ratings: No. in class		Damage ratings: Frequency	
	Rating 1	Rating 2	Rating 1	Rating 2
0-2	2	0	< 0.001	0
2-3	29	0	0.006	0
3-4	154	24	0.030	0.005
4-5	391	108	0.075	0.021
5-6	1002	379	0.193	0.073
6-7	1644	1017	0.316	0.196
7-8	1192	1579	0.229	0.304
8-9	786	2093	0.151	0.402
Total	5200			

* Based on a visual rating scale of 0-9 where 0 = no damage and 9 = plants dying or dead. Wiseman et al. (1966). Ratings 1 and 2 were generally made 4 and 6 days after infestation. Ratings were based on a plot rating of ca. 20-25 plants/entry.

Table 2. Ratings distribution of Yemen sorghum plant introductions to seedling damage by larvae of the FAW.*

Damage Ratings class	Damage ratings: No. in class		Damage ratings: Frequency	
	Rating 1	Rating 2	Rating 1	Rating 2
0-2	0	0	0	0
2-3	4	0	0.001	0
3-4	26	2	0.008	< 0.001
4-5	151	13	0.045	0.004
5-6	384	87	0.115	0.026
6-7	648	322	0.193	0.096
7-8	747	765	0.223	0.228
8-9	1390	2161	0.415	0.645
Total	3350			

* Based on a visual rating scale of 0-9 where 0 = no damage and 9 = plants dying or dead. Wiseman et al. (1966). See Table 1 for ratings 1 and 2. Ratings were made on a plot basis of ca. 20-25 plants/entry.

Table 3. Sorghum seedling damage ratings for resistance to the FAW in the best performing Ethiopia and Yemen lines in the second evaluation test.*

Entry	Rating 1	Rating 2
Huerin Inta (Susc. check)	9.00	9.00
1821 cm (Resist. check)	6.25	7.00
PI 452566	5.00	6.50
452554	5.00	6.00
474740	5.00	6.25
455018	5.00	6.00
453255	5.00	6.75
452825	4.75	7.00
456111	4.75	6.00
452571	4.75	6.00
452962	4.75	5.50
453281	4.75	6.50
453299	4.75	6.25
452987	4.75	5.75
452771	4.75	6.00
453130	4.50	5.25
457624	4.25	6.00
453356	4.25	5.50
454733	4.00	5.50

* Based on a visual rating scale of 0-9 where 0 = no damage and 9 = plants dying or dead. Wiseman et al. (1966). Ratings 1 and 2 were made 4 and 6 DAI. Ratings were based on a plot rating of ca. 20-25 plants/entry/replicate.

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RELATIONSHIP OF EUROPEAN CORN BORER[1] RESISTANCE IN SORGHUM TO HCN-p AND DIMBOA CONTENT IN LEAF AND SHEATH-COLLAR TISSUE[2]

W. D. Guthrie[3], F. A. Haskins[4], and H. J. Gorz[5]

Abstract: Both high- and low-HCN-p genotypes of sorghum, *Sorghum bicolor* (L) Moench, were resistant to the European corn borer (ECB), *Ostrinia nubilalis* (Hübner). It was not proven that HCN-p is or is not a chemical factor conditioning resistance to leaf feeding by first-generation ECB and resistance to sheath-collar feeding by second-generation ECB in sorghum. If HCN-p is a resistance factor, however, it is effective at very low levels because levels in the low-HCN-p genotypes were very low.

DIMBOA is not a chemical factor conditioning resistance in sorghum to the ECB because midwhorl leaves and sheath-collar tissue of both high- and low-HCN-p genotypes contained no DIMBOA.

Key Words: *Ostrinia nubilalis*, sorghum, host plant resistance, dhurrin, DIMBOA.

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During the period of egg deposition by first-generation European corn borers (ECB), *Ostrinia nubilalis* (Hübner), sorghum *Sorghum bicolor* (L) Moench, is in the whorl stage of plant development. Most larvae feed on leaf tissue in the moist area deep in the whorl of sorghum plants through 9 d after egg hatch. Most first-generation larval mortality of the ECB occurs during the first few days after egg hatch. Resistance to first-generation ECB on sorghum as in maize, *Zea mays* (L), is, therefore, leaf-feeding resistance; i.e., high antibiosis against first and second instars (Dharmalingam et al. 1984).

During the 1960's, F. F. Dicke (unpublished data) evaluated several varieties of sorghum under a low level of artificial ECB infestation (75 eggs/plant). During 1981-1983, Guthrie et al. (1985) evaluated 208 sorghum hybrids under a very high level of artificial ECB infestation (750 eggs/plant). All genotypes of sorghum were resistant to leaf feeding by first-generation ECB. The leaves on sorghum had pinholes (Fig. 1) indicating that some larvae fed for a short time on leaf tissue.

Beck and Lilly (1949) found that cyanogenetic content in whorl leaf tissue of sorghum plants is responsible, at least in part, for the high resistance of sorghum to ECB larvae during the whorl stage of plant development. One objective of our study was to determine if sorghum genotypes high in content of dhurrin [p-hydroxy-(S)-mandelonitrile-β-D-glucoside] and, thus, in hydrocyanic acid potential (HCN-p) in whorl leaf tissue are resistant to ECB and, conversely if sorghum genotypes with low levels of HCN-p are susceptible. DIMBOA (2, 4-dihydroxy-7-methoxy-(2H)-1,4-benzoxazin-3(4H)-one) is a chemical factor that conditions resistance in some genotypes of maize to leaf feeding by first-generation ECB (Tseng et al., 1984). A second objective of this study was to determine if whorl leaves of the sorghum genotypes contain DIMBOA.

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Fig. 1. A class 2 visual leaf rating showing pinhole feeding, typical of a highly resistant reaction to first-generation ECB on sorghum.

During the period of egg deposition by second-generation ECB, sorghum is in various stages of anthesis. Most larvae feed on sheath-collar tissue through 35 d after egg hatch. Resistance in sorghum as in maize, therefore, is resistance to sheath-collar feeding (Guthrie et al. 1984). Sorghum genotypes vary in degree of resistance-susceptibility when an infestation occurs during anthesis. Some genotypes are highly susceptible (Atkins et al., 1983; Ross et al. 1982). On susceptible genotypes of maize (infested during anthesis), ECB larvae tunnel throughout the whole plant. In contrast, ECB larvae rarely enter sorghum stalks below the peduncle, and only peduncles and heads are damaged. A third objective of our study was to determine if genotypes of sorghum containing different amounts of HCN-p and DIMBOA in sheath-collar tissue are resistant to second-generation ECB.

MATERIALS AND METHODS

The genotypes of sorghum evaluated were: (1) BKS8, a low-HCN-p (24 ppm, dry leaf tissue) parental line; (2) BN32, a high-HCN-p (858 ppm) parental line; (3) F_3-1, a line from a high-HCN-p (788 ppm) F_2 plant (28-4) from the cross, BSK8 × BN32; (4) F_2-2, a line from a low-HCN-p (33 ppm) F_3 plant (28-9) from the cross, BKS8 × BN32; (5) F_3-3, a line from a high-HCN-p (927 ppm) F_2 plant (36-7) from

the cross, BN32 × BKS8; (6) F_3-4, a line from a low-HCN-p (18 ppm) F_2 plant (36-10) from the cross, BN32 × BKS8; and (7) Check #2 (used only in 1986), an experimental grain sorghum hybrid known to be resistant to first-generation ECB and susceptible to second-generation ECB. The HCN-p values given are from plants grown at Lincoln, Nebraska, in 1984. A single major gene pair is primarily responsible for the large difference in HCN-p content between BKS8 and BN32. There were no obvious maternal effects, and F1's were generally intermediate in HCN-p level between the two parents, indicating that neither high nor low HCN-p was completely dominant (Gorz et al. 1986).

The genotypes were planted in single-row plots in two different experiments (randomized complete-block design with four replications for each experiment) at Ankeny, Iowa. Plots were planted 15 May 1985 and 17 May 1986. Rows were 3.3 m long, and distance between rows was 100 cm; stand was thinned to ca. 10 cm between plants when plants were ca. 15 cm high.

In the first experiment, six plants in each plot were artificially infested with 30 egg masses (ca. 750 eggs)/plant in five applications of six masses each spaced 1 d apart during the midwhorl stage of plant development. The same number of plants and egg masses were used for infestation during anthesis in a second experiment. Infestation and egg production techniques were reported by Atkins et al. (1983) and Guthrie et al. (1960, 1971).

In the first experiment, midwhorl leaves from six uninfested plants in each plot were taken for HCN-p analysis, and six plants were used for DIMBOA analysis; plants were cut above the growing point. For HCN-p analysis, whole leaves with midribs removed were used. For DIMBOA analysis, midwhorl leaves with 10 cm of tips removed were used. In the second experiment, sheath-collar tissue from each plot (at anthesis) was taken for HCN-p (six plants) and DIMBOA (six plants) analyses.

In the first experiment, leaf-feeding damage was rated on a plot basis 21 d after egg hatch, as described by Guthrie et al. (1960). In a 1-to-9 rating scale, classes 1 and 2 (Fig. 1) are highly resistant, classes 3 and 4 are resistant, classes 5 and 6 are intermediate, and classes 7 to 9 (Fig. 2) are susceptible. In the second experiment, cavity counts (cm of damage in peduncles and heads) were made from six plants in each plot 60 d after egg hatch as described by Atkins et al. (1983).

For HCN-p analysis, whorl leaves and sheath-collar tissue were cut into 2.5-cm pieces and dried for 4 h at 75°C. The dried samples were ground with a Wiley mill fitted with a 1-mm screen. A weighed portion from each plot was first extracted with water at room temperature to obtain dhurrin (the cyanogenic glucocide). An aliquot of each extract was treated with sodium hydroxide to hydrolyze the dhurrin, releasing cyanide into the solution. Cyanide content was then determined with the colorimetric reagents of Lambert et al. (1975) as described by Gorz et al. (1986).

For DIMBOA analysis, midwhorl leaves (75 cm in extended leaf height) and sheath-collar tissue from six plants in each plot were placed in plastic bags and frozen at -23°C until used. The frozen leaf and sheath-collar tissue were thawed, dried in an oven at 48°C, and ground into a fine powder for DIMBOA analysis. The chemical determinations were actually for MBOA (6-methoxybenzoxazolinone), expressed as milligrams of MBOA per gram of plant tissue. We used a modification of the procedures reported by Klun and Robinson (1969). Because DIMBOA is

chemically labile and decomposes stoichiometrically to MBOA, DIMBOA concentrations can be determined by chemical analysis of dried plant tissue for MBOA. Details of the MBOA extraction procedure were reported by Tseng (1984).

Fig. 2. A class 9 visual leaf rating showing numerous elongated lesions, typical of a highly susceptible reaction to first-generation ECB on maize.

For analysis of variance of plot means, total sum of squares for leaf-feeding ratings, cavities in peduncles and heads, HCN-p of leaf tissue, and HCN-p of sheath-collar tissue for each year were partitioned into components for replications (3df), genotypes (5 df in 1985, 6 df in 1986), and error (15 df in 1985, 18 df in 1986). LSD ($P < 0.05$) values were calculated as described by Steel and Torrie (1960) to determine the level of significance of differences between means.

RESULTS AND DISCUSSION

The analysis of variance showed no significant difference in leaf-feeding damage among genotypes in 1985. In 1986, BN32 and check #2 had significantly

less leaf-feeding damage than did the other five genotypes, but the difference was of little practical importance because, during the 2-year period, both high- and low-HCN-p genotypes of sorghum rated highly resistant (class 2) or resistant (class 4) to leaf feeding by first-generation ECB (Table 1). As in previous studies (Guthrie et al., 1985), whorl leaves of the sorghum genotypes in the present study had pinholes (Fig. 1) indicating that some larvae fed for a short time on leaf tissue, similar to those on resistant genotypes of maize. None of the genotypes had numerous elogated lesions typical of susceptible genotypes of maize (Fig. 2).

Table 1. ECB leaf-feeding damage, ECB peduncle and head damage, and HCN-p levels in whorl leaves and sheath-collar tissue in seven genotypes of sorghum, Ankeny, Iowa.

	Leaf feeding ratings*		HCN-p in leaf tissue‡		Cavities (cm)†		HCN-p in sheath collar tissue‡	
Genotype	1985	1986	1985	1986	1985	1986	1985	1986
1. BKS8	2.3	4.0	31	45	1.5	1.0	45	18
2. BN32	2.0	2.0	752	465	11.0	6.0	239	65
3. F_3-1	2.0	4.0	689	300	2.8	1.4	298	104
4. F_3-2	2.0	4.0	95	69	3.0	3.4	54	12
5. F_3-3	2.0	3.5	711	302	7.5	4.9	214	73
6. F_3-4	2.0	4.0	38	49	2.3	6.6	52	12
7. Check #2		2.3		475		15.4		60
LSD 0.05		0.8	94	76	4.5	2.6	52	26

* Leaf-feeding damage was rated on a 1-to-9 scale, with 1 indicating no damage and 9 indicating extensive damage to leaf tissue 21 d after egg hatch.

† Cavities (cm of damage in peduncles and heads) were determined 60 d after egg hatch. There were no cavities in stalks below the peduncle.

‡ mg HCN-p per kg of dry midwhorl leaf or sheath-collar tissue.

As expected, there were large differences among genotypes for HCN-p of midwhorl leaf tissue. Midwhorl leaves of most genotypes had higher HCN-p in 1985 than in 1986, but the differences between high- and low-HCN-p genotypes in both years were great (Table 1).

On genotypes of sorghum susceptible to second-generation ECB, the larvae survive on sheath-collar tissue (Fig. 3) through 35 d after egg hatch (Guthrie et al. 1984) and then enter peduncles and heads, causing extensive damage (Fig. 4). Cavity counts in peduncle and heads can be used, therefore, to measure resistance-susceptibility. Cavities (cm of damage) in peduncles and heads of high- and low-HCN-p genotypes of sorghum ranged from 1.5 to 11.0 in 1985 and from 1.0 to 6.6 in 1986. The susceptible check contained 15.4 cm of damage in 1986 (Fig. 4).

Genotypes with high levels of HCN-p in midwhorl leaves also had relatively high levels in sheath-collar tissue, and genotypes with low HCN-p levels in midwhorl leaves had low levels in sheath-collar tissue. Midwhorl leaves had more than twice the HCN-p of sheath-collar tissue. Sheath-collar tissue of all genotypes was higher in HCN-p in 1985 than in 1986 (Table 1).

We did not prove that HCN-p is or is not a chemical factor conditioning resistance to leaf feeding by first-generation ECB and resistance to sheath-collar feeding (as measured by damage in peduncles and heads) by second-generation

ECB. If HCN-p is, however, a resistant chemical factor, as indicated by Beck and Lilly (1949), it is effective at levels that are no higher than those observed for the low-HCN-p sorghum genotypes.

Fig. 3. Sheath-collar feeding damage by second-generation ECB on sorghum when infested at anthesis.

DIMBOA is not a chemical factor conditioning resistance in sorghum to the ECB because midwhorl leaves and sheath-collar tissue of both high- and low-HCN-p genotypes contained no DIMBOA. Guthrie et al. (1985) also found no DIMBOA in midwhorl leaves of four sorghum hybrids.

Fig. 4. Peduncle and head damage on a susceptible genotype of sorghum caused by second-generation ECB when infested at anthesis.

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THE EFFICACY AND USE OF AMITRAZ FOR THE CONTROL OF HOG LICE[1]

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Abstract: An evaluation was made of three concentrations of amitraz TAKTIC® 12.5% emulsifiable concentrate for controlling hog lice, *Haematopinus suis* (L.). Comparisons were made of single treatments and two treatments 14 days apart. A follow-up study was conducted on three privately owned farrow to finish swine facilities using a 0.05% rate of amitraz in an integrated treatment program.

In the initial study, all rates tested (0.025%, 0.05%, and 0.1%) reduced lice numbers on animals. However, a second treatment, at 14 days, was necessary to provide 100 percent control. In the follow-up study, the integrated treatment program consisted of an initial whole-herd treatment phase followed by scheduled treatments applied in conjunction with daily management of the operations. Total hog lice control ranged from 20 weeks at one farm through 53 weeks at another farm.

Key Words: Amitraz, hog lice, *Haematopinus suis*.

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Hog lice, *Haematopinus suis* (L.), along with sarcoptic mange mites, *Sarcoptes scabiei* var. *suis* (De Geer), are generally recognized as the most economically important ectoparasites of swine. In a survey of Indiana veterinarians, it was estimated that 51.5% of the farms visited had swine infested with lice (Wooten-Saadi et al. 1987). It is estimated that $40 million is lost annually in the United States by producers due to hog lice alone (Anonymous 1979). A 1979 survey of pesticide use in Nebraska indicated that 75% of pesticides used on swine were for hog louse control (Campbell and Kamble 1981). In a survey of market-weight swine in Indiana conducted during 1980 to 1981, hog lice were found on 18% of all animals examined (Wooten-Saadi et al. 1987).

Hog lice pass their entire life cycle on the host. Their blood-sucking activity results in an apparent irritation and discomfort to swine. Davis and Williams (1986) demonstrated physiological changes (blood chemistry) in growing pigs due to hog lice infestation.

Controlling hog lice on swine is directed at 1) eliminating both active adults and nymphs from the animals, 2) eliminating nymphs hatching from attached eggs, and 3) eliminating re-introduction of lice into the herd. Control should also be directed toward prevention of transmission of lice infestations not previously controlled in a herd. These steps of treatment and control should be integrated with the routine daily tasks of the producer. Various insecticides have been used for hog lice control (Williams et al. 1980) but many of these compounds have since been removed from the market or are marginal in effectiveness.

This report describes the use of a new insecticide, for controlling hog lice in swine. Amitraz (TAKTIC®) is a triazapentadiene which has shown high parasiticidal

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activity against various ectoparasites of livestock (Curtis 1985). This report is based on two studies: an initial study evaluating different rates of active ingredient and comparing single and double treatments; and a follow-up study on commercial swine herds in Indiana establishing an integrated program using amitraz to control hog lice. The control of sarcoptic mange in these studies was described by Gaafar et al. (1986).

MATERIALS AND METHODS

Study 1

Forty pigs, averaging 7.4 kg each, were divided into 8 similar weight groups of 5 pigs each and placed in separate pens. The treatments evaluated were amitraz 12.5% emulsifiable concentrate (EC) sprays at 0.025%, 0.05% (labeled rate), and 0.1% active ingredient (ai) comparing a single treatment (3 groups) with two treatments (3 groups). Two groups were used as untreated controls and sprayed with inert carrier solvent at 0.05% as a single treatment and two treatments. In the groups given two treatments, the second treatment was made 14 days after the initial spraying.

Treatments were applied with a 7.57 liter B & G hand-held pressure sprayer equipped with a Teejet #6502 fan-pattern nozzle at a rate of 1 to 2 liters of finished spray per animal in each group. Thorough body coverage was achieved including the inner surface of the ear pinna, the axillae and groin.

Population assessment of hog lice was obtained by making total body counts on each animal. Louse infestations were established by placing 50 to 100 lice per animal 3 weeks pre-treatment. After pre-treatment counts, louse populations were monitored on the pigs twice weekly for 4 weeks. Louse count data obtained were statistically analyzed in a one-way analysis of variance. Mean counts each day were compared using Duncan's New Multiple Range Test ($P < 0.05$) (Duncan 1955).

Sudy 2

In this study, three commercial farrow to finish swine operations were selected and a treatment program using amitraz was devised to control the hog lice on the animals at these farms. The swine herds used in this study were selected because they had a history of problems with hog lice and sarcoptic mange during previous years. Although these herds had been previously treated periodically with various chemicals, no animals had been treated for at least 2 months prior to initiation of the present trials.

The spray material for each farm was prepared from a stock solution containing 12.5% ai amitraz. Before spraying, all feeders and waters were removed, emptied or tightly covered to prevent contamination. All animals in each herd were initially sprayed with an emulsion of 0.05% ai in water. A second application of the same concentration of spray was made 7-14 days later. All animals were thoroughly sprayed to runoff with particular attention being given to the ear pinna, the axillae and groin. Approximately 2 liters of spray material were used on each animal with more solution used on larger sows and boars and less on the smaller animals. Newborn pigs, up to 3 weeks of age, were dipped in the same concentration of spray solution. The premises, including the walls, pen dividers, and equipment in the pens, were also sprayed. The equipment used for spraying consisted of power sprayers with coarse nozzles and wand spray handles.

Following the two comprehensive sprayings, a maintenance prevention program was initiated in each herd. This program focused on preventing infestation of the young piglets. At weaning and before piglets from various litters were mixed together, they were dipped in a 0.05% ai emulsion of amitraz. The dip solution was renewed after each 50 piglets. Boars were also sprayed once every 3 months. New animals added to the herd during this study were isolated and sprayed twice (at 7-10 day intervals) before being placed with the rest of the animals. Sows in late pregnancy were sprayed as they were being moved to the farrowing crates and again in the farrowing pen after weaning their piglets and before transferring to the gestation stalls.

Farm A, located in White County, Indiana, had approximately 2800 animals. All the animals in this herd were sprayed twice with amitraz. The first spraying was on 20, 23, and 24 May 1983 followed by a second treatment on 27, 31 May and 1 June 1983.

Farm B, located in Carroll County, Indiana, had approximately 2300 animals. The first spraying was on 20 June 1984 and the second spraying was made on 5 July 1984.

Farm C, located in Jasper County, Indiana, had approximately 2400 animals. The first spraying was on 26 June 1984 and the second spraying on 10 July 1984.

Before the initial sprayings, 20 animals from each farm were examined for hog lice. Five animals were randomly selected from each of four age groups of pigs (sows, nursery stock, early feeder pigs, and late feeder stage pigs). Lice counts consisted of total body counts on each animal. After the second spraying of the animals examinations for the presence of lice were made on 20 animals, chosen as above, at 1 week, 2 weeks and monthly thereafter until July 1985.

RESULTS AND DISCUSSION

Study 1

The mean numbers of hog lice from each treatment group over the duration of the study are shown in Table 1. It is evident from the data that amitraz was effective in reducing lice numbers on the animals treated with each spray concentration. However, after the first spray treatment, live lice were observed on the animals at 7 days post-treatment in all amitraz treatment groups. The majority of these lice were nymphs, probably having hatched from eggs after the initial treatments were made. In each amitraz group where a second treatment was made, no lice were found on the pigs after two weeks. Lice numbers in the groups where one treatment was made continued to increase in numbers for the remainder of the trial. However, pigs in all three groups treated once still had fewer lice on them than did pigs in the untreated groups after 4 weeks.

Study 2

Control of lice was achieved at each farm following the initial treatments (Table 2). On Farm A, all the animals examined before treatments had lice infestations. It was 20 weeks later before lice were again found on the animals sampled. Lice were found on most of the sampling dates thereafter. At Farm B, lice first reappeared at week 24, on two lactating sows, and were observed again at weeks 47 and 55. On Farm C, total lice control was achieved for the duration of the 53 weeks during which counts were made.

Table 1. Mean numbers of *Haematopinus* suis on swine treated with different concentrations of amitraz.*

	Single Treatment				1 and 14 Day Treatment			
Days Post-trt	Amitraz 0.025%	Amitraz 0.05%	Amitraz 0.1%	Control	Amitraz 0.025%	Amitraz 0.05%	Amitraz 0.1%	Control
0	13.0ab	21.0ab	6.4a	19.0bc	24.6bc	16.2ab	19.8ab	38.6c
3	0a	0a	0a	9.0b	0a	0a	0a	18.6c
7	6.4ab	1.2a	2.4a	17.6b	2.2a	0.6a	1.8a	44.8c
10	21.4ab	12.6ab	1.6a	32.0b	20.0ab	13.4ab	5.8ab	109.4c
14	34.4ab	21.8ab	5.4a	66.0b	34.0ab	17.6ab	25.4ab	247.6c
17	35.2a	35.8a	12.4a	95.6b	0a	0a	0a	358.0c
21	36.8a	87.8b	21.8a	141.0c	0a	0a	0a	443.2d
24	38.5ab	67.2b	23.6ab	141.0c	0a	0a	0a	495.8d
27	39.6a	62.2a	19.2a	202.4b	0a	0a	0a	510.0c

* Means within rows followed by the same letter are not significantly different ($P < 0.05$) - Duncan's New Multiple Range Test.

In these studies, it was demonstrated that amitraz is an effective insecticide for controlling hog lice on pigs, but only if applied twice with a 7-14 day interval between sprayings and if the maintenance program is strictly followed. This interval is needed to allow for nymphs to hatch from eggs. In the commercial farms used in Study 2, the maintenance program was effective in providing longer term lice control. It is essential, however, that the maintenance program be followed closely to insure proper control of lice. Single applications to new animals brought into the herds at Farms 1 and 2 probably led to the reinfestation of the herds.

Comparisons of pre-treatment and post-treatment production records at these farms, as presented by Gaafar et al. (1986), showed 10-15 days earlier maturity in finishing pig market weight, an average increase of 2.1 weaned pigs/litter, and decreases in piglet processing and nursing mortality following implementation of the amitraz treatment program for controlling both lice and sarcoptic mange mites.

Table 2. Mean percentage of swine from commercial farms infested with *Haematopinus suis* following treatments with amitraz.

Farm A		Farm B		Farm C	
Week	% Infested With Lice	Week	% Infested With Lice	Week	% Infested With Lice
0	100	0	20	0	5
3	0	3	0	3	0
4	0	4	0	4	0
8	0	6	0	7	0
12	0	10	0	11	0
16	0	15	0	16	0
20	10	20	0	19	0
24	5	24	10	24	0
28	20	26	0	28	0
31	35	32	0	32	0
36	0	35	0	36	0
39	40	43	0	40	0
44	25	47	40	44	0
49	5	51	0	48	0
53	35	55	10	53	0
57	40				
62	60				
66	0				
70	35				
75	60				
79	30				
82	35				
87	20				
92	60				
99	30				
103	20				
107	15				
111	0				

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CARBOSULFAN, FONOFOS, AND LINDANE WHEAT SEED TREATMENTS FOR CONTROL OF SUGARBEET WIREWORM[1,2]

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Abstract: Results from greenhouse and field trials showed that 'Daws' and 'Dirkwin' wheat seed treated with carbosulfan (1.25-10.0 g AI/kg seed) protected plants from damage by the sugarbeet wireworm (SBW), *Limonius californicus* (Mannerheim), and caused SBW mortality equal to or greater than lindane-treated seed. Fonofos-treated seed caused SBW mortality and protected plants, but phytotoxicity resulted in reduced plant survival. Lindane, though less toxic to SBW than carbosulfan or fonofos, still protected plants from SBW.

Key Words: Sugarbeet wireworm, *Limonius californicus*, wheat, carbosulfan, fonofos, lindane, seed treatment, control.

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The sugarbeet wireworm (SBW), *Limonius californicus* (Mannerheim), is one of the most economically important species of wireworms on wheat (Lane 1935) and is among the most common species in the Pacific Northwest (Toba et al. 1985). It is commonly found in irrigated cropland and in areas where annual precipitation is > 46 cm (Lane 1935).

Soil treatment with insecticides or fumigants may effectively control wireworms (Onsager et al. 1966); however, the high cost of such treatment limits their use in small grain pest management. Conversely, insecticide seed treatment is generally cost-effective in protecting seeds and young plants because of the small amounts of insecticide used and the low cost of application. In the past, a number of chlorinated hydrocarbon insecticides were used for seed treatment of small grains (Lange 1959), but presently, only lindane is registered for use on wheat seed for wireworm protection. Treatment of wheat seed with insecticides other than chlorinated hydrocarbons has been reported by Harwood et al. (1957), Golightly et al. (1969), Kulash (1953), and Perkins and Harwood (1979a, 1979b). In general, carbamate and organophosphate treatments have resulted in phytotoxicity or insufficient protection from wireworms. One exception is carbosulfan-treated seed which was shown to be as effective as lindane-treated seed against *Ctenicera destructor* (Brown) in Montana (Morrill 1984).

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[2] This paper reports the results of research only. Mention of proprietary products or pesticides does not constitute a recommendation for use by the USDA, nor does it imply registration under FIFRA as amended. Scientific paper no. 7854. College of Agriculture and Home Economics Research Center, Pullman. Project no. 0337. Idaho Agricultural Experiment Station paper no. 87746.

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Our objective was to evaluate the effectiveness of carbosulfan, fonofos, and lindane as wheat seed treatments for control of SBW, using winter and spring wheat varieties.

MATERIALS AND METHODS

Wheat Varieties

Seed of the spring wheat variety 'Dirkwin' was used in all spring field plantings, while seed of the winter wheat variety 'Daws' was used in all fall field plantings; both varieties were evaluated in greenhouse trials. The two varieties are of the soft white wheat type, a type widely grown in the Pacific Northwest.

Seed Treatment and Germination Tests

All seed was slurry treated according to Pike and Glazer (1980). The treatments consisted of various rates of carbosulfan (Advantage 25ST), fonofos (Dyfonate 4S or 5S), and lindane; lindane 40F (0.3 g AI/kg of seed) was used as a standard for comparison. Table 1-4 show the treatment rates evaluated. Unless otherwise indicated, all seeds were treated with Vitavax-200 fungicide (carboxin:thiram, 17%:17% flowable) at a rate of 2 ml formulation/kg of seed.

Seed germination tests were conducted by the Washington State University Seed Laboratory, Pullman, according to standard germination test procedures outlined by the Association of Official Seed Analysts (Copeland 1978). The germination for each treatment rate was evaluated from 4 samples of 100 seeds, each taken at random from a single seed source of 'Daws' or 'Dirkwin', with sprouts rated after 7 days.

Greenhouse Trials

Trials consisting of potted wheat seeds infested with SBW were conducted in the greenhouse to compare carbosulfan, lindane and fonofos seed treatment on plant survival and SBW mortality. Seeds were planted in moist soil in plastic pots (height 16 cm, diameter 14 cm at top and 12 cm at bottom) with 32-mesh screen attached to the bottom to insure wireworm confinement. Field-collected SBW (10-20 mm long) were introduced into each pot. Trials varied in the number of seeds/pot planted, number of wireworms/pot introduced, and wheat variety. The respective values for trials 1, 2, and 3 were: seeds/pot planted 4, 10, and 5; wireworms/pot 4, 5, and 10; wheat varieties 'Daws', 'Daws', and 'Dirkwin'. Each pot was considered a replicate, and each treatment was replicated five times. After 3 wk, the number of surviving plants and wireworms were recorded.

Field Trials

Trials were conducted at sites near Walla Walla and Prosser, WA, and Arbon Valley and Nampa, ID, on land naturally infested with wireworms. Wireworm infestation levels were determined by taking 20 soil core samples with a 15-cm diameter posthole digger to a depth of 45 cm within a 0.5 ha. area/site, and extracting the wireworms by sifting the soil through a series of screens. The number of damaged plants, plant stand, and grain yield were used to evaluate treatment performance. Wireworm damaged plants included dead or dried seedlings lying on the soil surface, or plants with distinctly weak or dead central tillers. The count on damaged plants and plant stands were recorded at 10 to 48 days postplanting,

depending upon the trial, and were based on 2 or 3 m of each row of 2-row plots or two center rows of 4-row plots (plot size/trial site described below).

At Prosser, where trials were conducted in both fall and spring, soil was classified as a Starbuck- Scooteney silt loam. In fall, 'Daws' seed was planted on 7 September 1982 in 4-row plots, 3.7 m long. In spring, 'Dirkwin' seed was planted on 22 March 1983 in 2-row plots, 2.1 m long. In both of these trials, treatments were replicated five times in a randomized complete block design; row spacing was 30.5 cm. Seeding rate was 41 seeds/m. Trials were not harvested because of inadvertent damage by livestock in late season.

At the Walla Walla site, soil was classified as a Yakima silt loam. 'Dirkwin' seed was planted on 24 April 1982 in 2-row plots, 3.7 m long. Treatments were replicated four times in a randomized complete block design; row spacing was 30.5 cm. Seeding rate was 41 seeds/m. The trial was harvested on 12 August 1982 using a plot combine.

In Idaho, two trials were conducted with spring-planted 'Dirkwin'. At Arbon Valley, where the soil was classified as Dale silt loam, seeds were planted on 5 May 1982. At Nampa, where the soil was classified as Purdam silt loam, seeds were planted on 23 March 1982. At both sites, plots consisted of four rows, 3.7 m long and 30.5 cm row spacing; treatments were replicated four times in a randomized complete block design. The seeding rate was 34 seeds/m. The yields were harvested from the two center rows in early- to mid-August 1982 using a plot combine.

Data were analyzed by analysis of variance and means were separated by Duncan's (1955) multiple range test. In greenhouse trials, percentages were transformed to arcsine $\sqrt{X}$ before analysis; actual means are shown in the table (Table 2).

RESULTS AND DISCUSSION

Germination and Sprouting

Carbosulfan treatment of 'Daws', at rates of 2.5 to 10.0 g AI/kg of seed (5 months posttreatment), resulted in some reductions in normal sprouts, with corresponding increases in weak and abnormal sprouts, compared to lower carbosulfan rates (0.63-1.25 g AI/kg of seed) and untreated controls (Table 1). With 'Dirkwin' seed, 0.3 or 10.5 months posttreatment, normal sprouting was not significantly affected by carbosulfan. All rates of fonofos, except 2.0 g AI/kg of seed (10.5 months posttreatment), significantly reduced the normal sprouts compared with the untreated controls. Increases in abnormal sprouts were also more evident with the fonofos. The lindane standard ('Daws', 5 months posttreatment; 'Dirkwin', 10.5 months posttreatment) did not significantly affect germination compared with the fungicide or untreated controls.

Greenhouse Trials

In Trial 1 (T_1, Table 2), insecticide treatment of 'Daws' seed caused no differential effects on potted plant survival. All insecticide treatments led to significantly higher survival of plants than untreated seed, even though in the standard laboratory germination tests, weak and abnormal sprouts were increased by carbosulfan (2.5 to 10.0 g AI/kg seed) (Table 1). However, SBW mortalities with treatment were relatively low; maximum was 25%.

Table 1. Effect of various insecticide seed treatments on wheat germination and sprouting.

Insecticide treatment	Rate (g AI/kg of seed)	Seed treated with fungicide†	Wheat variety	Percent sprouting ± SE*			
				Normal sprouts	Weak sprouts‡	Abnormal sprouts§	Ungerminated seed
Seed Evaluated 5 Months Posttreatment							
Carbosulfan 25ST	0.63	Yes	Daws	90.3 ± 1.5a	2.3 ± 0.5f	5.0 ± 1.5d	2.5 ± 0.6b
Carbosulfan 25ST	1.25	Yes	Daws	85.8 ± 2.0b	5.8 ± 1.4ef	7.5 ± 1.5cd	1.0 ± 0.6b
Carbosulfan 25ST	2.50	Yes	Daws	76.5 ± 1.3c	9.8 ± 0.9de	10.0 ± 0.9bcd	3.8 ± 1.0b
Carbosulfan 25ST	5.00	Yes	Daws	72.3 ± 3.4c	14.3 ± 2.5d	9.3 ± 1.9bcd	4.3 ± 0.3b
Carbosulfan 25ST	7.50	Yes	Daws	60.0 ± 1.7d	20.8 ± 2.1c	15.0 ± 0.9bc	4.3 ± 0.6b
Carbosulfan 25ST	10.00	Yes	Daws	36.5 ± 5.6f	21.8 ± 1.5c	32.0 ± 5.4a	9.8 ± 1.8a
Fonofos 4S	12.00	Yes	Daws	50.0 ± 2.3e	32.3 ± 2.3b	14.5 ± 2.1bc	3.3 ± 1.0b
Fonofos 4S	18.00	Yes	Daws	14.5 ± 4.9g	60.8 ± 4.5a	16.0 ± 3.0b	8.8 ± 1.5a
Lindane 40F	0.30	Yes	Daws	80.0 ± 3.3bc	6.8 ± 1.3ef	9.8 ± 2.4bcd	3.5 ± 1.0b
Untreated		Yes	Daws	88.0 ± 0.8ab	4.8 ± 0.8ef	6.0 ± 1.0d	1.3 ± 0.3b
Untreated		No	Daws	86.3 ± 1.0ab	2.5 ± 1.0f	8.8 ± 0.9bcd	2.5 ± 1.0b
Seed Evaluated 0.3 Months Posttreatment							
Carbosulfan 25ST	1.25	Yes	Dirkwin	97.3 ± 0.3a	1.8 ± 0.3e	0.5 ± 0.3d	0.5 ± 0.3b
Carbosulfan 25ST	2.50	Yes	Dirkwin	94.5 ± 0.6a	1.3 ± 0.5e	2.5 ± 0.6cd	1.8 ± 0.8b
Carbosulfan 25ST	5.00	Yes	Dirkwin	93.0 ± 2.0a	2.3 ± 1.0de	2.5 ± 0.6cd	2.3 ± 0.9b
Fonofos 5S	2.00	Yes	Dirkwin	82.8 ± 2.6b	7.8 ± 2.5d	7.3 ± 1.1c	2.3 ± 1.3b
Fonofos 5S	4.00	Yes	Dirkwin	60.3 ± 3.0c	24.3 ± 1.9c	12.3 ± 2.2b	3.3 ± 0.5b
Fonofos 5S	8.00	Yes	Dirkwin	28.0 ± 1.1d	34.0 ± 2.0b	34.5 ± 1.2a	3.5 ± 1.0b
Fonofos 5S	12.00	Yes	Dirkwin	11.0 ± 1.8e	43.3 ± 3.8a	37.0 ± 3.6a	9.0 ± 2.1a
Untreated		Yes	Dirkwin	95.8 ± 0.6a	0.8 ± 0.5e	1.5 ± 0.6d	2.0 ± 0.4b

				Seed Evaluated 10.5 Months Posttreatment			
Carbosulfan 25ST	2.50	Yes	Dirkwin	96.0 ± 0.8a	1.5 ± 0.0d	1.5 ± 0.5bc	1.0 ± 0.6b
Carbosulfan 25ST	5.00	Yes	Dirkwin	94.5 ± 1.3ab	2.3 ± 0.6cd	1.8 ± 0.9bc	1.5 ± 0.6ab
Carbosulfan 25ST	10.00	Yes	Dirkwin	93.8 ± 1.3ab	3.8 ± 1.2bcd	1.0 ± 0.7c	1.5 ± 1.2ab
Fonofos 5S	2.00	Yes	Dirkwin	90.0 ± 0.4bc	2.8 ± 0.9bcd	4.5 ± 0.6ab	2.8 ± 0.9ab
Fonofos 5S	4.00	Yes	Dirkwin	88.0 ± 2.1c	5.3 ± 1.2b	6.0 ± 1.5a	0.8 ± 0.5b
Fonofos 5S	8.00	Yes	Dirkwin	71.5 ± 3.1d	17.8 ± 1.1a	6.8 ± 1.7a	3.5 ± 0.9a
Lindane 40F	0.30	Yes	Dirkwin	91.4 ± 0.8abc	3.3 ± 0.7bcd	2.8 ± 0.6bc	2.4 ± 0.6ab
Untreated		Yes	Dirkwin	93.8 ± 0.6ab	2.5 ± 0.9bcd	2.0 ± 0.6bc	1.8 ± 0.6ab

* Column means followed by the same letter within each seed lot are not significantly different at $P = 0.05$ (Duncan's [1955] multiple range test).

† Vitavax-200 fungicide; rate: 2 ml/kg of seed.

‡ Shoot less than 3 cm long; seedling otherwise normal.

§ Split, empty coleoptile; some sprouts too small to evaluate, swollen shoot.

Table 2. Effect of carbosulfan, lindane, and fonofos seed treatment on seed germination (laboratory analysis), and plant survival and SBW mortality in greenhouse pot trials.

Seed treatment (g AI/kg seed)*	% Germination		% Plant survival†			% Wireworm mortality†		
	'Daws'	'Dirkwin'	T1	T2	T3	T1	T2	T3
Carbosulfan 0.63	93a‡	—	55a	65b	—	0a	24bc	—
Carbosulfan 1.25	—	99a	—	—	92a	—	—	42a
Carbosulfan 2.50	86a	96ab	80a	74b	84ab	25a	28ab	44a
Carbosulfan 5.00	87a	95ab	65a	83ab	88ab	25a	32ab	44a
Carbosulfan 10.00	58b	—	—	89a	—	—	56a	—
Fonofos 2.00	—	91b	—	—	53b	—	—	44a
Fonofos 4.00	—	85c	—	—	80ab	—	—	38a
Fonofos 8.00	—	62d	—	—	71ab	—	—	52a
Fonofos 12.00	—	54e	—	—	72ab	—	—	40a
Lindane 0.30	87a	95ab	70a	63b	60ab	5bc	44ab	18b
Vitavax-200	93a	97a	35ab	14c	4c	10abc	8c	6c
Untreated	89a	—	0b	—	—	0c	—	—

* All seeds were treated with Vitavax-200 (2.0 ml/kg seed), except in the untreated.

† 'Daws' winter wheat was evaluated in T1 and T2; 'Dirkwin' spring wheat was evaluated in T3.

‡ Column means followed by the same letter are not significantly different at $P = 0.05$ (Duncan's [1955] multiple range test).

In Trial 2 (T_2, Table 2), 'Daws' seed treated with the highest rate of carbosulfan (10.0 g AI/kg seed) and infested with SBW led to significantly higher plant survival than the lower rates (0.6 to 2.5 g AI/kg seed) or the lindane treatment. SBW mortality differed little among the insecticide-treated seeds, and those of all but the lowest rate of carbosulfan were significantly higher than that of the fungicide control.

In Trial 3 (T_3, Table 2), germination of 'Dirkwin' seed was affected by fonofos, but not by carbosulfan or lindane. All insecticide treatments produced significantly higher plant survival than the fungicide control. However, fonofos phytotoxicity was evident by the generally lower plant survival compared to carbosulfan-treated seed, although the differences were not significant. All insecticide treatments produced significantly higher SBW mortality than the fungicide control, but lindane was less toxic to SBW than either carbosulfan or fonofos.

These greenhouse pot trials demonstrated that carbosulfan is an effective seed treatment, of equal or higher efficacy than lindane in protecting wheat plants from wireworm damage. Fonofos, although relatively effective in controlling SBW, showed some phytotoxicity. Lindane was less effective in controlling wireworms than fonofos; nevertheless, it protected plants with little or no phytotoxicity. The afforded plant protection by lindane, with only low wireworm kill, suggested it repelled or was sublethal to the wireworms. Repellency to lindane-treated seed has been demonstrated previously. Long and Lilly (1958) showed that corn seed treated with lindane caused feeding inhibition or orientation of *Melanotus communis* (Gyllenhal) away from the treated seed.

Field Trials

At Prosser (Table 3), SBW density was estimated at 20.2/m². 'Daws' seed treated with lindane and the low rates of carbosulfan (0.6 to 5.0 g AI/kg seed) caused no adverse effects on germination compared with fungicide control. Fonofos and the highest rate of carbosulfan reduced seed germination. The number of SBW-damaged plants in plots of insecticide-treated 'Daw' seeds did not differ from each other (21 days postplanting), but were all significantly lower than the fungicide control. In the trial with spring-planted 'Dirkwin' (29 and 35 days postplanting), results followed a trend similar to the fall 'Daws' trial.

At Walla Walla (Table 3), SBW density was estimated at 48.4/m². Treatment of 'Dirkwin' seed with carbosulfan or lindane caused no adverse affects on germination, reduced plant damage, and produced significantly higher yields than the fungicide control. There were numerical increases in yield with increased rates of carbosulfan, but these were not significant increases.

At Arbon Valley (Table 4), the wireworm species were SBW (92%) and *Aeolus mellillus* (Say) (8%), and the density was estimated at 85.0/m². Plant stands in plots of insecticide-treated seeds did not differ from each other, but all were significantly higher than in the fungicide control. Similarly, yields in plots of insecticide-treated seeds were numerically higher than that of the fungicide control, although the difference was not significant.

At Nampa (Table 4), the SBW density was estimated at 52.1/m². No significant differences in plant stands were found between treatments. However, plots of carbosulfan-treated seeds produced significantly higher yields than that of the fungicide control, but not lindane-treated seeds. Also, plots with the lowest rate of carbosulfan-treated seeds produced significantly higher yield than that of lindane-treated seeds.

Results from the greenhouse and field trials were complimentary: wheat seed treatment with carbosulfan (up to 10 g/AI/kg seed) protected plants from SBW at levels comparable to or higher than lindane. Fonofos-treated seeds provided SBW plant protection, but not without some phytotoxic injury.

Another injurious species on wheat in the Pacific Northwest is the Great Basin wireworm (GBW), *Ctenicera pruinina* (Horn), which is commonly found in dryland farming areas where annual participation is < 38 cm (Lane 1935). Greenhouse tests showed that lindane-treated seeds afforded protection from GBW equal to that from SBW (Toba, unpublished data); therefore, similar protection can be expected with carbosulfan-treated seeds.

Table 3. Effect of carbosulfan, fonofos and lindane seed treatment on seed germination (laboratory analysis), and plant stand or SBW damaged plants of 'Daws' winter (fall 1982) or 'Dirkwin' spring wheat (1983) at Prosser, WA, and 'Dirkwin' spring wheat (1982) at Walla Walla, WA, in field trials.

			Prosser†			Walla Walla‡		
	% Germination		Wireworm damaged plants/m row, DPP§			Wireworm damaged plants/m row, DPP		
Seed treatment (g AI/kg seed)*	'Daws'	'Dirkwin'	21	34	48	10	19	Yield (kg/ha)
Carbosulfan 0.63	93a‖	—	0.4a	—	—	—	—	—
Carbosulfan 1.25	92a	99a	0.6a	0.3a	0.9a	—	—	—
Carbosulfan 2.50	86ab	96ab	0.3a	0.0a	1.0a	5.4a	10.1a	4277a
Carbosulfan 5.00	87ab	96ab	0.3a	0.0a	0.9a	3.5a	10.4a	4835a
Carbosulfan 10.00	58c	96ab	0.2a	0.0a	1.0a	3.0a	8.0a	5420a
Lindane 0.30	87ab	95ab	0.0a	0.0a	1.0a	2.3a	7.7a	5857a
Fonofos 2.00	82b	91b	0.1a	0.0a	1.1a	—	—	—
Vitavax-200	93a	97a	1.3b	3.7b	7.3b	13.8b	25.1b	2098b

* All seeds were treated with Vitavax-200 (2.0 ml/kg seed).

† One trial of 'Daws' wheat evaluated 21 days postplanting; second trial of 'Dirkwin' wheat evaluated 34 and 48 days postplanting.

‡ One trial of 'Dirkwin' wheat.

§ DPP denotes days postplanting.

‖ Column means followed by the same letter are not significantly different at $P = 0.05$ (Duncan's [1955] multiple range test).

Table 4. Effect of carbosulfan and lindane seed treatment on seed germination (laboratory analysis), plant stand and yield of 'Dirkwin' spring wheat in field trials at Arbon Valley and Nampa, ID, 1982.

Seed treatment (g AI/kg seed)*	% Germination	Plant stand (no. plants/m row)†		Yield (kg/ha)	
		Arbon Valley	Nampa	Arbon Valley	Nampa
Carbosulfan 0.63	96a‡	51.8a	77.7a	1270a	4947a
Carbosulfan 1.25	97a	51.8a	74.0a	1296a	4441ab
Carbosulfan 2.50	96a	55.5a	70.3a	1182a	4463ab
Lindane 0.30	95a	59.2a	70.3a	1328a	3674bc
Vitavax-200	96a	29.6b	66.6a	872a	3075c

* All seeds were treated with Vitavax-200 (2.0 ml/kg seed).

† Evaluated 29 and 35 days postplanting at Arbon Valley and Nampa, repectively.

‡ Column means followed by the same letter are not significantly different at $P = 0.05$ (Duncan's [1955] multiple range test).

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RELATIVE EFFICIENCY OF *MYZUS NICOTIANAE* AS A VECTOR OF TOBACCO ETCH VIRUS TO TOBACCO AND SICKLEPOD[1]

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Abstract: The relative transmissibility of tobacco etch virus (TEV) to tobacco, *Nicotiana tabacum* L., and sicklepod, *Cassia obtusifolia* L., by apterous *Myzus nicotianae* Blackman was examined. Multiple-aphid transfers were used to calculate the virus transmission rate from burley tobacco to two flue-cured tobacco varieties, 'NC 2326' and 'Coker 176', one burley tobacco variety, 'B-21', and sicklepod. *Myzus nicotianae* were able to transmit TEV readily to both burley and flue-cured tobaccos. No differences were found in the relative transmissibility of TEV to the three tobacco varieties tested (probabilities of transfer of 0.306, 0.316, and 0.256 for 'B-21', 'NC 2326', and 'Coker 176', respectively). Transmission of TEV to sicklepod was lower (probability of transfer of 0.041) than transmission of TEV to the three tobacco varieties tested.

Key Words: *Cassia obtusifolia, Myzus nicotianae, Myzus persicae, Nicotiana tabacum,* TEV.

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Tobacco etch virus (TEV) is found almost everywhere tobacco, *Nicotiana tabacum* L., is cultivated (Purcifull and Hiebert 1982). It is distributed throughout North and South America and causes reduction in tobacco yield and quality (Purcifull and Hiebert 1982). In North Carolina, crop losses in burley tobacco due to TEV were estimated at $350,000 in 1982, $280,000 in 1983, and $200,000 in 1984 (Powell 1985). Losses in flue-cured tobacco were estimated at $230,000 in 1984 (Main and Nusser 1986); thus, the combined total losses in flue-cured and burley tobacco were ca. 0.05% of the 1984 North Carolina production. TEV has a wide host range, but is most common in solanaceous crops (Gooding 1975). TEV does not survive long in dead host tissue under field conditions and overwinters in perennial weed hosts (i.e., horsenettle, *Solanum* spp., and ground cherry, *Physalis* spp. [Gooding 1975; Purcifull and Hiebert 1982]).

In the spring, dispersing alate aphids acquire TEV while probing infected overwintering hosts and transmit TEV to alternative weed hosts (horsenettle, ground cherry, jimsonweed [*Datura stramonium* L.], bull thistle [*Cirsium vulgare* (Savi) Tenore], lambsquarters [*Chenopodium album* L.], sicklepod [*Cassia obtusifolia* L.] [Purcifull and Hiebert 1982]) and cultivated crops (pepper, tobacco, tomato [Gooding 1975]) as they search for new host plants, thereby propagating the infection. Sicklepod, a natural host for TEV (Demski 1979), is one of the main weed problems in the production of soybeans in the southeastern United States (Bridges and Walker 1987). In 1981, sicklepod was estimated to infest ca. 81,000 ha of soybean in the southeastern North Carolina (Thompson and Sherman 1981),

[1] HOMOPTERA: Aphididae. Accepted for publication 25 February 1988.

and in a 1983 survey, it was rated the sixth most common weed in the southern region of the United States (Talbert 1984). Due to the prevalence of this weed, the difficulty of its control and its role as a natural host of TEV, the importance of sicklepod in the seasonal spread of TEV must not be overlooked.

TEV is transmitted from host to host in a nonpersistent manner by many species of aphids (Kennedy et al. 1962; Purcifull and Hiebert 1982). The green peach aphid (GPA), *Myzus persicae* (Sulzer), is a major vector of TEV (Swenson 1968; van Emden et al. 1969) and is commonly found on many weeds and cultivated crops throughout the southeastern United States (Palmer 1952). Recently a tobacco feeding-form of GPA has been identified (Blackman 1987). This tobacco-feeding form has distinctive morphometric characters and, due to its apparently anholocyclic life cycle, has been described as a distinct species, *Myzus nicotianae* Blackman. The status of *M. nicotianae* as a vector of TEV is unknown.

When testing the ability of an aphid to transfer viruses, the amount of time an aphid is allowed access to a virus-infected host plant and the number of aphids that are transferred to each indicator plant are important factors in the experimental design. Burrows (1987) and Swallow (1985) have discussed the efficiency of multiple aphid transfers to estimate the pathogen transmission rate of vectors, particularly when transmission rates are low. The objective of this study was to compare the relative transmission of *M. nicotianae* as a vector of TEV from infected tobacco to healthy tobacco and sicklepod. In order to compare transmission rates, the appropriate access time and number of aphids to transfer must also be determined.

MATERIALS AND METHODS

Seedling Preparation

All test plants (flue-cured tobacco 'NC 2326' and 'Coker 176', burley tobacco 'B-21', and sicklepod) were grown from seeds. Certified tobacco seeds were sown in trays filled with sterilized loam soil. Sicklepod seeds were field-collected from the Central Crops Research Station, Johnston County, N.C., in 1983. To surface sterilize the seeds and induce germination, sicklepod seeds were scarified with sulfuric acid (Hartmann and Kester 1983). Sicklepod seeds were sown in flats of Metro Mix (#220, Grace Horticultural Products, Cambridge, MA) to reduce root damage during the transplanting process. Host plants were grown in a greenhouse at ca. 30°C with a 16:8 (L:D) photoperiod, maintained through the use of artificial lights (500 watt, Norelco quartz floodlight, Hightstown, NJ). Flue-cured and burley tobacco plants required ca. 4 to 6 weeks to grow to transplantable size (4- to 6-leaf stage), while sicklepod required 2.5 to 3 weeks. Seeding was staggered to synchronize transplanting. Seedlings were transplanted into clay pots (10.5 cm diam × 10.3 cm deep) containing Metro Mix ca. one week prior to the test date. Sicklepod seedlings were selected for uniform height and expansion of the first two true leaves.

Aphid Collection

M. nicotianae were reared on flue-cured tobacco 'McNair 944' in screen cages (1.8 × 1.0 × 1.2 m), maintained in a greenhouse as described above. The colony was started 23 August 1983 from aphids field-collected on flue-cured tobacco 'McNair 944' at the Central Crops Research Station, Johnston County, N.C.

(previously considered GPA, Throne and Lampert 1985). Voucher specimens were deposited with the N.C. State University Insect Museum Collection.

Virus Source

The isolate of TEV used in these studies was collected from naturally infected flue-cured tobacco 'Speight G-28' in Duplin Co., N.C., in 1984. The virus was determined serologically to be TEV (Gooding 1975) and was found to react with antisera prepared against both the Kentucky and Simons' strains of TEV. The virus source in all tests was burley tobacco 'B-21' plants, aphid-inoculated with TEV. Source plants were kept in screen cages in the greenhouse maintained as described above. Source leaves for the transmission efficiency tests were the second or third fully expanded, symptomatic leaves from the top of the source plant. A single leaf was removed and used as the virus source for all tests conducted on a single day.

Test Procedure

Groups of 20 to 25 apterous, adult *M. nicotianae* were collected from the colony and starved in petri dishes at room temperature for 5 to 8 h. All aphids in an individual test were allowed access to the underside of a single TEV infected leaf. Groups of aphids corresponding to the number needed per test plant were fed on the source leaf. Only aphids that appeared to be probing during the access time were used in the transmission test.

Access Time and Group Transfer Tests

For purposes of this paper, access time is defined as the amount of time an aphid was allowed to remain upon a host plant, while acquisition time is defined as the actual amount of time spent probing the host plant. Access time, rather than acquisition time, was measured in these experiments.

To determine the appropriate access time, aphids were given access to a TEV source leaf for fixed periods of time (30, 60, or 120 seconds). Aphids were transferred from the source leaf to the test plant with a small brush and allowed to feed overnight. During this period, plants were isolated to prevent aphid movement from plant to plant. At the end of the feeding period (24 h), plants were sprayed with acephate (Orthene TIS®, 75% SP, Chevron Chem. Co.). Plants were then transferred to a greenhouse where they were treated with aldicarb (Temik®, 15G, Rhone-Poulenc) and allowed to grow until symptoms were evident (14 days for tobacco, and 28 days for sicklepod). The number of symptomatic plants was determined by visual inspection (Purcifull and Hiebert 1982) and recorded.

Transmission rate was determined by calculating the probability of transmission by an individual aphid (Burrows 1987). Probability of transfer ($\tilde{p}$) was calculated using the function of Burrows (1987):

$$\tilde{p} = 1-[(2kR + k-1)/(2kN + k-1)]^{1/k} \quad (1)$$

where: N = number of test plants, R = number of healthy plants, and k = number of aphids transferred to a single host plant.

When using Equation 1 to estimate $\tilde{p}$, it is desirable to have moderate levels of transmission, i.e., < 0.50, and since $\tilde{p}$ is a function of the number of aphids placed on the indicator plant, it was necessary to determine the appropriate number of

aphids to transfer to each host plant (Gibbs and Gower 1960; Swallow 1985). If too many aphids are transferred to the test plants, all plants will become infected. When this happens, Equation 1 become saturated and $\tilde{p}$ cannot be calculated (Swallow 1985; Burrows 1987). In tobacco tests, 2, 4, 6, and 10 aphids were transferred to each plant. For sicklepod, transfers of 4, 6, 8, and 10 aphids per plant were made. From these experiments, probability of transfer was calculated for each access time and number of aphids transferred. Results were then examined to determine the optimum number of aphids to transfer to each host plant (Swallow 1985).

TEV Transmission Efficiency Test

Transmission efficiency from burley tobacco 'B-21' to flue-cured tobacco ('Coker 176' and 'NC 2326') and sicklepod was compared. 'Coker 176' was added to the test because it is tobacco mosaic virus-resistant and is frequently used when studying aphid-borne viruses in the field. Seedlings were grown in the greenhouse to the appropriate size. From preliminary tests, access time and number of aphid transfers were determined for each recipient host. Aphid transmissions were made in the laboratory on 11-13 April 1986 using the procedures described above. For each replication, aphid transfers were made to seven plants of each host. Two replicates of the test were set up on a single day for three consecutive days (6 replicates total). In the greenhouse, recipient plants were placed on benches in a randomized fashion and blocked for location of the bench. Beginning two weeks post-treatment, plants were examined on a weekly basis for symptoms. After five weeks, all asymptomatic plants were considered to be uninfected.

Using Equation 1, the virus transmission rate was calculated for each test host. Data were subjected to an analysis of variance (ANOVA) (SAS Institute 1982) to determine significant differences due to the treatments, and the Waller-Duncan K-ratio t test ($K = 100$, $\alpha = 0.05$) was used to separate treatments. Treatments were test hosts, and blocks were bench locations combined with date of testing.

RESULTS AND DISCUSSION

Access Time and Group Transfer Tests

The calculated transmission rates of TEV by an individual aphid varied from 0.22 to 0.58 for tobacco, and from 0.02 to 0.22 for sicklepod (Table 1), depending upon the access time. In burley tobacco, *M. nicotianae* were very effective vectors and transmission was readily accomplished with only two aphids per plant. In the first burley test (3 October 1985), the virus source plant was in the flowering stage when the transmission efficiency was determined, while in tests 2 and 3 (5 and 26 Nov. 1985, respectively), the virus source plant was in the vegetative stage. An increase in transmission efficiency was noted when the younger virus source plant was used; however, these tests were conducted at different times of the year and the age of the virus infection in the source plant varied (ca. 25, 8, and 11 weeks, test 1, 2, and 3, respectively). Therefore, conclusions as to the effects of the age of the virus source plant or age of infection should not be drawn from these experiments. Using the same greenhouse conditions, Gray (1984) found the proportion of GPA capable of transmitting potato virus Y from infected burley tobacco 'B-21' to healthy burley tobacco varied from 0.07 to 0.83 when one aphid was transferred per plant. These results are in agreement, which is not unexpected, because these viruses are both

in the potyvirus group (Hollings and Brunt 1982) and, until recently, *M. nicotianae* was not separated from GPA. Two aphids per plant were selected as the most appropriate number for the burley tobacco 'B-21' recipient hosts.

In the flue-cured recipient 'NC 2326', a vegetative TEV source plant was used in all transmission efficiency tests. Considerable variation was observed in p̄ among the different access time and number of aphids transferred combinations (Table 1); however, all combinations were effective at transmitting TEV. Once again, two aphids were determined to be the most appropriate number of aphids to transfer for the same reasons cited earlier.

Transmission efficiency was considerably less to the sicklepod than to the tobacco tested (Table 1). The virus source plant was in the flowering stage when the first test was conducted (14 October 1985), while the source plant used in the second test (8 January 1986) was in the vegetative stage. Sicklepod was apparently an unsatisfactory food source for *M. nicotianae* reared on tobacco. After being placed on sicklepod, aphids would frequently wander off the plant, possibly contributing to the low transmission efficiency. Based on the transmission efficiency in the second test, four aphids per transfer were slected as the appropriate number of aphids to be transferred to sicklepod. Two aphids per transfer were also included for consistency with the tobacco.

Based on the results of previously discussed tests, an access time of 60 seconds was selected for all recipient hosts.

TEV Transmission Efficiency Test

Using the access time and number of aphids per multiple aphid transfer as determined above, a single test was conducted and all host plants were tested for relative transmission efficiency. Significant differences ($F = 8.59$; d.f. = 4, 20: $P = 0.0003$) in relative transmission efficiency of TEV by *M. nicotianae* were observed among the host plants. No significant differences in the transmission efficiency of TEV were observed among any of the tobacco types tested ($P > 0.05$), while all tobacco had a significantly greater transmission efficiency than the sicklepod ($P < 0.05$) (Table 2). No significant differences ($P > 0.05$) in transmission efficiency in the sicklepod were detected when either two or four aphids were used in the multiple transfers (Table 2).

These experiments indicated that *M. nicotianae* are efficient vectors of TEV from infected burley tobacco source plants to both burley and flue-cured tobacco, but a relatively poor vector to sicklepod. In sicklepod, the probability of transfer by a single aphid averaged 0.04 as compared with > 0.25 in all tobacco tested. Demski (1979) found sicklepod infected with TEV to be common along field borders of TEV-infected pepper grown in Georgia, with TEV apparently being moved from infected pepper to sicklepod. TEV was not found in sicklepod growing beyond 50 m from infected pepper plants. Demski concluded that sicklepod was not a major factor in the epidemiology of TEV in pepper, but the importance of sicklepod to the epidemiology of TEV through the environment was not examined. Although sicklepod was a relatively poor recipient of TEV, it is abundant throughout the southeastern United States and has the potential of being a major reservoir and component of the seasonal spread of TEV through its host complex.

Table 1. Preliminary tests to determine the appropriate number of aphids to transfer from burley tobacco to alternative hosts and appropriate access time for TEV transmission tests.

Recipient plant	Source age*	Date tested	No. plants tested (N)	No. of aphids per plant (k)	Access time (Sec)	No. healthy plants (R)	$\bar{p}$
Burley 'B-21'	old	3 Oct. 1985	10	6	30	2	0.22
Burley 'B-21'	old	3 Oct. 1985	10	6	60	2	0.22
Burley 'B-21'	old	3 Oct. 1985	10	6	120	0	undefined†
Burley 'B-21'	old	3 Oct. 1985	10	10	30	0	undefined
Burley 'B-21'	old	3 Oct. 1985	10	10	60	0	undefined
Burley 'B-21'	old	3 Oct. 1985	10	10	120	0	undefined
Burley 'B-21'	young	5 Nov. 1985	7	2	30	3	0.33
Burley 'B-21'	young	5 Nov. 1985	7	2	60	1	0.58
Burley 'B-21'	young	5 Nov. 1985	7	4	30	1	0.34
Burley 'B-21'	young	5 Nov. 1985	7	4	60	0	undefined
Burley 'B-21'	young	5 Nov. 1985	7	6	30	0	undefined
Burley 'B-21'	young	5 Nov. 1985	7	6	60	0	undefined
Burley 'B-21'	young	26 Nov. 1985	10	2	30	2	0.53
Burley 'B-21'	young	26 Nov. 1985	10	2	60	2	0.53
Burley 'B-21'	young	26 Nov. 1985	10	4	30	0	undefined
Burley 'B-21'	young	26 Nov. 1985	10	4	60	0	undefined
Burley 'B-21'	young	26 Nov. 1985	10	6	30	0	undefined
Burley 'B-21'	young	26 Nov. 1985	10	6	60	0	undefined
Flue-cured 'NC 2326'	young	29 Oct. 1985	7	6	30	0	undefined
Flue-cured 'NC 2326'	young	29 Oct. 1985	7	6	60	0	undefined
Flue-cured 'NC 2326'	young	29 Oct. 1985	7	6	120	0	undefined
Flue-cured 'NC 2326'	young	29 Oct. 1985	7	10	30	0	undefined

Flue-cured 'NC 2326'	young	29 Oct. 1985	7	10	60	0	undefined
Flue-cured 'NC 2326'	young	29 Oct. 1985	7	10	120	0	undefined
Flue-cured 'NC 2326'	young	19 Nov. 1985	7	2	30	2	0.44
Flue-cured 'NC 2326'	young	19 Nov. 1985	7	4	30	0	undefined
Flue-cured 'NC 2326'	young	19 Nov. 1985	7	6	30	1	0.24
Sicklepod	old	14 Oct. 1985	10	6	30	6	0.08
Sicklepod	old	14 Oct. 1985	10	6	60	6	0.08
Sicklepod	old	14 Oct. 1985	10	6	120	0	undefined
Sicklepod	old	14 Oct. 1985	10	10	30	4	0.09
Sicklepod	old	14 Oct. 1985	10	10	60	8	0.02
Sicklepod	old	14 Oct. 1985	10	10	120	8	0.02
Sicklepod	young	8 Jan. 1986	6	4	30	2	0.22
Sicklepod	young	8 Jan. 1986	6	4	60	2	0.22
Sicklepod	young	8 Jan. 1986	6	6	30	2	0.15
Sicklepod	young	8 Jan. 1986	6	6	60	3	0.10
Sicklepod	young	8 Jan. 1986	6	8	30	1	0.17
Sicklepod	young	8 Jan. 1986	6	8	60	0	undefined

* Old source plant aphid inoculation on 9 April 1985 and ratooned to prevent flowering, young source plant aphid inoculated on 11 Sept. 1985.
† Transmission was 100%; therefore, the probability of transfer, $\bar{p}$, could not be calculated (Swallow 1985).

Table 2. Relative transmission efficiency ($\tilde{p}$) of tobacco etch virus by *Myzus nicotianae*. Virus source = Burley 21, access time = 60 seconds.*

Host	No. aphids transferred per plant	Transmission efficiency ($\tilde{p}$)†
Burley 'B-21'	2	0.306 (0.049)a
Flue-cured 'Coker 176'	2	0.256 (0.080)a
Flue-cured 'NC 2326'	2	0.316 (0.073)a
Sicklepod	2	0.024 (0.015)b
Sicklepod	4	0.058 (0.017)b

* Probability of transfer calculated from 7 plants per treatment (host) per block with 2 or 4 aphid transfers per plant. Treatments arranged in a randomized complete-block design with 6 replicates.

† Mean ± SEM. Means followed by the same letter are not significantly different (Waller-Duncan K-ratio t test, $P > 0.05$) (SAS Institute 1982).

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SURVEY OF CATTLE LICE[1], GRUB[2], AND PSOROPTIC MITE[3] INFESTATIONS IN SOUTHEAST GEORGIA

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Abstract: Vacuum sampling detected the cattle biting louse, *Bovicola bovis* (L.), on 14.8% of 128 cattle in a Southeastern Georgia sales barn from 21 January through 18 March 1985. Vacuum sampling failed to detect the presence of *B. bovis* in 264 samples taken from early spring through fall. None of the samples were positive for the mite , *Psoroptes ovis* (Hering). The common cattle grub, *Hypoderma lineatum* (de Villers) was in the backs of 54% of sales barn cattle and an untreated herd examined 19 November to 4 March, with a peak of 13.5 grubs per infested animal in mid-January.

Key Words: Cattle lice, cattle grub, *Bovicola bovis*, *Hypoderma lineatum*, *Psoroptes ovis*, ectoparasites.

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In the past 20 years many changes in the management of cattle have occurred, thus warranting a survey of current ectoparasite incidence. Cattle from Georgia are a part of an extremely complex series of interstate livestock movements. In 1985, the Georgia Department of Agriculture issued 29,304 health certificates for cattle shipped into Georgia and certificates for 213,572 head shipped to 33 other states (J. A. Cobb, personal communication). The Georgia cattle population rose 1% to 1,750,000 head in 1985 with an estimated 780,000 calves born and only 10,900 calves slaughtered within the state (Snipes and Hammer 1986). State records and estimates indicate that the vast majority of the Georgia calf crop is destined for finishing operations in other states. Thus, ectoparasites of Georgia stocker cattle and calves are potential problems for the conditioning and final feed lots across the country.

In a survey of 23 herds of cattle in 19 counties, Roberts (1963) detected lice in 16 of the herds, January-March 1962. *Linognathus vituli* (L.) and *Solenopotes capillatus* Enderlein, both Anoplura, were detected in 57% and 50%, respectively, of the infested herds, while *Bovicola bovis* (L.), Mallophaga, was found in only two herds. Roberts (1963) reported no correlation between degree of infestation and location of the herds in Georgia. Cattle lice spend their entire life on the host, with populations declining and becoming cryptic during the spring and summer (Lewis and Christenson 1962; Bram 1978).

Bovine psoroptic mange outbreaks caused by *Psoroptes ovis* (Hering), the sheep scab mite, generally are confined to cattle in mid-western states and California (Meleney and Christy 1978; Meleney and Roberts 1979). However, the later paper reported the mange in Georgia in 1976. *Psoroptes ovis* live on the skin and feed on

[1] MALLOPHAGA: Trichodectidae. Accepted for publication 25 February 1988.
[2] DIPTERA: Oestridae.
[3] ACARI: Psoroptidae.
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tissue fluids by piercing the skin with their minute chelicerae. Serum, which exudes from the feeding site wounds, hardens and forms a scab (Sweatman 1958).

The common cattle grub, *Hypoderma lineatum* (de Villers), was found in 11 of 18 herds in Georgia with the average number of grubs per animal ranging from 0.3 in Clarke County to 8.8 in Spalding County (Roberts 1963). Cattle grub populations are generally higher in the Piedmont and upper to middle Coastal Plains than in other parts of Georgia (Roberts et al. 1964). The life cycle of the common cattle grub, *H. lineatum*, takes approximately 1 year.

The objective for our study was to survey cattle for ectoparasites in Southeast Georgia both in private herds and sales barns. Our survey emphasized detecting *B. bovis*, *P. ovis*, and *H. lineatum*. *Psoroptes ovis* was included even though seldom reported from Georgia because vacuum sampling (Callcott 1985; French and Callcott 1987) provided a technique to detect sparse populations that produce no clinical symptoms of psoroptic mange. The vacuum sampling method also warranted evaluation for detecting *B. bovis*.

MATERIALS AND METHODS

All animals were located in counties of the southeastern coastal plain of Georgia. Counts and samples were taken while cattle were crowded in a narrow alley leading to a head-gate or while being held in a head-gate. The vacuum method of Callcott (1985) and French and Callcott (1987) was used to survey for *B. bovis*, and *P. ovis*. We used a household vacuum cleaner, an in-line screen support for Whatman no. 4 filter paper and an individual collecting head for each sample. The collecting head was moved against the hair to facilitate skin contact. The vacuum was allowed to pull for 30-45 seconds. Except where noted, the standard sample was taken from the withers and areas extending along the top-line to the tailhead. Samples were processed by soaking in alcohol with eosin, rinsing on to lined filter paper and observing at 10-12 magnification with a stereoscopic microscope (Meleney et al. 1982; Callcott 1985; French and Callcott 1987). Skin scrapings were made by trimming ca. 25 mm^2 area of hair with scissors; a #22 scalpel blade was then used to scrape the sample into a wide-mouth jar (59 ml). The skin scraping samples were then processed as described above for vacuum samples.

The number of cattle grubs per animal was estimated by back palpation as recommended by Bram (1978). Either the entire back between the withers and the hips was palpated, or one side of the back was palpated and this number multiplied by two to approximate the total number of cattle grubs present. The second method was used in situations where only one side on the animal could be reached.

Scraping Versus Vacuum Sampling for B. bovis

On 5 calves known to have *B. bovis*, 10 paired samples were taken. A vacuum sample was first taken in a circular area ca. 35 mm in diameter. A scraping sample (ca. 25 mm^2) was subsequently taken within the vacuumed area.

Sales Barn Survey

During the winter, 20-30 vacuum samples and 10-20 palpation counts were taken every 2 weeks on unrestrained cattle in a crowded alleyway (21 January to 1

April 1985) in a sales barn located in Hagan, Evans County. The cattle in the sample were considered stocker grade with an estimated weight of 313.8 ± 13.9 kg; 79% were female. The summer and early fall survey for lice was done with cattle restrained by a head-gate in a sales barn in Statesboro, Bulloch County with 264 vacuum samples taken (20 June to 21 November 1985). The sample sites were poll (12 samples), ear (22), neck (6), withers (85), dewlap (18), bottom line from between front legs to udder or scrotum (45), tail set (56), tail and switch (10), and udder or scrotum (9).

Private Herd Survey

Six private herds of registered or controlled cross-bred cattle in Bulloch, Effingham, Screven and Tattnall Counties were sampled (See Table 1 for the number of samples and dates). An additional herd (EXP) in a psoroptic mange experiment was sampled from 19 November to 13 March 1985; these grade hereford heifer calves were purchased through the Statesboro sales barn, not treated for cattle grubs after purchase in October, and presumed to be untreated for insect parasites.

Arithmetic means are reported ± standard error of the mean (SEM).

RESULTS AND DISCUSSION

Scraping Versus Vacuum Sampling for B. bovis

Five of the 10 paired samples contained *B. bovis* in both vacuum and scraping samples. Two other vacuum samples contained *B. bovis* but not in the paired scraping sample; two other sample pairs contained lice collected by scraping but not by vacuuming; and one sample pair was negative for lice.

Lice Survey

Bovicola bovis were present in 19 of 148 vacuum samples collected from cattle in a sales barn during the winter survey. All cattle that tested positively for *B. bovis* had light infestations using the criteria established by Bram (1978). Less than 25% of cattle examined were infested per collection date (Table 1).

Table 1. Incidence of *Bovicola bovis* in vacuum samples collected from sales barn cattle in Southeast Georgia January-April 1985.

Collection Date	Number of Cattle Sampled	Number of Cattle Parasitized	Percent Parasitized
Jan 21	23	3	13.0
Feb 4	30	6	20.0
Feb 18	30	2	6.7
Mar 4	20	2	10.0
Mar 18	25	6	24.0
Apr 1	20	0	0.0

Of the adult and nymphal lice collected, about half of them were intact, while the remaining were fragmented. Most of the lice eggs collected were empty with a few containing embryos; the viability of the embryos was not determined. The percent of animals parasitized varied over the first 8 weeks and dropped from a high of

24% on 18 March to none on 1 April. The 1985 summer and early fall survey attempted to detect cryptic populations of *B. bovis*. Only four samples, from withers or neck, yielded *B. bovis* with no more than two specimens per sample.

No ectoparasites were collected in 180 vacuum samples from the six private herds. Five of 5 heifers in the EXP herd were positive for *B. bovis* in vacuum samples taken 2 January and this species was also in scraping samples from 7 of 8 EXP heifers on 16 January.

While vacuum sampling has been shown to be as effective as scraping for detection of *P. ovis* (French and Callcott 1987), *B. bovis* is more difficult to detect by vacuum due to adaptations to grasp or glue to hair. Hair parting, then counting (Bram 1978) and scraping require considerably more skill and are very difficult to perform in a rapidly moving sales barn operation. In the two sales barns we worked, prior to sales, lots or groups of cattle were crowded into an alleyway leading to a head-gate. A veterinarian and an assistant recorded or applied a state ear tag and took a blood sample from each potentially reproductive animal; this process of less than one minute per animal made it difficult to examine the cattle concurrently for ectoparasites. In the alleyway, the cattle were unrestrained and thus difficult to sample except by the vacuum method.

One half of the *B. bovis* specimens in vacuum samples were fragmented and most of the eggshells were empty, indicating that vacuum sampling did not determine the activity of a louse population, but indicated that *B. bovis* was present at some time in the season. During the winter, the vacuum sampling indicated that ca. 14% of the cattle passing through that sales barn, from 21 January through 21 March, had been exposed to *B. bovis*. In the six private herds, the evident superior management and ectoparasite control measures were effective as indicated by 180 negative samples. The EXP herd, bought through a sales barn in October and not treated for ectoparasites, showed 87.5% parasitism by *B. bovis* in January.

Mites, P. ovis

No *P. ovis* were found in the 333 vacuum samples taken during the late fall and winter from cattle in private herds and sales barns. We are confident that none were present on the top line of these cattle due to the efficacy of the vacuum method for *P. ovis* (Callcott 1985; French and Callcott 1987).

Cattle Grubs

The data from grub counts on cattle in sales barns and the EXP herd were similar and are combined in Table 2. Of the 122 examinations, 54% were positive for cattle grubs from 19 November to 4 March. The count was nearly constant through 18 February then declined on 26 February and 4 March; no grubs were found 13 and 18 March. In four private herds, only 1 of the 40 animals examined was infested (2 grubs). Roberts (1963) reported 9 of 14 herds had grubs with a parasitism rate of 38.5% (avg 3.5 grubs/animal) in a survey of cattle in 14 Georgia Counties. Our data from the sales barn and EXP herd indicated that a higher incidence of parasitism with more grubs (peak 13.5 grubs) per infested animal.

CONCLUSIONS

The vacuum sampling method of French and Callcott (1987) for the mite, *P. ovis*, was valuable in detecting the louse *B. bovis* in sales barns. Although not the

ultimate diagnostic tool, vacuum sampling was the most feasible in the sales barn situation and will indicate the incidence rate of *B. bovis* during the winter months. This sampling method failed to reveal cryptic populations from early spring through fall. None of the vacuum samples were positive for *P. ovis*. Cattle grubs, *H. lineatum*, were present in over one half of the cattle processed through a sales barn during the winter. Herds with well executed management programs were essentially free of all three parasites.

Table 2. Incidence of cattle grubs in Southeast Georgia, November 1984-March 1985.

sample dates	n	% with grubs	grubs/infested host	
			avg.	range
Nov 19	11	63.6	5.6	2-9
Jan 4-16	31	67.7	13.5	2-40
Jan 21-29	27	55.5	13.4	2-27
Feb 4-18	30	56.7	10.3	2-46
Feb 26-Mar 4	24	25.0	4.5	2-8
Mar 13-18	15	0	—	—

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ESTABLISHING AND DISCRIMINATING SEEDCORN MAGGOT[1] INJURY TO SOYBEAN

Leon G. Higley[2] and Ronald B. Hammond[3]

Abstract: Seedcorn maggots (SCM), *Delia platura*, are a sporadic pest of soybean usually occurring in damaging numbers only when conditions are such that soybean fields are attractive ovipositional sites. Consequently, various methods must be employed to reliably produce injurious SCM populations for field experimentation. Additionally, effects of SCM injury to soybean may be obscured by other factors causing seedling injury; therefore, specific evaluation methods must be employed to characterize SCM injury. These techniques are discussed in the context of evaluating insecticides against SCM. Two methods are described and compared for enhancing SCM oviposition in the field: baiting with meat and bonemeal and incorporating cover crops (alfalfa, rye, or wheat). Advantages and disadvantages of the approaches are discussed. Techniques for evaluating SCM injury to soybean also are described. Measures of SCM numbers are useful in establishing the presence of injurious SCM populations but may not directly reflect injury to soybean. Therefore, SCM population estimates used alone can be misleading. Stand reduction and plumule injury provide useful estimates of SCM injury but can be produced by other agents. Although various factors may produce cotyledon injury, these injuries are distinguishable from that caused by SCM. We conclude that the most thorough and convincing evidence in evaluating SCM injury is provided by measuring SCM numbers and different kinds of SCM injury, with emphasis on cotyledon injury.

Key Words: *Delia platura*, soybean, insect injury, insecticide testing.

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The seedcorn maggot (SCM), *Delia platura* (Meigen), is a pest on germinating seeds of a variety of crops. Among the most important North American plants subject to SCM injury are soybean, dry beans, potatoes, and corn. Seedcorn maggot eggs are laid in the soil, usually in soil crevices or under clods, and larvae may feed on organic matter in the soil (Miller and McClanahan 1960) as well as on germinating seeds.

In the north-central region of the United States, SCM is primarily a sporadic pest of soybean, producing considerable stand reduction, even complete stand loss within specific fields, but infrequently over wide areas. Such variability in SCM numbers presents problems in attempting to obtain injurious SCM levels for experimental purposes such as evaluating insecticides.

Other than brief reports in the early literature and recent work on ovipositional stimuli, few studies have considered SCM habitat affiliations. Research in Iowa indicated distinct habitat preferences by SCM adults (Higley and Pedigo 1984a), with a particular preference for cut alfalfa fields (Higley and Pedigo 1985). Emergent females have a variable response to stimuli, which may influence habitat selection. Initially, females respond to a carbohydrate food stimulus, then to a protein food stimulus, and finally to an ovipositional site stimulus (McLeod 1964). Both sexes of SCM adults are attracted to flowers and to recently disturbed soil

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(Funderburk et al. 1983; Miles 1950; Miller and McClanahan 1960; Reid 1940). Many workers have reported that SCM display an ovipositional preference for organic (decaying) matter, particularly decaying plant matter (Hawley 1922; Narukawa and Kumashiro 1930; Reid 1940; Miles 1950), although Higley and Pedigo (1984b) suggest that some associations of SCM females with decaying matter also may be to obtain proteinaceous food and not just for oviposition. Many studies have examined factors associated with SCM oviposition (e.g., Eckenrode et al. 1975; Yu et al. 1975; Weston and Miller 1987). Ovipositional preference differs between host plants (Ibrahim and Hower 1979; Yu et al. 1975) and may be stimulated by microorganisms associated with germinating seeds (Eckenrode et al. 1975). In the field, SCM females may oviposit in response to germinating seeds of highly preferred hosts such as lima bean (Yu et al. 1975). However, Funderburk et al (1983) observed no differences in SCM adult emergence from within-row or between-row locations in soybean fields and concluded that germinating soybeans were not attractive for oviposition under field conditions.

In this manuscript, we describe various techniques for reliably developing injurious SCM populations in the field. Additionally, we discuss methods for discriminating SCM injury to distinguish it from other seed and seedling injuries. Because SCM injury evaluations are frequently used as measures of insecticidal efficacy, we discuss these evaluations in that context. Although our discussion focuses on soybean, it is equally applicable to other species injured by SCM.

MATERIALS AND METHODS

Data from various experiments conducted in 1981-1986 in Iowa and Ohio were used to illustrate establishment and evaluation procedures for SCM. All experiments employed a randomized complete-block design with four replications (Ohio 1983 study used six replications). Additionally, all studies used a 76-cm row spacing, a planting rate of 30 plants/row-m (except the Iowa 1982 study, which used 22 plants/row-m), and were planted in mid- to late-May. In those studies in which SCM adult emergence was measured, emergence traps (Funderburk & Pedigo 1980) were placed over the row within two weeks after planting and were checked three to four weeks after placement. Stand counts were made after soybean emergence was complete, usually at the unifoliolate stage. When injury evaluations were taken, seedlings were examined between the cotyledonary and unifoliolate stages.

Ohio studies (see Table 1) were conducted at or near the Ohio Agricultural Research and Development Center at Wooster, Ohio. Comprehensive information on methods and materials for the Ohio 1981 study is provided by Hammond & Jeffers (1983) and for the Ohio 1982-83 study by Hammond (1984). For the Ohio 1981 study, wheat (cv. 'Ruler') was disked under and planted in soybean (cv. 'Pella') in 15-m by 4-row plots. For the Ohio 1982-1983 studies, a rye cover crop was disked twice and planted in soybean (cv. 'Pella') with a plot arrangement as in 1981. The Ohio 1983 study was established with agronomic procedures as in the other Ohio studies; however, alfalfa (stand established in 1982) was incorporated by plowing followed by repeated disking for adequate mixing of alfalfa residue and soil. Soybeans ('Amsoy 71') were planted into 7-m by 10-row plots.

Both Iowa studies were conducted at the Johnson Research Farm, near Ames, Iowa. All insecticide treatments were applied at planting. In the 1982 Iowa study (Tables 1 and 2), soybeans ('Amsoy 71') were planted in 15-m by 4-row plots. One week before planting, all plots were baited with 1 liter of meat and bonemeal bait per 5 row-m. Two SCM emergence traps were placed in each plot. Stand counts and injury evaluations were based on six 1 row-m samples from each plot. In the 1986 Iowa study (see Table 1), soybeans ('Corsoy 79') were planted into disked and rotary-tilled alfalfa in 7-m by 4-row plots. One SCM emergence trap was placed in each plot. Stand counts and injury evaluations were based on two 1-row-m samples from each plot.

RESULTS AND DISCUSSION

Establishing Injury

Certainly, one important factor in establishing damaging SCM populations is to time ovipositional baiting and planting concurrently with high SCM adult populations. Although a degree-day model can be used to determine this period, local variations in SCM populations can be substantial (Higley and Pedigo 1985). Consequently, better accuracy is provided by using the degree-day model to indicate when to start cone-trap sampling and then basing estimates of adult SCM populations on these samples. To minimize the effect of habitat and location on estimates of SCM numbers, sampling should be conducted at or close to the experimental site. A more accurate method is to consider ovarian dynamics of collected SCM females and to time planting and baiting with the presence of gravid females (Higley and Pedigo 1984b). However, SCM oviposition usually occurs over a sufficiently long interval that such a precise estimate of SCM ovipositional status is unnecessary.

Once gravid SCM females have appeared, establishing injurious larval SCM populations in experiment settings primarily depends on obtaining sufficient oviposition. Table 1 summarizes experiments employing various factors to promote

Table 1. Effectiveness of various seedcorn maggot (SCM) ovipositional baits with respect to mean SCM adult emergence and resulting mean soybean plant stands. Numbers followed by * indicate treatment was significantly different from the check by paired t-test at the 5% level.†.

Location	Year	Treatment	SCM/row-m	Plants/row-m
Ohio	1983	Check	—	26.8
		Alfalfa	—	14.9*
Iowa‡	1986	Alfalfa	42.2	6.9
Ohio	1982-83	Check	2.2	20.7
		Rye	16.6*	10.5*
Ohio	1981	Check	2.6	24.6
		Wheat	33.4*	14.9*
Iowa	1982	Check	4.9	21.5
		Meat & Bonemeal	49.6*	18.4

† Treatments planted in mid-May, 76-cm rows, 30 plants/row-m (Iowa 1982, 22 plants/row-m). Alfalfa, rye, and wheat disked into soil before planting; meat & bonemeal applied preplant 1 liter bait/5 row-m. Adult SCM emergence determined from 1.0 × 0.2 m emergence traps.

‡ SCM numbers are not collected in Ohio 1983 experiment; therefore, data from Iowa 1986 are presented to indicate magnitude of emerged SCM from alfalfa baiting (however, no check is available for comparison with these data).

SCM oviposition. In general, two approaches are possible: disking in a cover crop or applying a bait. Both methods produce SCM populations in the soil that are well in excess of those observed in unbaited checks. Applying meat and bonemeal bait is a standard technique for enhancing SCM oviposition (e.g., Eckenrode et al. 1973; Higley and Pedigo 1984b) and, as illustrated in Table 1, substantial increases in SCM numbers can be obtained. However, there are some limitations with this method. In particular, labor requirements associated with applying bait must be considered, as well as sustained attractiveness of the bait. Although meat and bonemeal bait seems to remain attractive to ovipositing SCM for 1-2 weeks, this period can be substantially shortened by prolonged periods of rain. If estimates of adult SCM populations are in error or if SCM oviposition is delayed by environmental factors (e.g., cool temperatures, prolonged rain), meat and bonemeal bait may not remain attractive long enough to ensure sufficient SCM oviposition.

As with the meat and bonemeal bait, incorporated crop treatments significantly increased numbers of emerging SCM, reduced plant stands, or both. Results in Table 1 suggest that some incorporated crop treatments may be more attractive than others; e.g., alfalfa versus wheat or rye. Although our experience suggests that alfalfa may be more attractive than other treatments, data in Table 1 are based on a variety of experiments at different locations and different times. Consequently, in the absence of direct comparisons of these different incorporated crops, it is impossible to identify an ovipositional preference among the three crops and meat and bonemeal.

One drawback to the use of crop incorporation for enhancing SCM oviposition is the need for disking and other tillage operations before planting. Although some seedbed preparation is required for any soybean planting, these operations are likely to be much more extensive when incorporating crops, particularly when incorporating alfalfa. For example, mowing or burn-down herbicide application may be required with alfalfa, followed by plowing or disking and rotary tilling to obtain an adequate seedbed. On the other hand, a benefit of disking alfalfa or other crops to enhance SCM oviposition is that incorporated crops seem to remain attractive to ovipositing SCM for a longer period than meat and bonemeal bait. For example, in the Iowa 1986 experiment (Table 1), disked alfalfa ultimately resulted in high numbers of emergent SCM despite the occurrence of frequent rainfall for approximately two weeks after planting. Although no-till planting does not appreciably increase SCM numbers (Funderburk et al. 1983; Hammond and Stinner 1987), naturally-occurring SCM infestations often occur when planting into a disked cover crop (Hammond and Jeffers 1983; Hammond 1984). Consequently, incorporating alfalfa, wheat, or rye into the soil has the added advantage of simulating natural conditions under which losses from SCM frequently occur.

Discriminating Injury

In most naturally-occurring, economic infestations, SCM numbers are so large and injury so widespread that distinguishing SCM injury from that caused by other factors is either unnecessary or obvious. However, in experimental settings, SCM numbers and corresponding injury are rarely as great, even with the use of techniques to enhance oviposition.

Consequently, once injurious SCM populations have been established, several criteria should be used to evaluate insecticide performance against SCM. In general, these criteria involve measuring SCM numbers, measuring injury by SCM,

or a combination of these approaches. Numbers of SCM per treatment may be obtained by soil sampling for larvae or pupae (e.g., Montecinos et al. 1986), which requires substantial labor, or by the use of SCM emergence traps (Funderburk and Pedigo 1980) for SCM adults. Emergence traps require far less labor than soil sampling, and identification of SCM adults is easier and more reliable than identification of larvae or pupae.

Estimates of SCM numbers are useful in confirming the presence of potentially injurious SCM populations but may prove unreliable as predictors of SCM damage. For example, Table 2 summarizes data from a field evaluation of various

Table 2. Effectiveness of various insecticide treatments against seedcorn maggot (SCM) on soybean as evaluated by four criteria: SCM adult emergence from 1.0 × 0.2 m emergence traps, plant emergence, cotyledon injury, and plumule destruction (Ames, Iowa, 1982). Injury evaluations based on six 1-row-m samples per plot (× four replications). Meat and bonemeal bait applied to all plots at 1 liter/5 row-m, one week before planting. Planting rate of 22 plants/row-m.*

			Mean #/row-m		Injury§	
Treatment†	Rate‡	Placement	SCM	Plants	Cotyledon	Plumule
Check	—	—	49.6a	18.5a	12.4a	0.63a
TF 3486	2.06	seed	39.4ab	18.5a	3.5b	0.08a
TF3486	3.12	seed	26.4bcd	17.9a	2.2b	0.17a
Agrox 3-Way	3.12	seed	20.2cd	17.3a	3.3b	0.04a
Counter 15G	2.19	in-furrow	13.5de	19.3a	4.4b	0.25a
Counter 15G	2.91	in-furrow	13.9de	19.0a	3.2b	0.17a
Dyfonate 20G	1.12	in-furrow	12.1de	21.2a	3.5b	0.04a

* Numbers followed by the same letter are not significantly different according to Duncan's Multiple Range Test at the 5% level.

† TF 3486 = chlorpyrifos; Agrox 3-way = combination of captan, lindane, and diazinon; Counter 15G = terbufos; and Dyfonate 20G = fonofos.

‡ Rate of seed treatments in g AI/kg seed and in-furrow treatments in kg AI/ha.

§ Injury expressed as number of injured plants/row-m.

insecticide treatments against SCM. Although in-furrow insecticide applications tended to reduce SCM numbers more than did seed treatments, these methods did not differ in measures of SCM injury. Additionally, although the TF 3486 treatment resulted in numbers of SCM comparable to that of the check, SCM injury was significantly less than the check. Banded insecticide treatments can reduce SCM emergence even more than in-furrow or seed treatments without producing differences in soybean injury (Hammond unpublished data). Presumably, reductions in SCM numbers are a function of how much soil area is treated with insecticide (with banding greater than in-furrow which is greater than seed treatment), whereas protection of germinating soybeans only depends on treating the soil immediately surrounding the seed. Thus, measures of SCM numbers alone do not provide an accurate indication of the effectiveness of a compound in reducing injury by SCM.

In feeding on germinating seeds SCM larvae can produce at least three forms of injury: complete seed destruction (resulting in stand loss), cotyledon gouging or scarring, and plumule destruction. The most important manifestation of SCM injury is

stand reduction; substantial, even total, reductions are possible (e.g., Hammond 1984; Hammond and Jeffers 1983; Monecinos et al. 1986). However, a variety of additional factors, including seed quality, damping-off diseases, planting depth, and soil crusting, may also reduce plant stands. Consequently, measurements of stand reductions may reflect the influence of various factors besides SCM injury. Agronomically, stand reduction frequently is the most important consequence of SCM injury to soybean, but given the variety of factors influencing soybean emergence, measures of stand reductions alone may not provide an adequate indication of SCM injury.

Because stand reduction occurs as a consequence of SCM larval feeding, we might expect reductions to be closely related to SCM populations in the soil. For most examples in Table 1, large emergent SCM populations are associated with reduced plant stands; however, this relationship was not evident for the Iowa 1982 experiment. In this instance, no significant differences in plant stand between baited and unbaited plots were noted despite an order of magnitude difference in the number of emerging SCM. Similarly, Pearson-product-moment correlations between SCM numbers and plant emergence were not significant for data from the Iowa 1982 (Table 2) or 1986 experiments. Because SCM do not seem to oviposit in response to germinating soybeans in the field (Funderburk et al. 1983), and because SCM can develop successfully without feeding on germinating seeds, the lack of a significant relationship between emergent SCM numbers and plant stand probably results from many emerging SCM not having been associated with soybean seeds. Additionally, other factors such as temperature or time of planting with respect to tillage can influence soybean emergence rates, thereby changing the likelihood of soybean injury from any given SCM population. Consequently, soybean stand reduction from SCM may not be reliably indicated by SCM numbers and, conversely, SCM numbers may not be indicated by stand reduction.

A more certain indication of SCM injury is provided by examination of emergent seedlings. Two manifestations of SCM injury on seedlings are plumule destruction and cotyledon scarring. Plumule destruction can occur when feeding SCM larvae do not destroy the seed but do injure or destroy the growing tip, or plumule, of the developing plant. In this instance, the axillary buds begin growing, resulting in a plant with two main stems. Although plumule injury is commonly associated with SCM feeding, this injury also may occur in association with preemergence damping-off produced by the fungal pathogen *Rhizoctonia solani*. Plumule injury caused by *R. solani* may be differentiated from SCM injury by the presence of other symptoms such as water-soaked leaves, lesions, or reddish-brown decay of the root (Sinclair and Shurtleff 1975). Some abiotic factors, such as soil crusting or rotary hoeing, may also injure the plumule. Consequently, although plumule destruction can be an important indication of SCM injury, it does not represent an absolute measure.

Cotyledon gouging is more diagnostic of SCM injury. Seedcorn maggots feeding on developing seeds create grooves or gouges in the outer surface of the cotyledons. Some surface discoloration or light scarring of the cotyledon may be associated with poor seed quality, but these do not include tissue removal. Preemergence diseases, such as damping-off, may produce scarring and tissue loss, but cotyledons usually display clear evidence of rot. Cotyledon injury also can occur from slug feeding, usually in no-till planted soybean (Hammond and Stinner 1987). However, slugs remove large sections or entire cotyledons, and slugs or

signs of slugs are readily apparent. Because injury from slugs usually occurs in no-till plantings and is manifested through substantial tissue removal over the entire cotyledon, not just in grooves as with SCM, it can be discriminated from SCM injury. Thus, cotyledon gouging by SCM is distinguishable from cotyledon injury by other factors and provides the best single criterion for establishing SCM injury. For example, because substantial crusting occurred in the Iowa 1986 experiment, attributing stand reduction and plumule injury solely to SCM injury would have been erroneous. However, cotyledon injury provided an indication of SCM injury independent of the effects of crusting and other factors.

For the evaluation of insecticidal efficacy or similar treatments, multiple criteria, including stand reduction, plumule injury, and cotyledon injury, provide the best indication of SCM injury. Of these, cotyledon injury provides the most reliable indicator. Measures of SCM numbers are useful in establishing the presence of injurious SCM populations, but used alone they can be misleading.

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ARTIFICIAL INFESTATION OF CATTLE IN SOUTHEASTERN USA WITH *PSOROPTES OVIS*[1,2]

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Abstract: Cattle were artificially infested with *Psoroptes ovis* to evaluate their susceptibility and to document development of psoroptic mange in southeastern coastal plain of Georgia, an area outside the normal distribution of this parasite. Sixteen Hereford heifer calves were purchased at a local stockyard and placed in an isolation facility near Statesboro, Bulloch Co., GA; five heifers were stanchioned under an open shed and 11 were confined in an adjacent, small feed lot. All calves were exposed to *P. ovis* three times: 19 December; 16 and 29 January. All five stanchioned calves developed extensive lesions. From 8 to 18 wk, after the last exposure to mites, were required for lesions to develop on at least 40% of the body of the stanchioned calves. No living mites were detected by scraping nor vacuum sampling methods on the 11 non-stanchioned calves in feed lot. Normal self-grooming probably prevented the establishment of *P. ovis* populations on these calves during the short, mild winter.

Key Words: *Psoroptes ovis*, cattle mange, mange, psoroptic mange.

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Psoroptic mange of cattle is caused by *Psoroptes ovis* (Hering). Most psoroptic mange outbreaks in cattle have been within an area from northern Texas through South Dakota, between the Mississippi River and the Rocky Mountains, and with few reported cases in southeastern USA (Meleney and Christy 1978; Hourrigan 1979; Meleney and Roberts 1979). Since cattle are freely and frequently shipped throughout the USA, it is possible that climate may be the prime factor determining the geographical distribution of the reported cases. Guillot and Cole (1984) under feed lot conditions, compared the transmission and development of mange on cattle from an endemic northern Texas area with cattle from a non-endemic area in central Texas. These workers found that northern feed lot cattle that experience colder winter temperatures were significantly more affected by the mange than those cattle from a warmer climate. The rarity of clinical psoroptic mange in the southeast may be partially explained by the mild winter climate.

However, other factors also contribute to the survival of *P. ovis* on cattle. Summer climatic conditions are associated with reduction of *P. ovis* populations and mange symptoms (Downing 1936a). Summer hair coat and general skin conditions have been suggested as factors contributing to the seasonal decline of *P. ovis* populations (Downing 1936b; Guillot 1981a). Gullot (1981a) reported that

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ambient temperature and humidity did not limit development of large populations of *P. ovis* on stanchioned cattle; concurrently, unstanchioned cattle in a similar covered corral had very low populations of mites. Cattle removed from stanchions and allowed to groom had dramatic declines in mite populations, whereas large populations developed under stanchioning of cattle with low populations (Guillot 1981a).

This study, under southeastern USA weather conditions, was conducted (1) to evaluate the transmission of psoroptic mange to cattle restricted in stanchions and to cattle in a small feed lot, and (2) to document the development of psoroptic mange in cattle in this non-endemic region.

MATERIALS AND METHODS

Sixteen Hereford heifer calves weighing 136-220 kg were purchased in November 1984, through a stockyard in Bulloch County in the eastern coastal plain of Georgia. Conditioning of the calves for the experiment near Statesboro, GA, included acclimatization to new location, diet, and handling. All were treated with thiabendazole paste for gastrointestinal roundworms and vaccinated against *Clostridium* (5 spp.), *Haemophilus* (1 sp.), *Leptospira* (5 spp.), *Pasteurella* (2 pp.), and the viruses of bovine rhinotracheitis and diarrhea-parainfluenza.

On 5 December, 16 heifers were divided into two groups: five confined in individual stanchions under an open shed and 11 in an adjacent, small feed lot. The five stanchions were centered under the roof of an open-sided shed. A slightly sloping corrugated metal roof, 3 × 8 m, was centered over the decking of 2 × 4 lumber. Each stanchion side was 1.1 m long and consisted of two horizontal steel pipes. An adjustable head gate closed one end of the stanchion. The internal width of a stanchion was 0.6 m with 0.6 m between stanchions. The five heifers were continuously confined within stanchions until the body lesions exceeded 50% of the skin surface; the minimum confinement was 17 wk and the maximum 25 wk. In stanchions, heifers could lie down or stand and could rub laterally against the top horizontal pipe of each side. The head gate prevented oral grooming except around the mouth. The feed lot was ca. 121 m^2 and was constructed of woven livestock wire and wooden posts. Within the lot were five water oak trees with trunks 14-24 cm diam. and a wooden feeding bunker. All heifers had access to automatic watering devices and those in the feed lot were fed from the common bunker. The average ration consumed per calf was 2.7 kg of 12-14% protein, custom-ground feed per day with bermuda hay free-choice.

All calves were infested with *P. ovis* obtained from the US Livestock Insects Laboratory, Kerrville, TX. The mites were shipped in rice paper packages, resembling tea bags, (Wright and Riner 1979). Approximately 100-200 mites were released on the withers of each heifer three times: 19 December, 16 and 29 January.

All calves were treated for chewing louse, *Bovicola bovis* (L.), on 9 January 1985, with one gallon of 0.05% lindane applied by a 1-gal hand sprayer. Each of the five stanchioned calves was retreated 8 February with 1 gal of 0.5% malathion applied by a hand sprayer for the sucking louse, *Linognathus vituli* (L.)

Sampling for mites was done (1) by scraping a 25 mm^2 area of skin and (2) by vacuum (Callcott 1985; French and Callcott 1987). A household vacuum cleaner was used with an in-line screen support for Whatman® #4 filter paper and an

individual collecting head for each sample. Individual vacuum samples collected on filter paper and scraping samples were processed by soaking in 70% ethyl alcohol with 1% eosin, rinsing the sample on to lined filter paper, and observing at 10-12X with a stereoscopic microscope (Meleney et al. 1982; Callcott 1985; French and Callcott 1987).

Lesions on the five stanchioned calves were mapped weekly from 19 March to 6 June, following the grid mapping procedure of Guillot (1981a, b). Skin surface temperature of lesions and adjacent non-lesion areas was recorded by a Bailey Microprobe Thermometer, Model BAT-4. The foot of the probe was pressed against the skin for 30 s and the maximum temperature was recorded for 10 paired-samples taken from four stanchioned calves on 9 April, with body lesions from ca. 5 to 41%. Temperature and relative humidity were recorded by a hygrothermograph located under the open shed at 1.7 m above the floor decking.

Arithmetic mean of sample and standard error of the mean of the sample (SEM) were computed for number of mites per skin scraping and skin temperature data; the latter was also analyzed by the paired t-test.

RESULTS AND DISCUSSION

The weather conditions during this experiment differed from the data published by Guillot and Cole (1984). Maximum and minimum temperature and relative humidity recorded at Kerrville (Guillot and Cole 1984) and at Statesboro, in our experiment, are presented in Table 1. During the first 4 wk post-inoculation with *P. ovis* the mean maximum daily temperatures of 19.4°C at Statesboro exceeded that at Kerrville by 2.9°C and the daily low of 6.5°C was 1.8°C higher than at Kerrville; the relative humidities were essentially the same (Table 1).

Table 1. Mean daily temperatures and humidities at Statesboro, GA and Kerrville*, TX.

Week	Max. (°C) Stat†	Max. (°C) Kerr‡	Min. (°C) Stat	Min. (°C) Kerr	RH Max. (%) Stat	RH Max. (%) Kerr	RH Min. (%) Stat	RH Min. (%) Kerr
1§	20.8	24.4	11.6	12.2	99.9	100.0	69.0	41.0
2	15.3	12.2	2.5	3.3	90.4	100.0	40.9	55.7
3	17.5	18.2	1.7	3.2	100.0	100.0	31.6	37.3
4	24.0	11.3	10.2	0.4	100.0	100.0	53.3	44.8
5	22.2	19.9	9.6	0.9	98.6	100.0	45.6	25.8
6	24.4	13.2	2.5	3.1	90.4	100.0	40.9	40.2
7	20.1	6.6	1.7	-2.6	100.0	100.0	31.6	51.0
Mean Avg.	20.6	15.1	5.7	2.9	97.0	100.0	44.7	42.3

* Data from Guillot and Cole (1984).
† Stat = Statesboro, GA.
‡ Kerr = Kerrville, TX.
§ Wk 1 starting = Statesboro 29 January, Kerrville 18 November.

We were unable to determine the consequences of the unfortunate use of lindane to control lice in January. Control of *B bovis* was required to avoid problems with this mallophagan which may reduce *P. ovis* populations (Meleney et al. 1982). Although lindane is not currently recommended for control of psoroptic mange on cattle, it has been recommended in the past and some control has been reported

(Kemper and Peterson 1953; Wright 1980). The lindane, as applied, did not control the Anoplura and 1 mo later malathion was used to control *L. vituli* on the five stanchioned calves. Mites were applied 3 wk prior and 1 and 3 wk after the lindane spray. The lindane treatment may have negated the first inoculation with *P. ovis* and possibly the second; the third inoculation on 29 January, 20 d post-lindane spray, should have been successful in establishing mite populations on the unstanchioned calves. The first confirmed lesion with *P. ovis* was detected 7 wk after the lindane treatment on a stanchioned calf (4 wk after last mite inoculation = 2.8 mite life cycles).

Development of Psoroptic Mange on Stanchioned Heifers

Lesions with mites were confirmed 26 February on calf V, 4 wk after the last inoculation with *P. ovis*, and by 7 wk all five stanchioned calves had developed observable lesions. The initial lesions were scattered along the top line of the back and withers; later, other lesions developed along flanks and hips. Development of lesions varied considerably among the five calves (Table 2). Calf I had mange on 41% of the body 18 wk after the final inoculation. The other four calves responded more quickly with at least 50% of the body with lesions by wk 8, 11, 11, and 13 after final inoculation.

We define self-grooming as those behavioral actions taken by unrestrained cattle to maintain a healthy skin and hair coat. The principle grooming actions are tongue licks and rubbing against solid vertical objects (eg., tree trunks, fence post) (Kemper and Peterson 1953). Photographs were taken of calves III and IV, 1 h after release from stanchions on 1 May. The photographic slides were projected on a grid and lesions were estimated to cover 55 and 50% of the lateral aspects respectively, approximating the ratings of 57.8 and 54.9% on 30 April (Table 2). Bright red, highly vascular granulomas had developed under the scabs and were visable, in the photograph, where the scabs had been removed. In 1 h after release from stanchions, calves III and IV had removed ca. 10 and 20% of the scabs, respectively. We observed these calves attacking the scabs primarily with their tongues for the first h of unrestrained self-grooming and occasionally using a tree or fence post to rub accessable areas. A similar self-grooming behavior was observed with all calves when released from stanchions. To further test the effect of grooming, after 68.7% of body with lesions was recorded on calf V, it was released into a small feedlot and allowed to self-groom. After a 4 wk period when the active lesion rating had decreased to 0.5% (Table 2), calf V was restanchioned; 6 wk later the restanchioned calf had lesions over 46% of the body. Our data support the theory that self-grooming in cattle is a primary factor preventing development of psoroptic mange.

Mapping of the lesions was used to document the progression of mange. All five stanchioned calves were sampled by skin scraping at least twice between April 10 and 5 June; 33 of 46 samples were positive, $\bar{X} = 30.9 \pm 6.8$ SEM, range 1-206 mites. Free eggs were identified in 27 of the 33 positive samples. These mite counts were considerably lower than those of Guillot (1981a) whose counts exceeded 950 per scraping, and Guillot and Cole (1984) who reported mean counts exceeding 145 for wk 3, 7 and 10 post-exposure. The extent of the scabies lesions reported in those papers did not differ greatly from the data reported herein.

Inflammation due to psoroptic mange caused a localized and significant ($t = 2.432$, d.f. = 9, $P \leq 0.05$) increase in skin temperature. The surface of active skin

lesions had a mean temperature of 0.98°C higher than adjacent non-lesion skin. The lesion temperature mean was 35.8°C ± 0.35 SEM, n = 10, and the non-lesion mean was 34.8°C ± 0.15 SEM, n = 10. No *P. ovis*-free calves, stanchioned for extended periods of time, were available to check the effect of prolonged confinement on skin surface temperature. However, we observed on 9 April, that the skin temperature measurements were lowest on calf I with a 5.2% lesion rating and 0 mite count in skin scraping samples. Since psoroptic mites cause inflammation, a general increase in skin temperature was expected (Runnels et al. 1967). Our study demonstrates that psoroptic mange will develop on stanchioned cattle in southeastern Georgia similar to that demonstrated in central Texas (Fisher and Wright 1981; Guillot 1981a) despite some difference in environmental temperature.

Table 2. Percent of body with lesions/mite count* for stanchioned claves inoculated with *P. ovis* on 19 December, 16 and 29 January.

	Calf				
Date	I	II	III	IV	V
March 19	1.9	8.8	7.0	0.8	24.6
March 27	2.9	24.4	15.0	1.8	68.7
April 2	4.9	28.0	25.9	3.1	r†
April 9	5.2/0*	40.7/81	36.0/35	10.1/10	r/5
April 16	5.7	58.6	51.8	29.8	r
April 24	9.3	r	53.9	47.9	0.5
April 30	10.4	r	57.8	54.9/206	3.4
May 7	12.2	r	r	r	9.6
May 14	14.5	r	r	r	17.1
May 21	18.9/24	r/1	r/18	r/1	18.1/14
June 5	40.9/49	r	r	r	45.6/24

* total count of all stages, egg through adult, scraping sample.
† r = calf was released from the stanchion.

Transmission Attempt with Feed Lot Heifers

Calves allowed normal self-grooming in the feed lot showed no clinical signs of psoroptic mange, as no lesions were observed and no living mites were collected. These calves were sampled three times by standard scraping methods, 4, 16 January and 26 February. The vacuum method was also utilized on the last sampling date. The test was terminated on 26 February 4 wk after the final incoulation (= 2.8 life cycles of *P. ovis*) and 11 wk after the first exposure (= 7.7 life cycles of *P. ovis*). At termination of this phase of the study, winter hair coats had been shed and the weather was warm (see wk 4, Table 1).

The Statesboro feed lot infection test did not succeed in successful transmission whereas the Kerrville test did (Guillot and Cole 1984). There was a difference in method of exposing the cattle but the numbers of mites per animal were similar. The Kerrville test utilized a 1000 mite dose on each of two steers penned with six other steers (3 replicate pens), thus ca. 2000 mites for eight steers (= 250 mites/steer). In Statesboro each of the 11 heifers in the small feed lot received an estimated 100-200 mites. At Kerrville the six steers, receiving ca. 1000 mites, had mite populations of 145.2 ± 51.7 per skin scraping 3 wk post-exposure and nine of 18 steers penned with the six infested cattle developed psoroptic mange by wk 13. The number of mites transferred to an animal may greatly influence the

speed of extensive mange development. We terminated the feed lot test at the end of wk 4 because no living mites were detected by either the scraping or vacuum methods, the winter hair coats had been shed, of the onset of milder weather, and for economic reasons.

CONCLUSIONS

Our studies show that psoroptic mange will develop on stanchioned cattle in southeastern Georgia, similar to that demonstrated in central Texas (Guillot 1981a), also a non-endemic area. Self-grooming, after release from a stanchion, dramatically reduces the psoroptic lesions; however, restanchioning will again promote development of extensive lesions. Psoroptic lesions have a surface temperature of ca. 1°C higher than adjacent non-lesioned skin.

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NOTE

EFFECT OF COLONY DISTURBANCE ON RED IMPORTED FIRE ANT CONTROL[1]

Key Words: Red imported fire ant, *Solenopsis invicta*, colony movement, insecticide, Formicidae.

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Single mound treatments of red imported fire ants (RIFA), *Solenopsis invicta* Buren (Hymenoptera: Formicidae), may result in the lateral movement of treated colonies a few meters from the original site (Franke, O. F. 1983. Efficacy tests of single-mound treatments for control of red imported fire ants, *Solenopsis invicta* Buren. Southwest. Entomol. 8: 42-45). This lateral movement of treated colonies following insecticide application is a common complaint of homeowners. However, in many cases the homeowners disrupt the colony with a tool such as a rake before treatment [Lemke, L. A. 1986. Biological studies, control investigations, and public attitudes regarding the red imported fire ant, *Solenopsis invicta* Buren (Hymenoptera: Formicidae), in South Carolina. Ph.D. Dissert., Clemson Univ., Clemson, SC]. This study was conducted in order to evaluate the severity of mound disturbance on control of RIFA colonies and relocation of treated colonies.

The study was conducted in a Santee, SC, cattle pasture which contained 112 mounds per ha. On 27 May 1985, 36 circular plots measuring 30.5 m in diam were delineated with the location of both active and inactive mounds in each plot mapped (as described by Horton, P. M., J. B. Kissam, S. B. Hays, and G. W. Query. 1982. Chlorpyrifos aerosol mound injectors for control of the red imported fire ant. J. Georgia Entomol. Soc. 17: 478-484). Plots were separated from each other by a 6.1 m buffer zone. After being opened with a rake, all active mounds in plots used for the disturbed treatment were observed to contain both major and minor workers, as well as, sexual and worker brood and pupae upon being opened. Two mounds outside of each undisturbed plot were opened to check for colony structure in the field. All of these mounds also contained all castes.

Treatments consisted of Sevin® 80W, Spectracide® 2E, Dursban® 4E, Cessco® Accudose Aerosol (chlorpyrifos), and controls (no treatment or water only), in conjunction with the disturbance or non-disturbance of all active colonies in the plot. All insecticides were applied at the labelled rates, with drenches being applied at the rate of 3.78 liters of solution to mounds $\leq$ 0.305 m in diam and 7.56 liters to mounds $>$ 0.305 m in diam. One treatment was randomly assigned to each plot.

Colony activity was determined by slightly probing the mounds during the early morning hours, between 0800 and 1000 h, with a thin metal rod. Probing was conducted in the center of the mound as well as on 4 sides. A mound was considered active if ten or more ants came to the surface within three minutes of the disturbance created by the probing. All probing of mounds occurred when ambient air temperature was 70-75°F. Pretreatment determinations were made 4 h before the insecticides were applied.

Colonies were disturbed by raking through the mound for approximately 30 sec or until the mound was levelled in the case of extremely small mounds (.10 m in

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Table 1. Percent of disturbed and undisturbed red imported fire ant colonies which became inactive or relocated following treatment with one of four insecticides.

Treatment	No. mounds treated	Disturbed*	Weeks after treatment†				Percent relocation
			1	2	3	4	
Spectracide 2E (diazinon)	16	N	95.8a‡	95.8a	95.8a	95.8a	4.2d
Spectracide 2E	15	Y	86.7ab	86.7abc	86.7abc	86.7abc	13.3bcd
Dursban 4E (chlorpyrifos)	18	N	89.6ab	89.6ab	89.6ab	89.6ab	10.4bcd
Dursban 4E	19	Y	70.9cd	74.5bc	74.5bc	74.3bc	21.7b
Sevin 80WP (carbaryl)	20	N	93.3a	93.2a	89.8ab	89.8ab	6.7cd
Sevin 80WP	19	Y	58.9d	70.8c	70.8c	70.5c	19.1bc
Cessco Accudose Aerosol (chlorpyrifos)	21	N	74.8bcd	74.8bc	74.8bc	70.2c	20.9bc
Cessco Accudose Aerosol	23	Y	29.6e	29.5d	21.3d	17.2d	59.4a
Water	17	N	2.1f	2.4e	2.4e	1.4d	3.3d
Water	19	Y	4.0f	6.7e	6.7de	3.4d	7.5bcd
Control	18	N	0.0f	0.0e	0.0e	0.0d	0.0d
Control	23	Y	0.5f	5.4e	5.4de	4.5d	6.7cd

* Y indicates pretreatment disturbance. N indicates no pretreatment disturbance.

† Treatments were made on 28 May 1985.

‡ Means within the same column followed by the same letter, are not significantly different ($P > 0.05$); least squares difference test.

diam); the treatment was then immediately applied. all treatments were applied on 28 May 1985 between 1100 and 1400 h. Ambient air temperature was 82°F.

Colonies were evaluated for activity weekly for 4 wk following treatment. Relocation of all colonies in plots was recorded. Colonies were considered to have relocated if a new colony appeared within 10 m of an active colony which had been treated and had since become inactive. No new colonies, including those considered to have relocated, were noted in any plot 2 wk following treatment.

Data represent either the number of colonies that became inactive or the number of colonies that relocated after the insecticide application. These data were corrected for mortality in the control plots using Abbott's formula (Abbott, W. S. 1925. Method of computing the effectiveness of an insecticide. J. Econ. Entomol. 18: 265-267) and then subjected to analysis of variance (ANOVA) at the $P = 0.05$ level followed by the least squares difference test. Analysis was conducted on actual number of inactive or relocated colonies but for clarity mean number of colonies which became inactive or relocated following treatment have been reported in Table 1 as the percent of treated colonies which became inactive or relocated. Colonies which relocated and remained viable throughout the test were recorded as being viable and thus unaffected by the treatment.

The results are presented in Table 1. The disturbance of the colony before treatment with Spectracide 2E did not significantly ($P > 0.05$) affect control of treated colonies through 4 wk posttreatment. Control of disturbed colonies treated with Dursban increased during the second wk posttreatment. At 3 wk posttreatment, the mortality of disturbed and undisturbed colonies treated with Dursban was not significantly different. The other two treatments (Sevin 80W and Cessco Accudose Aerosol) were not as effective on disturbed colonies.

Mound disturbances, in addition to insecticide treatments, had a significant affect on relocation of colonies (Table 1). The highest percentage of relocation (59%) was in disturbed colonies that were treated with the aerosol. Colony movement caused by the other insecticide applications after disturbance of the mound was not significantly different ($P > 0.05$) from undisturbed treated colonies. Although no significant difference was observed, in all cases the percent of treated colonies which relocated was greater when they had been disturbed than when they were undisturbed. All treatments including water caused a certain amount of colony relocation regardless of whether the colony had been disturbed or not. It appears from this study that disturbing colonies prior to treatment with some insecticides can decrease the efficacy of some applications.

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AN IMPROVED ARTIFICIAL DIET FOR MEXICAN BEAN BEETLES[1] BASED ON HOST PREFERENCE[2]

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Abstract: Mexican bean beetle (MBB), *Epilachna varivestis* (Mulsant), larvae were simultaneously presented with either 2 or 4 modifications of Kogan's artificial diet. The diet most preferred by MBB in each test was then further altered and the resulting modifications tested for MBB preference. Based on a series of these free choice tests, a more attractive variation of Kogan's diet was produced. This modified diet reduced the amount of casein hydrolysate from 10 ml to 2 ml of a 10g/100 ml water suspension and substituted soybean protein and wheat germ oil for casein and corn oil, respectively. In addition, the modified diet contained 0.1 g of *Phaseolus lunatus* leaf powder. MBB larval development on this modified soybean protein diet was compared with MBB development on Kogan's diet with the wheat germ oil substitution and *P. lunatus* leaves. MBB developed best on *P. lunatus* leaves. Of the artificial diets, larvae developed better on the modified soybean protein diet, producing heavier pupae with shorter pupal development times.

Key Words: Mexican bean beetle, *Epilachna varivestis*, artificial diet, feeding preference.

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The Mexican bean beetle (MBB) is a pest of many leguminous plants in the United States. *Pediobius foveolatus*, a non-overwintering eulophid parasitoid, has been shown to be an effective control of MBB (Stevens et al. 1975; Reichelderfer 1979). Several generations of this parasitoid are possible each season and *P. foveolatus* has the potential of attaining high levels of parasitism among the MBB population (Stevens et al. 1975). However, in order for *P. foveolatus* to be used as an alternative to chemical insecticide control, large numbers of MBB must be produced to facilitate mass production of the parasitoid. Presently, MBB are reared on live plants of the genus *Phaseolus*. This method of production is costly, labor intensive, and requires a large amount of greenhouse space for production of bean plants. An artificial diet for the MBB would reduce these rearing problems.

An artificial media completely suitable for the production of MBB larvae and subsequent production of its parasitoid does not exist. However, Kogan (1971) developed a semi-liquid diet for MBB, based on a media formulated for chrysomelid beetles (Kogan 1969). Kogan's diet is marginally successfully in rearing 3rd and 4th instar MBB larvae although earlier instars cannot complete larval development on this media. Substituting wheat germ oil for corn oil in Kogan's diet slightly improves its quality for larval development (Culbertson 1984).

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The objective of this study was to evaluate the attractiveness of one modified diet media over another in order to make changes to Kogan's (1971) artificial diet, producing a diet more acceptable to the MBB. In addition, the most acceptable diet developed from these tests was evaluated for its nutritional adequacy. The data show that using soybean protein and wheat germ oil instead of casein and corn oil improves the attractiveness of Kogan's artificial diet. The addition of *Phaseolus lunatus* leaf powder further enhances the acceptability of the soybean protein diet. This soybean protein diet with leaf powder is also more nutritionally effective than Kogan's diet with the wheat germ oil substitution only.

MATERIALS AND METHODS

Early fourth instar larvae, reared on *Phaseolus lunatus* foliage in a greenhouse, were starved for 24 hours prior to tests. Diet modifications were compared in five experiments performed using either a two-choice or four-choice situation. Experiments were run in an environmental chamber at a constant 25°C with a 14 h light:10 h dark photoperiod. For feeding tests, flexible u-well microtiter filtration plates were cut into 5 × 5 well sections. The four corner wells were removed and seventeen of the resulting twenty-one wells were covered with labeling tape while the remaining four wells were left open. Sections of cellulose sponge, cut to fit into the wells, were soaked in the diet solution and deposited randomly into each of the four open wells. The twenty-one well section was then placed in a 60 × 15 mm petri dish lined with moistened cotton to maintain high humidity and to reduce diet evaporation. One larva was deposited per petri dish prepared in this manner. Each free choice test was replicated 36 times using one insect per replicate. Observations were made hourly for eight hours to determine the feeding preference of the larvae and the total number of feeding responses for each diet recorded. The most preferred diet in each free choice test was further modified and evaluated for preference.

The diets used in these experiments are modifications of Kogan's diet (K1) (Kogan 1971) with wheat germ oil substituted for corn oil (Tables 1 and 2). Modifications were based upon suggestions by Moore (1985) on how to improve a diet media. In the first free choice experiment, *E. varivestis* larvae were given the choice of either K1 or K1 with 1 ml of an acetone extract of *P. lunatus* leaves added. The acetone extract was prepared by adding 20 g of *P. lunatus* leaves to a blender containing 100 ml of distilled acetone. The mixture was pureed and then vacuum filtered through a Buchner funnel with qualitative filter paper. One ml of this extract was then substituted for 1 ml of water in the K + WGO diet. *E. varivestis* larvae were given the choice of four different diets in the second feeding choice experiment, K1 and three other diets (K2, K3, K4) with various amounts of casein and casein hydrolysate. In the third free choice experiment, K3 was modified by substituting soybean protein for casein and egg albumin for casein hydrolysate (diets K5, SP1, SP2). The fourth free choice experiment involved the addition of various amounts of *P. lunatus* leaf powder to SP1 (diets SPLP1, SPLP2, SPLP3). The leaf powder was prepared by air drying *P. lunatus* leaves, crushing them in a mortar, and sifting the crushed leaves through a no. 2 mesh screen to remove the stems. The leaf powder was added directly to the diet. In the final free choice experiment, SPLP1 was modified by the substitution of stigmasterol for cholesterol (SPLP4).

Table 1. Base constituents of all experimental diets for *E. varivestis* larvae.

Constituent	Quantity
Bactoagar	0.2 g
Alphacel	3.0 g
Wesson's salt mixture	1.0 g
Meso-inositol	0.1 g
B-vitamin mixture*	2.0 ml
Ascorbic acid	0.1 g
Formaldehyde	1.0 ml
Methyl paraben	0.4 g
Tetracycline	0.004 g
Sucrose	4.52 g
Distilled water	100 ml
KOH (1N)	to adjust pH to 7.2 - 7.4

* Mixture contained 2 g of Vanderzant B-vitamins and 0.05 g of choline chloride per 2 ml of water.

Based on the above tests, the most preferred diet, SPLP1, was evaluated as a nutritional replacement of K1 (Kogan's diet + wheat germ oil). Larvae were reared in a greenhouse on *P. lunatus* until early 3rd instar. Experimental larvae were placed singly in 60 × 15 mm petri dishes lined with filter paper. A 3.8 cm roll of cotton dental wicking was cut in half, dipped into one of the diet solutions, and deposited into the appropriate petri dish. The larvae fed on *P. lunatus* foliage were deposited singly into identical petri dishes lined with filter paper moistened with distilled water. Three leaf discs (1.5 cm diameter) were deposited into each petri dish prepared in this manner. The filter paper, dental wicking, and diet were changed daily as was *P. lunatus* foliage. There were 30 larvae for each of the two artificial diets and the *P. lunatus* check for a total of 90 larvae. Larvae were held in an environmental chamber at a constant temperature of 25°C and weighed individually on a Mettler balance at fourth instar, prepupal and pupal stanges, and at adult emergence. Larvae were observed for developmental times of each instar as well as for percent pupation and percent emergence. The data were analyzed with Waller-Duncan's Bayesian k-ratio t-test (k=100). All reductions in sample size were due to mortality during the course of the experiment.

RESULTS

Larvae in the first experiment preferred K1 over K1 containing a 1 ml acetone extract of *P. lunatus* leaves. Based on the total number of feeding responses, the larvae chose K1 an average of 72.5% of the time as opposed to 27.5% for the K1 with acetone extract. In the second experiment, K3 (with casein reduced to 1.0 g and casein hydrolysate reduced to 2 ml) was preferred by *E. varivestis* larvae over the other three alternatives (Table 3). In the third free choice experiment, MBB larvae showed preference for SP1, containing 1.0 g of soybean protein rather than casein and casein hydrolysate rather than egg albumin (Table 4). In the fourth test, the *E. varivestis* larvae showed a preference for SPLP1, containing 0.1 g of leaf powder (Table 5). In the fifth experiment, SPLP1 was preferred over SPLP4 (with stigmasterol in place of cholesterol) (Table 6).

ERRATUM

A line was omitted from Table 1 on page 84 of the Vol. 5 No. 1, January 1988 issue. The authors apologize for this ommision and any inconvenience it may have caused. Table 1 should be as follows:

Table 1. Base constituents of all experimental diets for *E. varivestis* larvae.

Constituent	Quantity
Bactoagar	0.2 g
Alphacel	3.0 g
Wesson's salt mixture	1.0 g
Meso-inositol	0.1 g
B-vitamin mixture*	2.0 ml
Ascorbic acid	0.1 g
Formaldehyde	1.0 ml
Methyl paraben	0.4 g
Tetracycline	0.004 g
Sucrose	4.52 g
Wheat germ oil	1.0 ml
Distilled water	100 ml
KOH (1N)	to adjust pH to 7.2-7.4

* Mixture contained 2 g of Vanderzant B-vitamins and 0.05 g of choline chloride per 2 ml of water.

Table 2. Composition of experimental diets. Experimental constituents below were added to base constituents to form experimental diets.

Constituents	K1	K2	K3	K4	K5	SP1	SP2	SPLP1	SPLP2	SPLP3	SPLP4
Casein (g)	1.0	2.5	1.0	0.5	1.0	—	—	—	—	—	—
Soybean protein (g)	—	—	—	—	—	1.0	1.0	1.0	1.0	1.0	1.0
Casein hydrolysate† (ml)	10	5	2	1	—	2	—	2	2	2	2
Egg albumin (ml)	—	—	—	—	2	—	2	—	—	—	—
Leaf powder‡ (g)	—	—	—	—	—	—	—	0.1	0.2	0.5	0.1
Cholesterol acetate (mg)	50	50	50	50	50	50	50	50	50	50	—
Stigmasterol (mg)	—	—	—	—	—	—	—	—	—	—	50

* Materials added to base constituents shown in Table 1.

† ml of a 10 g/100 ml suspension.

‡ *Phaseolus lunatus* leaves were air dried, crushed, and passed through a sieve.

Table 3. Percentage of total feeding responses elicited from *E. varivestis* larvae in a four-way free choice diet test over an 8-hour period.

Diet*	% of total responses
K1	16.21
K2	21.74
K3	44.83
K4	17.22

* As defined in Table 2.

Table 4. Percentage of total feeding responses elicited from *E. varivestis* larvae in a four-way free choice diet test over an 8-hour period.

Diet*	% of total responses
K3	17.95
K5	20.40
SP1	40.76
SP2	20.89

* As defined in Table 2.

Table 5. Percentage of total feeding responses elicited from *E. varivestis* larvae in a four-way free choice diet test over an 8-hour period.

Diet*	% of total responses
SP1	27.39
SPLP1	33.14
SPLP2	18.55
SPLP3	20.92

* As defined in Table 2.

Table 6. Percentage of total feeding responses elicited from *E. varivestis* larvae in a two-way free choice diet test over an 8-hour period.

Diet*	% of total responses
SPLP1	58.70
SPLP4	41.30

* As defined in Table 2.

As a result of feeding preference tests above, SPLP1 was compared with the K1 and the host plant *P. lunatus* in developmental studies of *E. varivestis*. The larvae, whether on the artificial diets or on leaves, reached fourth instar approximately two days after being deposited on their respective diets. Larvae fed *P. lunatus* had significantly larger mean fourth instar, prepupal, pupal, and adults weights than larvae fed either artificial diet (Table 7). There were no significant differences in mean fourth instar, prepupal, or adults weights among larvae fed artificial diets. However, MBB larvae fed on SPLP1 produced larger pupae than larvae fed on K1. The mean fourth instar, prepupal, and pupal developmental times of larvae fed *P. lunatus* were significantly shorter than those of larvae fed either of the artificial diets (Table 8). There were no significant differences in mean fourth instar or prepupal developmental times between larvae fed the

artificial diets, but larvae fed on SPLP1 had significantly shorter mean developmental times than larvae fed K1.

The mean percent pupation was significantly greater for those MBB reared on *P. lunatus* foliage (Table 8). There was no significant difference in the percentage pupation of MBB fed either of the two diet solutions although more MBB pupated from those fed SPLP1 than those fed K1. There was no significant difference in percent emergence of MBB adults in any of the test groups (Table 9).

Table 7. Mean weights of 4th-instar, prepupal, and adult *E. varivestis* reared on two artificial diets and a *P. lunatus* control.

Diet*	n	Mean weight (mg ± S.E.)†			
		4th-instar	Prepupal	Pupal	Adult
P. lunatus	30	18.84 ± 1.62a	31.53 ± 0.61a	30.90 ± 0.60a	29.54 ± 0.65a
SPLP1	30	9.94 ± 0.44b	16.07 ± 1.35b	17.99 ± 0.42b	17.89 ± 0.93b
K1	30	10.80 ± 0.76b	18.4 ± 0.89b	15.07 ± 1.37c	15.12 ± 1.95b

* As defined in Table 2.

† Means followed by the same letter are not significantly different by the Waller-Duncan Bayesian k-ratio t-test (k = 100).

Table 8. Mean developmental times of 4th-instar, prepupal, and pupal *E. varivestis* reared on two artificial diets and a P. lunatus control.

Diet*	n	Mean developmental weight (days ± S.E.)†		
		4th-instar	Prepupal	Pupal
P. lunatus	24	4.67 ± 0.26a	1.92 ± 0.08a	6.16 ± 0.39a
SPLP1	15	13.00 ± 0.53b	4.07 ± 0.37b	7.30 ± 0.47a
K1	8	13.00 ± 1.08b	4.25 ± 0.76b	10.20 ± 1.99b

* As defined in Table 2.

† Means followed by the same letter are not significantly different by the Waller-Duncan Bayesian k-ratio t-test (k = 100).

Table 9. Mean percent pupation and adult emergence of *E. varivestis* reared on each of two artificial diets and a *P. lunatus* control.

Diet*	n	% pupation (± S.E.)†	n	% adult emergence†
P. lunatus	30	80.00 ± 7.5 a	24	79.17 ± 8.47a
SPLP1	30	50.00 ± 9.30b	9	66.67 ± 12.60a
K1	30	30.00 ± 8.53b	15	55.56 ± 17.56a

* As defined in Table 2.

† Means followed by the same letter are not significantly different by the Waller-Duncan Bayesian k-ratio t-test (k = 100).

DISCUSSION

Kogan's diet is an artificial media suitable for rearing third and fourth instar MBB larvae to adulthood (Kogan 1971). The initial diet for comparisons in the preference tests was Kogan's diet with the substitution of wheat germ oil for corn oil (K1) (Table 1 and 2). While this modification substantially improves the quality of MBB adults produced from the artificial diet (Culbertson 1984), neither Kogan's nor K1 can consistently rear early instar larvae to adulthood. These preference

tests were conducted to screen out less acceptable diet modifications and to find a more acceptable diet which might be more nutritionally effective than the K1 diet.

Chemical stimuli for feeding can be nutritive or non-nutritive (Moore 1985). MBB are stimulated to feed by the nutrients sucrose and amino acids (Augustine et al. 1964; Sutherland 1977; Davis 1968). These stimulants, however, are only effective within a narrow range of concentrations. For example, sucrose maximally stimulates MBB feeding at a concentration of 0.1 N and either higher or lower concentrations reduce the feeding response (Kogan 1971). The non-nutritive compound phaseolunatin can also stimulate MBB feeding at low concentrations (Lippold 1957; Nayar and Fraenkel 1963) but has an inhibitory effect at higher doses (Nayar and Fraenkel 1963).

Changing protein concentration and source may often improve a diet's quality (Moore 1985). Reducing casein and casein hydrolysate to 1.0 g and to 2 ml of a 10 g/100 ml water solution, respectively, substantially increased MBB preference for K1 (Table 3), suggesting that initial concentrations of these components may have inhibited feeding. Further reductions in casein and casein hydrolysate made the artificial diet less acceptable. Exchanging soybean protein for casein produced a more acceptable diet, but substitution of egg albumin for casein hydrolysate did not increase preference (Table 4). Altering the sterol in the diet made the diet less acceptable (Table 6).

Possible non-nutritive feeding stimulants were added to the artificial diet via an acetone extract or a dried leaf powder of *P. lunatus* leaves. The addition of 1 ml of the acetone extract made the diet much less desirable to MBB larvae. The extract may have contained inhibitory concentrations of nutritive or non-nutritive compounds, but it is possible that the small amoung of acetone itself added to the diet was sufficient to reduce the diet's acceptability. Soybean protein diet with 0.1 g of leaf powder was preferred over unaltered soybean diet or diets with more than 0.1 g of leaf powder (Table 5). Therefore, ca. 0.1 g of leaf powder appears to contain desirable quantities of non-nutritive and/or nutritive compounds.

Based on the preference tests, the soybean protein diet with 0.1 g leaf powder (Table 1 and 2) was the most acceptable diet formulation. Acceptance of the diet by MBB larvae, however, does not assure that the diet is nutritionally adequate (Jones et al. 1981). The development test evaluated the nutritional adequacy of the 'preferred' soybean diet (SPLP1) as compared to *P. lunatus* and the K1 diet. Results showed that the preferred diet was not as nutritionally effective as *P. lunatus* but was better than the K1 diet as this diet produced heavier pupae with shorter pupal development times (Tables 8 and 9).

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OBITUARY

Miss Frances McAlister

1909-1987

Members of the South Carolina Entomological Society were saddened by the passing of 'Miss Frances', 78, on Thursday, November 12, 1987. Services were held on Saturday, November 14, 1987, at the First Baptist Church of Pendleton.

Miss Frances was a Winthrop College graduate who worked for 44 years in the Department of Entomology at Clemson University. She assisted Mr. Franklin Sherman in establishing, building, and maintaining the Clemson University Arthropod Collection and South Carolina Faunal Survey; she had primary responsibility for the Collection and Survey for 20 years from 1954 until she retired in June of 1974. Over the years, she assisted in numerous research projects and provided much-appreciated professional help to many students and faculty.

From its very beginning in 1956, and over the years, Miss Frances was closely associated with the South Carolina Entomological Society, and was a Charter Member.

Born in Pendleton, South Carolina, she was the daughter of the late Lawrence and Minnie Kay McAlister. Surviving are brothers, Harold J. McAlister of Paduca, Kentucky, and William L. McAlister of Clemson.

Memorials may be made to the First Baptist Church of Pendleton, where she served as church clerk for many years, or to any charity.

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DISTRIBUTION AND MONITORING LESSER MEALWORMS, HIDE BEETLES, AND OTHER FAUNA IN HIGH-RISE, CAGED-LAYER POULTRY HOUSES[1]

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Abstract: Distribution of the lesser mealworm, *Alphitobius diaperinus* (Panzer), the hide beetle *Dermestes maculatus* DeGeer, and several fly predators were determined from cross-sectional and surface sampling of manure cones within environmentally controlled high-rise, caged-layer poultry houses. Most of the lesser mealworm adults and larvae were recovered from the lower-outer regions of the manure cone and the aisle between manure rows. Most of the fly predators, *Carcinops pumilio* (Erichson), *Macrocheles muscaedomesticae* (Scopoli), and an uropodid mite, were recovered at the crest and top side of the manure surface. The anthocorid, *Lyctocoris campestris* (F.), was found to be equally distributed over the manure surface.

The Arends tube trap, consisting of rolled, corrugated cardboard inserted into a length of polyvinyl chloride pipe, was evaluated as a monitoring technique for the lesser mealworm and hide beetle. Significantly greater numbers of lesser mealworm and hide beetle adults and lesser mealworm larvae were recovered from traps placed on top of the manure cone ($P < 0.05$) than three other positions within the manure pit (floor, post, and wall ledge). Manure tube trap sample sizes for monitoring lesser mealworm densities at five levels of reliability were calculated. Recovery of *C. pumilio*, and *L. campestris*, in the traps suggested population trends of other fauna could also be monitored with the tube trap.

Key Words: Lesser mealworm, hide beetle, Arends tube trap, high-rise poultry house, sampling.

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Two beetles associated with poultry manure and litter, the lesser mealworm or darkling beetle, *Alphitobius diaperinus* (Panzer), and the hide beetle, *Dermestes maculatus* DeGeer, are serious pests in Pennsylvania environmentally controlled rw,03high-rise, caged-layer poultry houses. Both species normally live in the accumulated manure, but large numbers may migrate throughout a house in search of a safe pupation site or to escape unfavorable conditions in the manure pit. These beetles cause structural damage when last instar larvae tunnel into insulation and structural materials to pupate (Jefferies 1979; Wildey and Wayman 1979; Ichinose et al. 1980; Vaughan et al. 1984). Large beetle populations may also become a public nuisance at manure clean-out because of migration from fields into nearby residential areas. Lesser mealworms are often reported to be the most abundant beetle inhabiting manure and litter in commercial poultry operations (MacCreary and Catts 1954; Legner and Olton 1970; Pfeiffer and Axtell 1980). Only moderate numbers of hide beetles have been reported in commercial poultry operations (Legner and Olton 1970; Pfeiffer and Axtell 1980).

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The lesser mealworm has also been shown to harbor several poultry pathogens such as *Escherichia coli*, *Salmonella typhimurium* (De las Casa et al. 1968), and the viral agents causing Marek's disease (Eidson et al. 1965). Lesser mealworms also serve as an intermediate host for several helminths (Alicata 1939, Case and Ackert 1940). Hide beetles are scavengers, attacking bones, carcasses, furs and hides, and structural damage has been reported by several other industries (Snyder 1920; Brimblecomb 1938; Hinton 1945).

Evaluations of insecticide applications and development of alternative control strategies for lesser mealworms and hide beetles requires a convenient, effective sampling method. Safrit and Axtell (1984) evaluated several relative sampling devices and found tube traps (suggested by J. J. Arends) consisting of rolled, corrugated cardboard inserted into sections of polyvinyl chloride (PVS) pipe were effective for sampling lesser mealworms in the litter of turkey and broiler houses. While placement of sampling devices was shown to affect recovery of lesser mealworm adults and larvae, Safrit and Axtell (1984) concluded the relative changes in mean beetle numbers would be useful indicators for monitoring control application needs. They calculated sample size estimates for monitoring lesser mealworm populations and found 10 traps adequate for routine monitoring at high beetle densities.

Based upon the success of the Arends tube trap in turkey and broiler houses, the trap was used to survey beetle populations in several Pennsylvania environmentally controlled high-rise, caged-layer poultry houses. The objectives of these studies were to: 1) examine the distributions of lesser mealworms, hide beetles, and other fauna within the manure pits, 2) examine Arends tube trap position effects in monitoring beetle populations and migration activity, and 3) determine sample sizes for monitoring with the tube trap.

MATERIALS AND METHODS

Studies were conducted at two environmentally controlled high-rise, caged-layer poultry houses in Lancaster county, Pennsylvania, and at an environmentally controlled high-rise, caged-layer poultry house in Union county which had abundant lesser mealworm and/or hide beetle populations. These high-rise houses were two-story, closed-sided structures. Laying hens were housed on the upper story in banks of tiered cages separated by walkways. The first and second Lancaster county houses held 64,000 and 50,000 hens, respectively. The Union county house held 45,800 hens. Manure accumulated in cone-shaped rows beneath the cage banks in a concrete pit on the first floor. The manure pits in the first and second Lancaster county houses measured 13.4 by 152.4 m and 13.4 by 121.9 m, respectively, while the Union county house measured 10.7 by 137.2 m. Pit walls were 2.4 m high: concrete blocks on the lower half and technifoam insulation, fiber board, particle board, or unpainted wood paneling on the upper half. Half the length of the upper wall in the first Lancaster county house was composed of unpainted wood while the newer half was composed of technifoam insulation. Ventilation was provided by fans mounted in the pit walls.

Beetle Distribution Within the Manure

Manure cone cross-sectional studies were conducted at the two Lancaster county houses and a manure surface study was conducted at the Union county house to determine lesser mealworm and hide beetle distributions.

Manure core samples were taken 28 October 1981 at five different locations among the manure rows in the pit of the first Lancaster county house. A sheet of galvanized metal was used to laterally divide the cone-shaped row of manure. While the metal sheet remained in position, manure was cleared from one side to expose the manure cone and provide room to take horizontal core samples. The exposed cross section of the manure cone was divided into 17 areas (Fig. 1, Inset). A second cut was made 0.30 m away and a 15.3 cm diam, 0.30 m deep horizontal core sample was taken at the center of each area. The samples were returned to the laboratory where adults and larvae of the lesser mealworm and hide beetle were manually extracted and counted. Manure had accumulated 28 weeks prior to sampling and the house had a high hide beetle population.

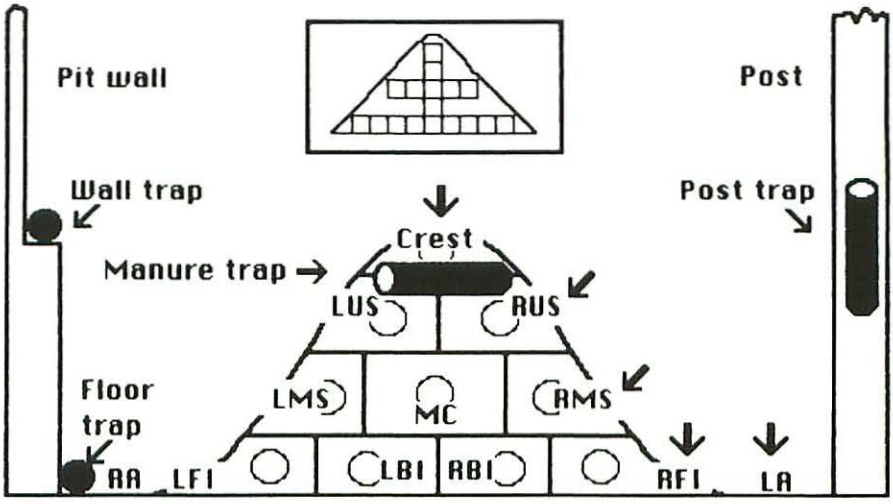

Fig. 1. Arends tube trap locations in the pit of a high-rise poultry house and sampling positions within the accumulated manure cone (Crest, upper side - US, middle side - MS, manure - floor interface -Fl, middle center - MC, bottom interior - Bl, and aisle - A. Right and left sides of the manure cone indicated by R and L prefix, respectively). Circles represent locations of horizontal core samples in the second cross-sectional study. Inset diagram represents interior core sample locations in first cross-sectional study. Arrows represent location of surface manure-profile samples.

Similar core samples were taken 8 October 1982 from two pit locations in the second Lancaster county house. The exposed cross sections of the manure cone were equally divided into seven outer and three inner areas (Fig. 1). In addition to lesser mealworm and hide beetle, distribution data were also collected for the histerid beetle, *Carcinops pumilio* (Erichson), a house fly egg and larval predator. Manure had accumulated 24 weeks prior to sampling and the house had high lesser mealworm and hide beetle populations.

At the Union county house, beetles and other fauna were sampled at four positions along the surface of the manure cone profile (Crest, upper side - US,

middle side - MS, and at the manure-concrete floor interface - Fl) and in the aisle (A) between manure rows (Fig. 1). A sample was taken at each position along the profile at five random locations (i.e. n = 5 each position) among the manure rows on 19 July and 13 August 1985. Manure was removed to a depth of ca. 6 cm using a 30 cm length of 5.1 cm diameter pipe to delineate the sample area (20.3 cm^2). All debris (i.e. feathers, manure), if less than 6 cm deep, was removed to the concrete surface at the aisle position. Samples consisting of only two replications at each position were taken 22 October and 12 November 1985 due to increased flooding in the pit from leaking waterers. Manure had accumulated ca. 16 weeks when the first samples were taken. Samples were placed into plastic bags and returned to the laboratory for separation, identification, and enumeration. Beetles were extracted using Berlese-Tullgren funnels consisting of a 4-mesh screen (ca. 10 cm^2) placed within a steel funnel (ca. 23 cm top diam.) below a 60 w incandescent bulb. Screw cap jars (62 ml) containing 80% ethanol were placed below the funnels to receive the arthropod specimens.

Trap Position and Beetle Migration

Since the number of beetles recovered in each sample by a particular sampling method is dependent upon their spatial distributions, Arends tube trap beetle recoveries were evaluated at several positions within the manure pit. The Arends tube trap consisted of a 43.2 by 22.9 cm piece of corrugated cardboard (brooder guard) rolled inside a 30.5 by 5.1 cm diameter PVC pipe. Each week, the cardboard rolls were removed from the tubes, sealed in plastic bags, and a new roll inserted into the pipe. Collected trap materials were returned to the laboratory, frozen for at least 48 hours, and the contents identified and counted. Weekly mean counts for each trap position were calculated.

An Arends tube trap was placed on the manure surface, aisle floor, and wall ledge at 10 stations around the pit periphery of the Union county house from 23 October 1984 to 26 March 1985 (Fig. 1). Manure had accumulated ca. 28 weeks prior to the start of the study. Ten additional traps were attached to support posts along the center axis of the pit. Traps on the surface of the manure were placed below cage support beams to minimize manure accumulation on top of the traps. Traps on the floor were placed against the cinderblock wall. Wall traps were placed on top of the cinderblock wall and traps on the posts were hung vertically from a nail in the post ca. 1-1.5 m above the pit floor. Manure and floor traps provided a measure of beetle distribution and relative abundance. Wall and post trap catches were expected to measure migrational activity and were compared with numbers of migrating beetles observed at wall and post counting stations.

Counting stations (0.093 m^2 or 1 ft^2) provided a measure of beetle activity or migration independent of the tube traps. Station counts were made by taking 'instantaneous' counts of beetles crawling within the 0.093 m^2 stations marked on pit walls and posts. Wall stations alternated between paneling (upper pit wall, 10 stations) and cinderblocks (lower pit wall, 10 stations). Ten stations were also located on the central support posts. As a measure of tube trap effectiveness in monitoring beetle migratory activity, the mean number of beetles at the wall and post counting stations each week were compared with wall and post tube trap weekly mean catches by Spearman's rank correlation, r_s (Steel and Torrie 1980).

Number of Samples

The number of tube traps required for sampling lesser mealworm adults and larvae at a given level of reliability was calculated from Union county house manure tube trap data obtained in the trap position study and from monitoring from 14 May 1985 to 21 October 1986. Monitoring was interrupted from 21 January 1986 to 17 June 1986 because of an outbreak of avian influenza in the area. Using reliability defined as the coefficient of variability C, which is the standard error of the mean divided by the mean, optimum sample sizes can be described for the general case by: $n = (1/C^2)\ (s^2/\bar{x}^2)$ (Karandinos 1976). If the form of the parent distribution is known (e.g. Poisson, Negative binomial, or Binomial), optimum sample size formulae may be given in terms of the parameters (Karandinos 1976). However, Taylor (1961) related the variance (s^2) and the mean ($\bar{x}$) in the form of a power function ($s^2 = a\bar{x}^{\,b}$) which is independent of the parent distribution and is applicable to insect populations conforming to a variety of mathematical distributions. Optimum sample size-density estimate curves were calculated by substituting the variance term in the optimum sample size formula with $a\bar{x}^{\,b}$. The parameters a and b were estimated for both adult and larvae by the intercept and regression coefficient, respectively, of the least-square regression line of the log of the sample variance on the log of the sample mean.

Differences in the number of recovered beetles between trap positions were detected by analysis of variance (ANOVA) and Fisher's protected least significant difference (LSD) procedure (Steel and Torrie 1980). The Mann-Whitney U test was used to compare adult and larval recoveries at each trap position and the Kruskal-Wallis one-way analysis by ranks was used to detect differences in the distribution along the manure profile. All procedures except the LSD test were performed with StatWorks™ (Rafferty et al. 1985).

RESULTS AND DISCUSSION

Beetle Distribution Within the Manure

A total of 679 lesser mealworm adults and 1,725 lesser mealworm larvae were recovered from the five manure cross-sections at the first Lancaster county house. Most of the lesser mealworm adults (64.9%) and larvae (47.5%) were distributed in the bottom row at the four outside positions of the manure cone. The number of lesser mealworms decreased towards the center of the manure cone, although a large proportion of adults (17.1%) and larvae (20.6%) were recovered from the bottom row at the four inside positions. Only 4.3% of the adults and 9.0% of the larvae were recovered from the five interior, vertical positions. From the four middle positions, 12.7% of the adult and 22.0% of the larval lesser mealworms were recovered.

In the second Lancaster county house, a total of 1,672 lesser mealworm adults and 19,365 lesser mealworm larvae were recovered from two manure cross-sections. Again, most of the lesser mealworm adults (77.5%) and larvae (62.6%) were distributed through the lower and outer portions of the manure pile (MS and Fl positions on both sides of the manure cone). A smaller proportion of lesser mealworm adults (14.6%) and larvae (24.5%) were recovered from the top half of the manure cone (Crest, US, both sides). Only 7.9% of the adults and 12.8% of the larvae were recovered from the three interior positions (MC,Bl, both sides).

Similar results were observed in the proportion of lesser mealworms recovered along the 4 manure surface profile and aisle positions at the Union county house

(Table 1). There was significant difference in the distribution of lesser mealworm adults and larvae along the profile (Kruskal-Wallis H = 30.74, 20.32, adults and larvae, respectively, P < 0.001). Most of the lesser mealworm adults (42.2%) and larvae (53.9%) were recovered from the manure-floor interface. However, a large proportion (adults, 23.0%, larvae, 23.6%) were also recovered from the aisle. The lesser mealworm was the most abundant beetle at the Union county house.

Table 1. Mean number and percent of lesser mealworm (*Alphitobius diapernius*) adults and larvae and fly predators collected from manure-debris samples at five positions along the manure cone profile at the Union County high-rise, caged-layer poultry house (n = 14).

	$\bar{x}$ (± SEM) (%)				
Species*	Crest	Upper side	Lower side	Floor-interface	Aisle
Alphitobius diapernius adult	5.0 ± 1.0 (10.9)	7.5 ± 1.9 (16.3)	3.5 ± 1.0 (7.6)	19.4 ± 2.7 (42.2)	10.6 ± 1.5 (23.0)
Alphitobius diapernius larva	9.6 ± 1.6 (3.2)	22.8 ± 5.8 (15.9)	4.8 ± 1.3 (3.3)	77.4 ± 16.5 (53.9)	33.9 ± 12.3 (23.6)
Carcinops pumilio adult	14.4 ± 2.8 (38.7)	7.1 ± 2.5 (19.3)	9.2 ± 1.6 (24.9)	4.1 ± 1.0 (11.2)	2.2 ± 0.9 (6.0)
Carcinops pumilio larva	13.9 ± 2.6 (29.2)	12.2 ± 2.5 (25.6)	16.6 ± 3.3 (34.9)	4.6 ± 2.1 (9.6)	0.3 ± 0.2 (0.6)
Macrocheles muscaedomesticae	116.8 ± 24.9 (52.2)	52.1 ± 18.8 (23.3)	29.1 ± 10.5 (13.0)	14.1 ± 3.7 (6.3)	11.7 ± 8.4 (5.2)
Uropodidae	23.1 ± 7.8 (44.3)	9.1 ± 5.8 (17.4)	10.4 ± 7.1 (19.9)	8.1 ± 3.7 (15.6)	1.5 ± 1.1 (2.9)
Lyctorcoris campestris	6.1 ± 3.0 (19.3)	7.1 ± 3.0 (22.7)	5.6 ± 2.0 (18.0)	8.8 ± 2.4 (28.0)	3.8 ± 1.6 (12.1)

* Means along the manure profile for all species except *L campestris* were significantly different by Kruskal-Wallis one-way analysis by ranks (P < 0.05), Steel and Torrie (1980).

There were also significant differences in fly predator distributions along the manure surface-aisle profile (H = 13.00 to 32.50, P < 0.001) (Table 1). Most of the predators were recovered at the crest and upper side of the manure (*C. pumilio* adults, 58.0%; *C. pumilio* larvae, 54.9%; *Macrocheles muscaedomesticae* (Scopoli), 75.5%; Uropodidae, 61.7%). Predator numbers declined towards the base of the accumulated manure and aisle. Therefore, the location of core samples would strongly influence indices of predator abundance. There was no significant position effect for the anthocorid, *Lyctocoris campestris* (F.) (H = 3.62, P = 0.46).

Geden (1984) found similar predator distributions from a manure surface profile at 7 surface positions and a cross-sectional profile at 7 exterior and 7 interior positions of ca. 12 week old accumulated poultry manure. For example, the uppermost positions (Crest, LUS, RUS) were favored by 53.3% of the *C. pumilio* and 76.9% of the *M. muscaedomesticae* in the cross-sectional study (Geden 1984). In contrast to the surface distributions at the Union county house and in Geden's (1984) work, 410 of 606 *C. pumilio* adults (67.7%) and 184 of 314 larvae (58.6%) were recovered from the four lower and outer positions of the manure cone (MS and Fl, both sides) in the cross-sectional study at the second Lancaster county house. Only 132 (21.8%) *C. pumilio* adults and 74 (23.4%) *C. pumilio* larvae were recovered from the crest and upper sides of the manure. Many *C. pumilio* adults (21.8%) and larvae (23.6%) were also recovered from the three interior positions of the manure cone.

Faunal distribution on and within poultry manure is probably determined, to a large degree, by physiochemical gradients within the manure and the distribution of food or prey. Accumulated manure in high-rise poultry houses show temperature gradients from the surface to the interior of the manure and temperatures may exceed arthropod tolerances (Armitage 1985, Stafford and Collison 1987). The environment in the interior of the manure cone may also become thick, sticky, and anerobic; conditions unsuitable for foraging scavengers, predators, or their prey. The outer 8-10 cm of accumulated poultry manure appears to be the primary site of house fly, *Musca domestica* L., activity and subsequently that of predators of house fly immatures (Willis and Axtell 1968). Nearly all *C. pumilio* and *M. muscaedomesticae*, which prey upon house fly eggs or first instar larvae, were recovered by Geden (1984) in a narrow band on or below the manure surface. Predators would also be active near the manure cone peak because of fresh manure accumulations and house fly egg deposition near the cone peak (Willis and Axtell 1968; Stafford and Bay 1987). In contrast *L. campestris* was recovered from all sampled surface positions. However, the role of the anthocorid as a predator is unknown. The concentration of *C. pumilio* at the basal areas of the manure cone and its presence within the interior of the manure cone at the second Lancaster county house demonstrates other variables may alter their usual distribution. These beetles may have been responding to changes in prey distributions, feeding on alternative prey, or responding to lesser mealworm activity.

Unlike the predators, lesser mealworms were most abundant at the base of the manure cone and many, although only a small portion of the total recovered, were found deep within the manure cone. Lesser mealworms feed primarily on grain or stored food products (Sarin and Saxena 1975), although Alicata (1944) reported the beetles actually feeding on poultry manure. Spilled feed would tend to accumulate primarily at the lower edges of the manure cone. The tunneling activity of lesser mealworm larvae may aerate and dry the interior of the manure as it accumulates. This activity may also have permitted penetration into the manure cone by *C. pumilio*.

Extremely few hide beetle adults or larvae were recovered within the interior of the manure cone in the cross sectional studies, although both Lancaster county poultry houses had relatively large hide beetle populations. Cloud and Collison (1986) concluded hide beetle larvae should be most numerous in the aisles or along the edges of the manure cone where their primary food sources, spilled feed and eggs, should collect. The lesser mealworm population had surpassed the hide beetle population at the second Lancaster county house when the cross-sectional samples were taken (unpublished data) and may have influenced the hide beetle distributions. At the Union county house which had a large lesser mealworm population, only a small number of hide beetles were recovered from the manure surface samples. Distributional differences along the cone profile were not clear-cut. There were no significant position differences for hide beetle adults or larvae (adults, $P = 0.10$, larvae, $P = 0.43$), although a large proportion of the adults were recovered at the crest of the manure.

Trap Position and Beetle Migration

Arends tube traps were most effective in sampling beetle populations when placed on the manure surface. Significantly greater numbers of lesser mealworm adults, larvae, and hide beetle adults were recovered from traps placed on the

manure surface than all other trap positions (Table 2). There were no significant differences in the number of lesser mealworms recovered from tube traps located on the floor, posts, or walls. Like the manure profile study, the tube traps showed the hide beetle population at the Union county house to be extremely low. Weekly trap means never exceeded 3 and 6 adults and larvae per manure trap, respectively. There was no difference between the manure and floor trap catches of hide beetle larvae.

Table 2. Mean number of lesser mealworm and hide beetle adults and larvae collected from Arends tube traps placed at four positions in the pit of the Union county high-rise poultry house for 22 weeks from October 1984 through March 1985 (manure n = 213, floor n = 220, wall n = 220, and post n = 218).

Trap position	$\bar{x}$ (± SEM) trap catch*			
	LMW adult	LMW larva	HB adult	HB larva
Manure	156.25 ± 20.80a	11.31 ± 2.42a	0.60 ± 0.10a	1.12 ± 0.13a
Floor	5.01 ± 1.10b	0.83 ± 0.32b	0.23 ± 0.05b	1.15 ± 0.16a
Wall	0.64 ± 0.25b	0.07 ± 0.03b	0.07 ± 0.02bc	0.25 ± 0.04b
Post	3.50 ± 0.89b	0.15 ± 0.45b	0.04 ± 0.01c	0.48 ± 0.06b

* Means separated by the same letter in the same column were not significantly different by Fisher's protected LSD (α = 0.05),Steel and Torrie (1980).
LMW = Lesser mealworm, HB = Hide beetle.

Significantly greater mean numbers of lesser mealworm adults than larvae were recovered at all trap positions (Mann-Whitney U = 23.0, 53.0, 113.5, and 45.5 for the manure, floor, wall, and post traps, respectively, $P < 0.05$ for all traps) (Table 2). In contrast, the ratio of larvae to adults was 2.5:1 and 11.6:1 in the first and second manure cross-section studies, respectively. In the manure surface profile study, 2,008 lesser mealworm larvae and only 644 adults were recovered. Therefore, tube traps showed an adult lesser mealworm bias. Based on their longevity, adult lesser mealworm numbers could be expected to accumulate in the manure. Adults lived 77-703 days in the laboratory (Preiss and Davidson 1971), while the larval period averaged 48.5 days (Lancaster and Simco 1967).

Examination of temporal trends for the four trap positions reveal some population trends in the manure and floor and migratory activity on the pit walls and posts (Fig. 2 and 3). There was a steady and substantial increase in the number of lesser mealworm adults recovered in the manure traps and a smaller increase in the number of larvae recovered. Increased adult and larval lesser mealworm recoveries from the floor, post, and wall traps as the population in the manure increased suggested density pressures may be partly responsible for the migrational activity. However, peaks at the beginning of the study and around the tenth week indicates other factors may also be influencing beetle migration.

Comparison of the adult lesser mealworm post trap catches with counts of migrating lesser mealworm adults on the 0.093 m^2 post counting stations reveal similar trends which were significantly correlated (r_s = 0.536, P = 0.01). Adult lesser mealworm wall trap catches and wall station counts were not significantly correlated (r_s = 0.390, P = 0.07). Since the open ends of the wall traps were horizontal rather than vertical, they may not have been efficient enough to accurately

reflect the beetle population migrating up the walls and some other trap design should be developed for monitoring migrating populations. The time of day wall station counts were made may also influence wall station results because of possible diurnal cycles in beetle migratory activity. Trap counts, however, supported visual impressions of more lesser mealworm adults than larvae present on the walls and posts. Results obtained by Despins (1987) suggested larvae were not as capable as adults in crawling on a vertical concrete surface. Larvae may also be finding sufficient places to pupate in the manure, since pupae can be found 3-4 cm beneath the surface of manure cones where they have a safe pupation site (Vaughan et al. 1984). However, studies have shown larvae are responsible for the damage to insulation with adults following the larvae into the tunnels (Ichinose et al. 1980; Vaughan et al. 1984).

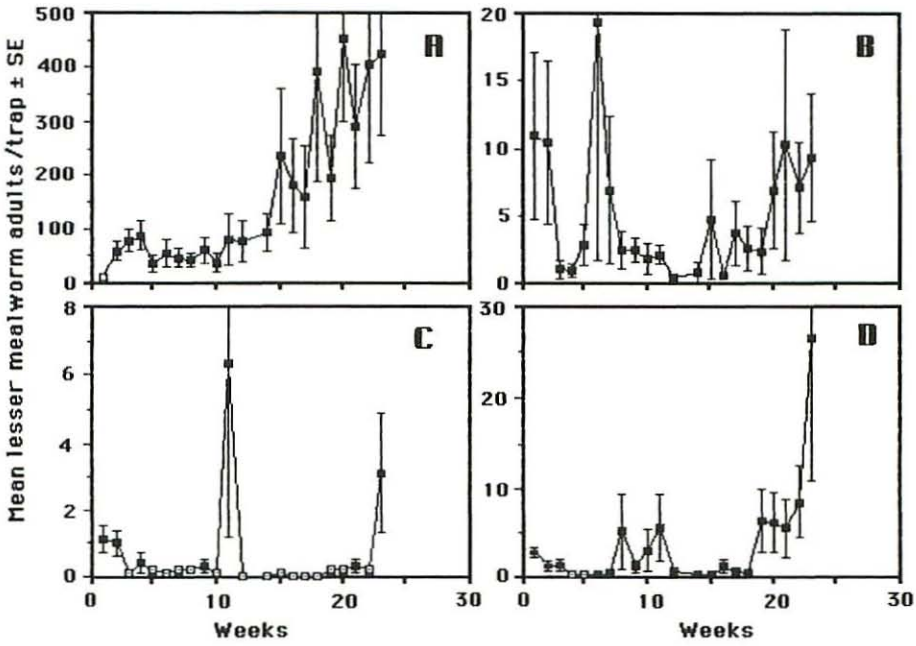

Fig. 2. Lesser mealworm adult population trends over the 22 week study period based on mean number of beetles collected from tube traps at 4 positions in the manure pit of a high-rise poultry house (A = Manure, B = Floor, C = Wall, D = Post).

The adult hide beetle population fluctuated during the same period of study, but appeared to increase slightly. Larval population fluctuations were erratic. During the first half of the study period, there was a large amount of larval floor, post, and wall activity. As the population increased in the manure, there was another increase in migration activity. Floor, wall, and post trap catches for both hide beetle stages generally corresponded with manure trap population trends.

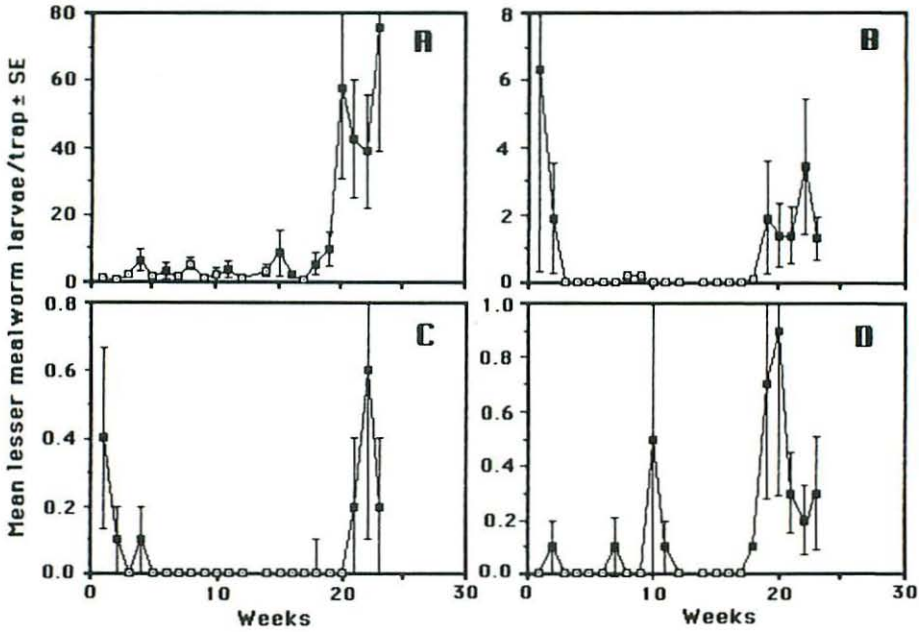

Fig. 3. Lesser mealworm larva population trends over the 22 week study period based on mean number of beetles collected from tube traps at 4 positions in the manure pit of a high-rise poultry house (A = Manure, B = Floor, C = Wall, and D = Post).

Number of Samples

An excellent fit of the regression lines, log s^2 on log $\bar{x}$, was obtained for lesser mealworm adults ($r^2 = 0.97$) and larvae ($r^2 = 0.96$). Since no significant differences between the residual variances, slopes, and elevations of the two regression lines were found ($P < 0.05$) (Snedecor and Cochran 1967), the data for both adults and larvae were pooled, producing a single regression of log s^2 on log $\bar{x}$ ($r^2 = 0.97$). This gave $s^2 = 2.897\bar{x}^{1.876}$ for Taylor's power law which was substituted into the equation for calculating optimum sample sizes.

Optimum tube trap sample sizes for estimating lesser mealworm densities are given in Table 3. The same levels of reliability and beetle densities given by Safrit and Axtell (1984) for turkey and broiler houses were used to allow direct comparisons. The number of traps required at any given level of reliability decreases with higher beetle densities. With a standard error of 20% of the mean, ca. 54-72 traps would be required at low beetle densities (≤ 10 beetles per sample) in high-rise houses, while ca. 35 traps would be required at high densities (≥ 350 beetles per sample). Safrit and Axtell (1984) found only ca. 10 traps were required at high beetle densities (≥ 350) for a reliability of 20%, but ca. 49-132 traps would be necessary at low beetle densities (≤ 10 beetles). Since a greater number of traps were required at high densities for high-rise houses, beetle densities were more variable at high densities in the high-rise houses than in the turkey and broiler houses. In contrasts, beetle

densities were less variable at low densities in the high-rise houses than in the turkey and broiler houses Safrit and Axtell (1984) studied. Therefore, lesser mealworm populations appear to be more aggregated within the pits of high-rise houses than in the turkey and broiler houses. Using only 10 traps to routinely monitor beetle populations, a reliability of only ca. 40-50% would be obtained for the beetle trap numbers typically recorded at the Union county house. An error of 30% of the mean could be obtained by increasing the number of traps on the manure to ca. 20-25 traps.

Table 3. Number of Arends tube trap samples required at different adult and larval lesser mealworm densities (mean per sample) for 5 levels of reliability within a high-rise poultry house.

Beetle density	Number of traps				
	10%	20%	30%	40%	50%
1	289.7	72.4	32.2	18.1	11.6
5	237.3	59.3	26.4	14.8	9.5
10	217.7	54.4	24.2	13.6	8.7
20	199.8	49.9	22.2	12.5	8.0
40	183.4	45.8	20.4	11.5	7.3
60	174.4	43.6	19.4	10.9	7.0
80	168.3	42.1	18.7	10.5	6.7
100	163.7	40.9	18.1	10.2	6.5
150	155.6	38.9	17.3	9.7	6.2
200	150.2	37.5	16.7	9.4	6.0
250	146.1	36.5	16.2	9.1	5.8
300	142.8	35.7	15.9	8.9	5.7
350	140.1	35.0	15.6	8.8	5.6
400	137.8	34.5	15.3	8.6	5.5

In summary, most of the lesser mealworm adults and larvae were recovered in samples from the lower regions of the accumulated manure and aisle. With the exception of *C. pumilio* in one study, the fly predators, *C. pumilio*, *M. muscaedomesticae*, and the uropodid, were recovered at the crest and top side of the manure. Cross-sectional sampling of manure cones in houses with high hide beetle populations indicated that hide beetles do not inhabit the interior of the manure cone. Tube traps placed on the manure surface, where adults and larvae of the lesser mealworm and hide beetle are active, were the most effective. The tube trap was more effective in monitoring adult lesser mealworms than larvae. Although tube traps positioned on the crest and top side area of the manure cone were not located in the area of manure with the highest lesser mealworm population, relative changes in mean numbers per sample per week should be useful indicators of population trends and for evaluating control measures. Since lesser mealworm densities in the manure pits of high-rise houses are highly variable, ca. 20-25 tube traps would be required for sampling with a 30% reliability. Traps located on the crest and top side area of the manure cone should also provide information on relative population trends for *C. pumilio* and *L. campestris*.

ACKNOWLEDGMENTS

Permission by the poultry producers to use their farms in this study is sincerely acknowledged. A special thanks is given to Joseph Cowfer for making many trips to collect the tube traps and count trap catches. The assistance of June Ferry in handling data was also appreciated. Thanks are also extended to Alfred G. Wheeler, Jr. for identifying the anthocorid, *Lyctocoris campestris*. Authorized for publication as Paper No. 7743 in the Journal Series of Pa. State Univ. Agric. Exp. Stn.

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ERRATUM

In the article "Distribution and Monitoring Lesser Mealworms, Hide Beetles, and Other Fauna in High-Rise Caged-Layer Poultry Houses" by Stafford, et al. which appeared in the Volume 5, Number 2, April 1988 issue of *Journal of Agricultural Entomology* on pages 80-101:

Line 4 on page 89 should read "high-rise, caged-layer poultry houses. Both species normally live in the"

Line 12 on page 90 should read "corrugated cardboard inserted into sections of polyvinyl chloride (PVC) pipe were "

Line 11 on page 93 should read "sample size formulae may be given in terms of their parameters (Karandinos 1976)."

Line 1 on page 94 should read "(Table 1). There was a significant difference in the distribution of lesser mealworm."

Line 1 of footnote to Table 1 on page 94 should read "* Means along the manure profile for all species except *L. campestris* were significantly different by"

Line 2 of footnote to Table 2 on page 96 should be "protected LSD (α = 0.05). Steel and Torrie (1980)."

Line 5 from bottom of page 98 should be "(1984) found only ca. 10 traps were required at at high beetle densities ($\geq$ 350 beetles) for a"

Last line on page 98 should be "rise houses than the turkey and broiler houses. In contrast, beetle"

NOTE: These are not author's errors. The author had marked these changes in the final proof, and the correction was made on the final galley; however, when the final typeset copy was produced, the old disk was inadvertently utilized. The editor regrets any inconvenience caused by these errors.

AN INEXPENSIVE SUCTION TRAP AND ITS USE IN AN APHID MONITORING NETWORK[1]

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Abstract: An inexpensive, easily constructed suction trap is described and depicted. Notes are included on its use in a network for the detection and monitoring of flights of aphids injurious to crops.

Key Words: Suction trap, insect, aphid, trap network.

J. Agric. Entomol. 5(2): 103-107 (April 1988)

In the early 1950's suction traps were first developed and shown to be an effective tool for sampling migrating insects (Johnson 1950a and 1950b; Taylor 1951; Johnson and Taylor 1955). Since then, traps with various modifications have been designed and used in insect surveys (van Ark and Pienaar 1970; Taylor and Palmer 1972; Takagi 1978; Wainhouse 1980; Goodenough et al. 1983; Bidlingmayer and Evans 1985; Kimsey and Brittnacher 1985; Taubert and Hertl 1985).

In 1983, we began to set up a network of suction traps in eastern Washington State to monitor the seasonal distribution and abundance of aphids injurious to crops. The trap that we designed for this network is simple and inexpensive to build and can easily be erected without special equipment. Construction and erection of the trap require approximately $300 worth of materials (1987 prices) and 20 man-hours of labor. Currently 17 of the traps are in use in the network; 13 in Washington State, and four in Oregon, Wyoming and California. An additional 36 are in use in several other networks in the western United States.

Trap design (Fig. 1). A fan draws air down an 8 m tube (sufficiently long to reach above most of the local insect populations [Taylor and Palmer 1972]) and through a screen funnel, where airborne arthropods are filtered out and collected in a jar of ethylene glycol. The housing consists of a 1.5 m section of 38 cm diameter polyvinyl chloride (PVC) plastic pipe attached with a coupler to a 6 m high section of 30 cm diameter PVC pipe. The screen funnel, made of aluminum or saran cloth (0.3 mm dia. strands, 12 strands/cm), is 85 cm long and has an upper opening of 38 cm and a lower opening of 5 cm. The upper end of the screen is attached with flexible steel strapping to the housing of the trap. A canning jar ring is attached to the base of the screen with silicon sealant and supported by brackets mounted on the housing. The ring holds a pint canning jar or a 500 ml plastic jar (Nalge, #2117, Rochester, NY). Access to the sample jar is through a door in the housing (20 cm × 25 cm). A 30 cm 3-wing aluminum fan (Dayton, #2C842, Chicago, IL) is mounted below the jar and is powered by an electric motor (Dayton, #3M568) with an output of 38 watts.

[1] Scientific paper no. 7912, College of Agriculture and Home Economics Research Center, Washington State University, Pullman. Project no. 0337. Accepted for publication 29 April 1988.

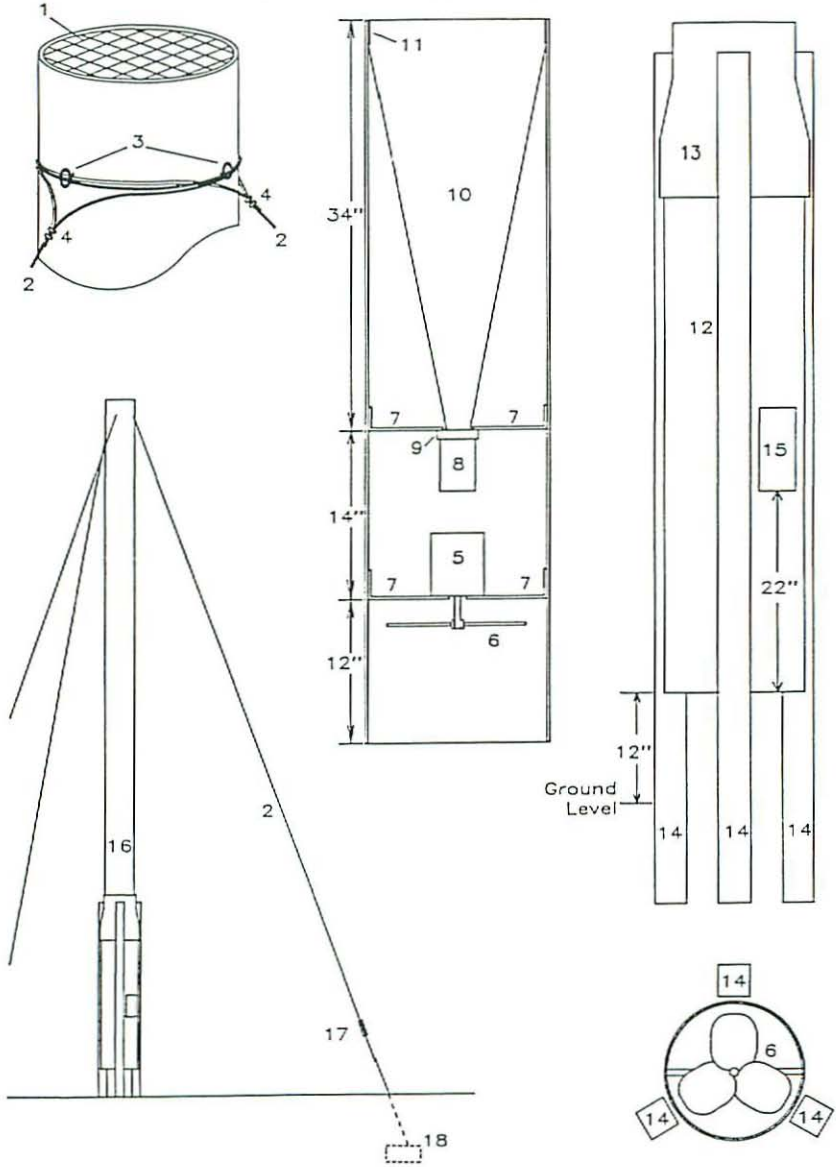
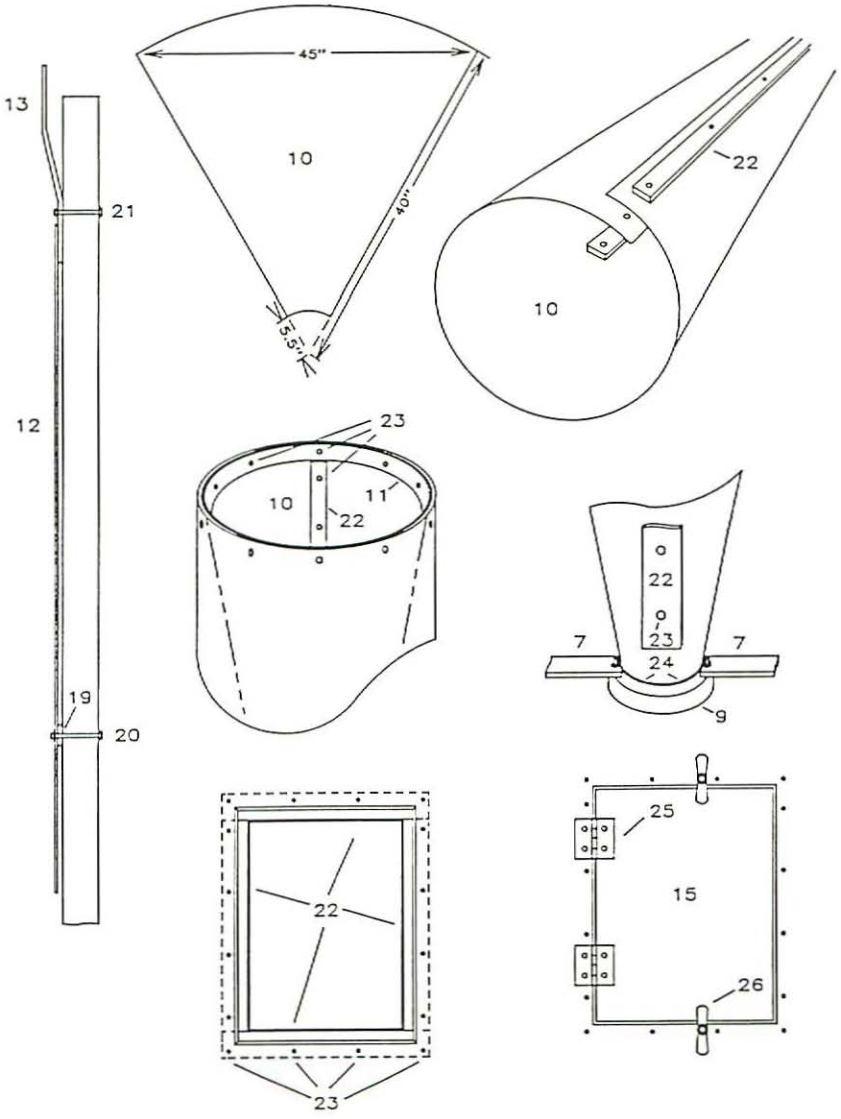


Fig. 1. **[1]** 25mm (1″) mesh hardware cloth **[2]** 3 mm (1/8″) aircraft cable **[3]** 2-15 mm × 6 mm (1/2″ × 1/4″) eye bolts w/ washers & nuts **[4]** Wire rope clips **[5]**Shaded pole motor, Dayton #3M568, 38 watt (1/20 HP) output **[6]** 30 cm (12″) 3-wing aluminum fan blade, Dayton #2C842 **[7]** 25 mm × 2 mm (1″ × 1/16″) strap steel (all steel should be painted) **[8]** 500 ml jar, Nalge #2117 or 1 pt canning jar **[9]** Narrow mouth canning jar ring **[10]** Aluminum wire or Saran cloth, .3 mm dia strands, 12 strands/cm (.012″, 30 mesh) **[11]** 25 mm × 30 mm (1-1/4″) steel strapping **[12]** 1.5 m (60″) length of low head 38 cm (15″) PVC pipe **[13]** PVC reducing coupler **[14]** 10 cm



× 10 cm × 2.5 m (4″ × 4″ × 8′) pressure treated posts **[15]** 18 cm × 22 cm (7″ × 9″) PVC door **[16]** 6 m (20′) length of low head 30 cm (12″) PVC pipe **[17]** Turnbuckle **[18]** Concrete block **[19]** Spacer from scrap PVC **[20]** 120 mm × 10 mm (5″ × 3/8″) hex cap bolt w/washers & nut **[21]** 115 mm × 10 mm (4-1/2″ × 3/8″) hex cap bolt w/washers & nut **[22]** 25 mm × 2 mm (1″ × 1/16″) aluminum strips **[23]** 3 mm (1/8″) blind rivets **[24]** Silicon seal, screen extends 3 cm (1″) below ring **[25]** 4 cm (1-1/2″) hinges **[26]** 4.5 cm (1-3/4″) turn buttons **[27]** Door frame

The trap is held 30 cm above the ground by three 10 cm × 10 cm × 2.5 m rot resistant wooden posts bolted to the sides of the base. Three lengths of 3 mm aircraft cable are wound around and fastened to the top of the trap and anchored to concrete blocks buried 1 m deep, 3 m from the posts. Turnbuckles are used to adjust cable tension.

The volume of air sampled by the trap was calculated by measuring the airflow with a hot wire anemometer (Hastings-Raydinst, #RA-1, Hampton, VA) inserted into the 30 cm PVC pipe at a level 2.5 m above the fan. Measurements were taken at a number of different ambient wind speeds (0 to 5 m/sec) and the following linear relationship developed: air sampled (m^3/min) = 14 - 2.2 × wind speed (m/sec) (r^2 = .82, df = 4, p = .95). Taking into account the average wind speed in this area (3.2 km/hr) the traps were estimated to sample ca. 570 m^3/hour, ca. 1/5 of the 2960 m^3/hr sampled by the traps used in the Rothamsted Insect Survey (Taylor and Palmer 1972). A sample of this size, while easy to sort and identify, may not be sufficient to characterize flights of aphid species that occur in small numbers; however it is adequate for monitoring many of the economically important species.

Information use. The traps are established on farms or agricultural research stations. The sample jars are changed weekly from April to November and sent to the Washington State University Agricultural Research Center in Prosser. As the samples are received, the aphids are removed and identified, without further preparation, using binocular microscopes of 12x-100x magnification. Identification is usually made to species level. All of the aphids from the samples are retained and representatives of each species are mounted on slides.

Each week a report is prepared and sent to interested scientists, extension personnel, consultants and farmers. The report includes the current week's suction trap counts of economically important aphids, comparisons of current flights with flights of previous years, and, at appropriate times, field counts of certain aphid species, especially grain infesting aphids. When available, information on economic population thresholds of aphids on some crops is also included.

The information acquired to date permits us to begin to characterize the flight and population trends of the region's aphids, allowing us to better interpret current information and to make observations and predictions of immediate value to farmers.

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MONITORING PYRETHROID RESISTANCE IN TOBACCO BUDWORM[1] IN LOUISIANA DURING 1987: RESISTANCE MANAGEMENT IMPLICATIONS[2]

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Abstract: The responses of over 2600 tobacco budworm, *Heliothis virescens* (F.), male moths collected in Louisiana from May through September 1987 to a discriminating dose (10 µg/vial) of cypermethrin indicated that the pyrethroid-resistance management plan recommended and adopted by Arkansas, Louisiana and Mississippi was successful in helping reduce resistant genotypes. During August and September 1986, survival of male moths in vials dosed with 10 µg of cypermethrin was 33-37%. During 1987, survival at the same dose was 20, 13, 18, 12 and 15% for males collected during May, June, July, August and September, respectively. The frequency of pyrethroid-resistant genotypes was generally highest in the northern part of the cotton production areas where most of the cotton acreage is located. Dosage-mortality data (LC_{50}'s and LC_{90}'s) on selected populations of tobacco budworm confirmed the results obtained using the 10 µg/vial discriminating dose.

Key Words: Tobacco budworm, pyrethroid resistance, insecticide resistance monitoring.

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Pyrethroid insecticides were introduced in the late 1970's for field use against cotton insect pests, particularly tobacco budworms, *Heliothis virescens* (F.), and bollworm, *H. zea* (Boddie). Previous laboratory and field research demonstrated that the pyrethroids were relatively more toxic to the tobacco budworm-bollworm complex than organophosphates and carbamates (Palazzo 1978; Pieters 1979; Twine and Reynolds 1980; Herzog and Ottens 1982; Martinez-Carrillo and Reynolds 1983). Although some field populations of tobacco budworms were shown to have an increased tolerance to pyrethroids via laboratory studies (Davis et al. 1977; Harding et al. 1977; Crowder et al. 1979; Twine and Reynolds 1980; Plapp 1981, 1984; Martinez-Carrillo and Reynolds 1983; Staetz 1985), no serious field control failures were documented in the laboratory until 1985. However Crowder et al. (1984) demonstrated in the laboratory that the tobacco budworm could become resistant to pyrethroids when selected for several generations.

During the 1985 season, tobacco budworm field control failures with pyrethroids were reported from areas in West Texas. Plapp and Campanhola (1986) later confirmed in laboratory tests that these field control failures were due to resistance. In 1986, pyrethroid resistance in tobacco budworm was documented in Arkansas (Plapp et al. 1987), Mississippi (Roush and Luttrell 1987), Louisiana (Leonard et al. 1987) and Texas (Allen et al. 1987; Plapp et al. 1987) using a variety of bioassay techniques. Moreover, pyrethroid resistance was confirmed in these states by

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Plapp et al. (1987) using a glass vial technique and field-collected tobacco budworm male moths. The primary resistance mechanism appeared to be kdr (knockdown resistance), which apparently is incompletely recessive in nature (Roush and Luttrell 1987; Payne 1987; Plapp and Campanhola 1986). Because of its recessive nature, this type of resistance mechanism (kdr) affords an excellent opportunity for resistance management.

In an effort to prolong the usefulness of the pyrethroids, pyrethroid-resistance management plans were developed for the mid-South states of Arkansas, Louisiana and Mississippi (Anonymous 1986) and for Texas (Plapp 1987). To determine if the pyrethroid-resistance management plans were successful, widespread monitoring of male tobacco budworm for pyrethroid resistance was conducted in these states during 1987 using Plapp's (1987) glass vial technique. Results of the pyrethroid-resistance monitoring program in Louisiana are reported herein.

MATERIALS AND METHODS

Wire cone traps (Hartstack 1979) baited with sex pheromone (Hendricks et al. 1987) were used to collect tobacco budworm male moths from May through September 1987. Although males were collected from most cotton production areas, more intense sampling was conducted on or near Dean Lee Research Station (Rapides Parish), Northeast Research Station-St. Joseph Location (Tensas Parish), Northeast Research Station-Macon Ridge Location(Franklin Parish) and Red River Research Station (Bossier Parish).

The *Heliothis* adult resistance monitoring technique developed by Plapp et al. 1987 was utilized to monitor the response of tobacco budworm males to cypermethrin. Briefly, the interiors of glass scintillation vials (20 ml) were coated with a residual film of cypermethrin (dosage levels ranged from 0.2 to 25 µg). The 10 µg/vial dose is known to be lethal to homozygous pyrethroid-susceptible moths as well as moths heterozygous for pyrethroid resistance (Plapp 1987). Since only homozygous pyrethroid-resistant moths survive this dose, it was used as a discriminating dose. Vials were stored in a dark area (to prevent photodegradation of cypermethrin) and used within one month after preparation. Acetone-treated vials were used as controls for natural moth mortality, which was generally less than 10%.

Male moths were removed from the traps early in the morning to prevent dessication. Only those males that appeared to be young and healthy were used in these tests. Two males were placed in each vial and held at room temperature (ca. 27°C) for 24 h. Mortality was determined by removing the moths from the vials and tossing them into the air. Moths that were unable to fly or could only fly a short distance (< 3 meters) were recorded as dead. All data were corrected for control mortality using Abbott's (1925) formula. Dosage-mortality data were analyzed using a micro-computer based probit analysis (MicroProbit 3.0, Sparks, T. C., and A. P. Sparks, unpublished, Louisiana State University, Baton Rouge, LA) after the method of Finney (1971).

RESULTS AND DISCUSSION

Over 2600 male tobacco budworm moths were bioassayed from May through September 1987 against the 10 µg/vial discriminating dose of cypermethrin (Table 1). Examination of these data revealed that (1) numbers of moths tested in 17 of the 34 bioassays were less than 50 (50 individuals required to detect a 10% level of resistance—see Roush and Miller 1986) and (2) variation in the levels of resistance was associated with location and date of bioassay.

Table 1. Responses of tobacco budworm male moths collected during 1987 to a discriminating dose (10 µg/vial) of cypermethrin.

Month	Date	Location	Number Tested	% Alive*
May	23	Red River Res. Stn.	58	17
May	26	Franklin Parish	35	28
June	9	Rapides Parish	18	11
June	10	Tensas Parish	60	3
June	10,12	Franklin Parish	104	17
June	12	Richland Parish	60	16
June	16	Rapides Parish	40	13
June	18	Gilliam	34	20
June	18	Red River Res. Stn.	20	6
June	27	Baton Rouge	18	7
July	25,28,30	Baton Rouge	38	0
July	28	Red River Res. Stn.	320	31
July	28	Gilliam	100	9
July	23,26,27,29,30	Dean Lee Res. Stn.	55	5
July	30	Franklin Parish	20	0
July	30	East Carroll Parish	12	0
July	21,22,28	Tensas Parish	129	6
August	18,19	Red River Res. Stn.	72	22
August	18	Gilliam	69	9
August	18	Derry	48	9
August	17,18	Dean Lee Res. Stn.	24	5
August	18	Cheneyville	18	17
August	6	Ouachita Parish	18	14
August	12,18	Tensas Parish	244	15
August	6,14,19	Franklin Parish	196	11
Sept.	1	Cheneyville	17	6
Sept.	1	Dean Lee Res. Stn.	38	3
Sept.	1	Derry	60	6
Sept.	1	Red River Res. Stn.	266	23
Sept.	1	Richland Parish	160	21
Sept.	9,10	Morehouse Parish	40	16
Sept.	17	Red River Res. Stn.	106	11
Sept.	26,30	Baton Rouge	42	2
Sept.	29,30	Red River Res. Stn.	58	16

* % alive is an estimate of the % homozygous pyrethroid-resistant males in each population sampled.

Summarizing the 1987 bioassay data by month (life cycle is about a month) and comparing it with similar data obtained in 1986 revealed that the overall level of resistance in August and September has decreased by about 50% (Table 2). Most of this decrease occurred by June 1987. Thus these data indicate that the pyrethroid-resistance management plan recommended and adopted by Arkansas, Louisiana and Mississippi was successful in lowering the frequency of pyrethroid-resistant tobacco budworms. The authors estimate that about 95% of the Louisiana cotton producers followed the pyrethroid-resistance management plan, a key component of which was to avoid the use of pyrethroids on cotton until July.

Summarizing the 1986 and 1987 bioassay data by locations revealed that the frequency of pyrethroid-resistant moths was generally higher in the northern part of the Louisiana cotton production areas than in the southern part (Table 3). This was expected since the proportion of the total acreage that is devoted to cotton

is less in the southern cotton production areas (hence less insecticide selection pressure). In an area where cotton is no longer commercially produced (Baton Rouge), the frequency of pyrethroid-resistant moths was only 2%. The frequency of resistant moths was 20% or greater in only two locations (20% in Richland Parish, an area where field control problems were experienced in 1986, and 23% at the Red River Research Station).

Table 2. Monthly summary of cypermethrin resistance monitoring data obtained using a discriminating dose of 10 µg/vial.

Month	Year	Number Tested	% Alive*
August	1986	40	33
September	1986	30	37
May	1987	93	20
June	1987	354	13
July	1987	674	18
August	1987	699	12
September	1987	787	15

* % alive is an estimate of the % homozygous pyrethroid-resistant males present.

Table 3. Summary by location of cypermethrin resistance monitoring data obtained using a discriminating dose of 10 µg/vial.

Location	Year	Number Tested	% Alive*
Tensas Parish	1986	30	37
Red River Res. Stn.	1986	40	33
Baton Rouge	1987	98	2
Rapides Parish	1987	210	8
Tensas Parish	1987	433	11
Franklin Parish	1987	355	14
Richland Parish	1987	220	20
Ouachita Parish	1987	28	14
Morehouse Parish	1987	40	16
East Carroll Parish	1987	12	0
Natchitoches Parish	1987	108	7
Caddo Parish	1987	203	11
Red River Res. Stn.	1987	900	23

* % alive is an estimate of the % homozygous pyrethroid-resistant males in each population sampled.

LC_{50}'s and LC_{90}'s were obtained for tobacco budworm populations from two locations in 1986 and six locations in 1987 (Table 4). With one exception, LC_{50}'s obtained in 1987 were lower than those derived in 1986. As expected, male tobacco budworm moths from the Baton Rouge area (low insecticide use area) exhibited the lowest LC_{50} (1.3 µg/vial). In contrast, LC_{90}'s of all populations examined in 1987 were considerably lower than those bioassayed in 1986 and tracked the data obtained using the discriminating dose (10 µg/vial) better than the LC_{50}'s. As reported by Roush and Miller (1986), comparison of LC_{50}'s can be misleading in interpreting resistance problems. Comparisons of LC_{90}'s (or higher points on the ld-p line) provide a better understanding of the frequency of resistant genotypes in the population sampled.

The pyrethroid-resistance management plan (Anonymous 1986) recommended by the Cooperative Extension Services of Arkansas, Louisiana and Mississippi was designed to delay further development of resistance in tobacco budworms by (1) avoiding the use of pyrethroids against any cotton pests until July, (2) using ovicides during periods of heavy oviposition by *Heliothis* spp. (ovicides appear to be equally effective against eggs of pyrethroid-resistant (RR) moths, pyrethroid-susceptible moths (SS) and heterozygotes (RS) and (3) using pyrethroid-synergist mixtures such as chlordimeform (pyrethroid-chlordimeform mixture is lethal to tobacco budworms that are heterozygous (RS) with respect to pyrethroid resistance-see Campanhola and Plapp 1987). Theoretically, all of these approaches should serve to reduce the number of pyrethroid-resistant (RR) tobacco budworms in a population, particularly in the case of the kdr type resistance mechanism, which is essentially recessive from a genetic standpoint (Payne 1987; Plapp 1987).

Table 4. Toxicological responses (LC_{50} and LC_{90} levels) of tobacco budworm male moths from various locations in Louisiana to cypermethrin.

Location	Date	Number Treated	LC_{50} µg/vial	LC_{50} 95% C.L.	LC_{90} µg/vial	LC_{90} 95% C.L.	Slope
Red River Res. Stn.	8/26/86	184	5.4	(2.37- 8.0)	31.1	(21.1- 66.7)	1.7
Northeast Res. Stn.	9/11/86	138	5.9	(0.20-15.5)	601.0	(90.5->9999)	0.6
Dean Lee Res. Stn.	8/17/87	34	3.1	(0.09- 5.5)	11.3	(6.2- 1416)	2.3
Dean Lee Res. Stn	9/ 1/87	98	2.9	(0.09- 4.9)	9.6	(6.5- 248.9)	2.5
Franklin Parish	8/19/87	200	3.0	(1.25- 4.2)	10.0	(7.9- 14.9)	2.4
Northeast Res. Stn.	8/12-14/87	210	2.7	(0.86- 3.9)	12.6	(9.3- 29.1)	1.9
Northeast Res. Stn.	8/18/87	134	4.8	(4.33- 5.3)	10.1	(8.7- 12.4)	4.0
Oak Ridge	9/20/87	213	4.0	(3.23- 4.8)	12.2	(9.4- 20.1)	2.6
Red River Res. Stn.	8/19/87	80	2.5	(0.08- 5.2)	15.2	(8.8->9999)	1.6
Red River Res. Stn.	9/ 1/87	430	6.5	(0.02- 9.1)	16.1	(11.1->9999)	3.3
Red River Res. Stn.	9/16/87	164	4.2	(0.46- 5.9)	12.8	(10.1- 38.2)	2.6
Red River Res. Stn.	9/28/87	156	2.7	(1.99- 3.4)	8.1	(6.2- 13.3)	2.7
Baton Rouge	9/26-30/87	372	1.3	(0.99- 1.6)	6.0	(4.4- 9.3)	1.9

Almost unanimous adoption of the pyrethroid resistance management plan by Louisiana cotton producers during 1987 was successful in helping reduce the number of homozygously resistant tobacco budworms by 50% or more in comparison with levels of resistant genotypes reported in 1986. Furthermore, there were no documented *Heliothis* spp. control failures with pyrethroids in 1987. This is in contrast to numerous reports of control failures in 1986, several of which were documented by laboratory bioassays as being due to pyrethroid-resistance (Leonard et al. 1987). However, it must be noted that *Heliothis* spp. infestation levels in cotton during 1987 were much lower than observed in 1986, particularly during the critical months of July and August. The most important implication of the pyrethroid resistance monitoring data herein presented is that resistance can be managed in some situations. Resistance to pyrethroids in *Heliothis armigera* (Hubner) has been successfully managed in Australia for several years (Gunning et al. 1984).

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ACTIVITY OF CHITIN SYNTHESIS INHIBITORS ON THE CAT FLEA, *CTENOCEPHALIDES FELIS* BOUCHE[1]

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Abstract: Three chitin synthesis inhibitors, alsystin, diflubenzuron, and cyromazine were tested against cat flea, *Ctenocephalides felis* Bouche, larvae. The chemicals were incorporated into the larval rearing media of 1.5, 2.5, and 3.5 day-old larvae. LC_{50}'s of 0.36, 0.09, and 0.94 ppm for alsystin, diflubenzuron, and cyromazine, respectively, were determined by probit analysis. As larval age increased, susceptibility to these chemicals decreased with diflubenzuron and cyromazine being toxic only when applied to the first two ages.

Key Words: *Ctenocephalides felis*, chitin synthesis inhibitors, diflubenzuron, cyromazine, alsystin.

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Juvenile hormone analogs, such as methoprene, offer considerable potential for flea control, as has been demonstrated with the oriental rat flea, *Xenopsylla cheopis* Rothschild (Chamberlain 1975, 1979; Chamberlain and Becker 1977). Several insect growth regulators (IGRs) with juvenile hormone analog activity have been reported to kill the cat flea, *Ctenocephalides felis* Bouche, larvae and pupae effectively, when tested in the laboratory (El-Gazzar et al. 1986). They incorporated IGRs into cat flea larval rearing medium and reported 99% reduction in emerging adult fleas using 13.3 ppb of methoprene, 15.9 ppb of fenoxycarb, 410 ppb of hydroprene, or 700 ppb of Pro-drone. However, alsystin, diflubenzuron, and cyromazine, three other IGRs with chitin synthesis inhibitor activity, provided little or no mortality of cat flea larvae or pupae in concentrations up to 20 ppm.

Diflubenzuron and alsystin are benzoylphenyl urea chitin synthesis inhibitors that kill insects by preventing the normal deposition of the cuticle (Hazzar and Casida 1979; Kramer and McGregor 1979; Mass et al. 1981). Diflubenzuron, for instance, causes mortality of immature insects during ecdysis (Mulder and Gijswijt 1973) and kills dipterans by preventing pupal cuticular formation (Wright 1974). Cyromazine is a substituted melamine insect growth regulator that kills dipterans by preventing normal pupation (Bloomcamp et al. 1987; Williams and Berry 1980), but secondarily has been documented as having chitin synthesis inhibitor activity (Miller et al. 1981). Since chitin synthesis inhibitors were reported to be most effective against newly hatched larvae (Friedel and Mcdonnell 1985) and since all larvae used by El-Gazzar et al (1986) were ca. 3.5 d-old (late second or early third instar) when exposure to these chemicals was initiated, we re-evaluated alsystin, cyromazine, and diflubenzuron with younger ages of cat flea larvae.

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MATERIALS AND METHODS

Three IGRs, alsystin, diflubenzuron, and cyromazine, were evaluated against three age groups of cat flea larvae (1.5 [late first instar], 2.5 [second instar], and 3.5 [late second or early third instar] d-old). The chemicals were applied as technical active ingredients dissolved in reagent grade acetone. Serial dilutions were formulated to provide a range of concentrations between 0.01 and 20 ppm (5 discrete doses for cyromazine (0.125-20.000 ppm) and 6 discrete doses for diflubenzuron (0.01-6.00 ppm) and alsystin (0.25-10.00) for each stage) in the larval rearing medium. The composition of the larval rearing medium was 125:20:3:2 portions of sand, pulverized laboratory rat chow (Purina rodent chow #2), dried blood meal, and brewer's yeast. The chemicals were applied using the same method of El-Gazzar et al. (1986) by applying 3 ml of the appropriate dilution in 75 g of rearing medium and shaking the mixture in a 500 ml Mason jar. The mixture was transferred to 250-ml waxed paper cups, and the acetone was allowed to evaporate under a fume hood for 30 min. Appropriate-age larvae (25-200) were then placed on the medium in each cup, and the cup was covered with orandy cloth secured with a rubber band. Treatments were replicated either 2 or 3 times to achieve numbers of > 500 larvae per age group for each chemical.

After 5-6 wk of incubation at 27°C and 75 ± 5% RH, the cups were uncovered and the numbers of cocoons were counted, and pupae and adults wree observed for morphological abnormalities. Data was adjusted for control mortality with Abbott's (1925) formula, and lethal concentrations were determined by probit analysis (Finney 1971).

RESULTS AND DISCUSSION

Table 1 presents the LC_{50}'s and LC_{90}'s for alsystin, cyromazine, and diflubenzuron when applied at different larval ages of the cat flea. Cyromazine was effective only when initially applied to media containing 1.5 or 2.5 d-old larvae. The LC_{50} for the 3.5 d-old larvae was higher than 100 ppm compared to 0.94 and 5.46 ppm for the 1.5 and 3.5 d-old larvae, respectively. A range of 4-90% of the emerged adults from cyromazine treatments of 2.5-20.0 ppm were elongated; whereas, the untreated adults were normal morphologically. At the higher doses, adults reached a maximum length ca. 1.5 times the length of a normal flea. Degree of elongation appeared to be dose related as reported for cyromazine in house flies that produce elongated pupae that failed to eclose (Bloomcamp et al. 1987; Mulla and Axelrod 1983a, 1983b).

Diflubenzuron was active only when applied initially to media containing 1.5 and 2.5 d-old larvae. The LC_{50}'s were 0.09 and 2.22 ppm for 1.5 and 2.5 d-old larvae, respectively. Diflubenzuron was reported by Chamberlain and Becker (1977) to inhibit cocoon formation completely of oriental rat fleas at 5 ppm in the diet of second instar larvae. Our data indicate that diflubenzuron is similarly as active against cat fleas with 90% inhibition of cocoon formation at 8.58 ppm in the diet of equivalent stage larvae.

Alsystin was more active against young larvae than older larvae (Table 1). The LC_{50}'s were 0.36, 4.15, and 12.80 ppm for treatments made to media containing 1.5, 2.5, and 3.5 d-old larvae, respectively. In the case of 1.5 d-old larvae. complete inhibition of cocoon formation was achieved at doses > 2.5 ppm.

Table 1. Lethal concentrations of three chitin synthesis inhibitors to cat flea larvae.

Age (d)	n	LC_{50} (ppm)	95% C.I.	LC_{90} (ppm)	95% C.I.	Slope
			Alsystin			
1.5	700	0.36	0.20- 0.53	2.15	1.42- 6.37	0.72
2.5	530	4.15	3.64- 4.76	13.47	10.76- 18.31	1.09
3.5	1,600	12.80	8.75-23.56	131.57	55.29->500	0.59
			Cyromazine			
1.5	600	0.94	0.88- 1.00	1.33	1.23- 1.51	3.67
2.5	850	5.97	4.47- 8.18	23.90	15.06- 57.23	0.73
3.5	940	ineffective*	----------	--------	--------	--------
			Diflubenzuron			
1.5	700	0.09	0.03- 0.14	0.51	0.33- 1.09	0.73
2.5	530	2.22	1.57- 3.12	8.58	5.33- 21.58	0.95
3.5	1,200	ineffective*	----------	--------	--------	--------

* Probit analyses with LC_{50} values > 100 ppm were considered ineffective.

The chitin synthesis inhibitors are potentially effective toxicants for control of the cat flea. However, bioassays of chitin synthesis inhibitors can be biased since first and second instar larvae are more susceptible than third stage larvae. Juvenile hormone analogs are most active just before pupation, and consequently, IGR's have been tested against late stage larvae regardless of whether the IGR is a juvenile hormone analog or a chitin synthesis inhibitor (El-Gazzar et al. 1986). The results of this study indicate that chitin synthesis inhibitors are most active against first instar larvae and bioassays of these compounds should be conducted with first instar larvae.

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FIELD EVALUATIONS OF DIFLUBENZURON AND TRIFLUMURON FOR CONTROL OF THE RICE WATER WEEVIL[1] IN CALIFORNIA RICE FIELDS

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Abstract: Two benzoylphenyl ureas (diflubenzuron and triflumuron) were evaluated at different rates and times of application in a 3 year field study to determine effectiveness in controlling natural populations of the rice water weevil, *Lissorhoptrus oryzophilus* Kuschel. Application timing was based on the number of days beyond ca. 50% mean rice emergence from the water surface in a continuously flooded field (ca. 10 cm depth). The chemicals were applied as single applications at 4, 5, and 7 days and as double applications at 4 and 10 days, and 7 and 14 days post rice emergence respectively. Three rates were tested: 0.14, 0.28, and 0.42 kg (AI)/ha. Both compounds caused significant reduction in immature weevil populations when applied at 0.28 kg (AI)/ha, 4 to 5 days following rice emergence in a continuously flooded field.

Key Words: Benzoylphenyl urea, diflubenzuron, triflumuron, *Lissorhoptrus oryzophilus*, rice water weevil, field tests.

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Diflubenzuron and triflumuron are benzoylphenyl urea (BPU's) compounds that inhibit the synthesis of chitin in arthropods. Numerous studies have indicated that these BPU's primarily affect the egg or larval stages of insects and that ingestion of these materials is generally required for activity (Retnakaran et al. 1985; Wright and Retnakaran 1987).

The rice water weevil (RWW), *Lissorhoptrus oryzophilus* Kuschel, is the major economic pest of rice in the United States during the initial growth and tillering stages of rice (Grigarick 1984). Larval feeding on rice roots causes major damage of economic importance and conventional insecticides are employed to control this life stage.

Smith et al. (1985) demonstrated that both diflubenzuron and triflumuron have ovicidal activity on the RWW. Eclosion was significantly decreased by the ingestion of or exposure to treated rice foliage by the adult female weevil. Reduced eclosion was also observed by exposing 0-1 day old oviposited eggs to treated water.

The purpose of this study was to evaluate diflubenzuron and triflumuron in the field for protection of the rice plant against the RWW. Timing of application was based on the following three facts: (1) maximum weevil oviposition in the field in California occurs during the first two weeks after rice emerges through the water, (2) maximum injury to the rice plant by the weevil is initiated during the same period (Grigarick unpubl. data), and (3) both BPU's have ovicidal activity against the RWW. Rates of application were based on greenhouse and small field studies which suggested that rates ranging from 0.14 to 0.42 kg (AI)/ha caused acceptable RWW immature reductions (Grigarick unpubl. data).

[1] COLEOPTERA: Curculionidae. Accepted for publication 13 May 1988.

MATERIALS AND METHODS

This study was conducted during the rice growing seasons of 1982, 1985, and 1986. Evaluations were based on number of RWW immatures found and grain yields. Possible effects on plant growth by either BPU, weevil larvae or both were analyzed by measuring plant wet weight, height, number of leaves and tillers, root length, and wet and dry root weight.

The experiments were conducted at the Rice Research Facility (Biggs, California). Twelve paddies measuring 4.3 m by 31.7 m in 1982 and 6.1 m by 44.5 in 1985 and 1986 infested with natural populations of the RWW were used for these experiments.

Standard cultural procedures recommended for rice fields in California were followed. All paddies were flooded to a depth of ca. 10 cm and seeded at a rate of 146 kg/ha using the rice variety M-201. In 1982 and 1985, paddies were hand-seeded 24 h after the initial flood with seed pre-soaked for 24 h in tap water. In 1986 paddies were machine dry seeded one day before flood.

In 1982 and 1986 copper sulfate was applied at 11.2 kg (AI)/ha for algae and tadpole shrimp (*Triops longicaudatus* [L.]) control one week following seeding. MCPA (52.1% EC, Dow, Midland, Mich.) was applied at 0.84 kg/ha 8 weeks following seeding for weed control.

BPU's were applied to the paddies with a CO_2 pressurized backpack sprayer utilizing a spray boom that spanned the width of the paddy from levee to levee. Spray suspensions were applied at 112 liter H_2O/ha.

In 1982 each paddy was divided in half by metal partitions placed in the center of each plot. A 30 cm space was left open on each side of the partition to direct water movement along the lateral margins of the paddies. Control plots and the lower rates were placed on the inlet half of each plot to prevent contamination of the control and to allow water in the low rate plot to flow along the margins of the high rate plot. Six treatments were randomized (within these constraints) in each of four blocks (each block contained 3 paddies [6 plots], one plot for each treatment).

All treatments were applied when the rice was 3 to 5 cm above the water surface (ca. 7 days post mean emergence). Triflumuron (25% WP, Mobay, Kansas City, MO) was applied at the rate of 0.14 kg (AI)/ha as a single application and also as a double application (7 and 14 days post mean emergence). Triflumuron at 0.42 kg (AI)/ha was applied as a single application. Diflubenzuron (25% WP, Thompson-Hayward, Kansas City, KS) was applied at the rate of 0.42 kg (AI)/ha as a single application and as a double application. The control plots were sprayed with water only. Plyac® (Fisher, Fair Lawn, NJ) was added as sticker to all spray suspensions at the rate of 0.33 ml Plyac/liter of spray.

Twenty five days after the first treatment, ten soil core samples (10.2 cm in diameter) with one plant per core were removed from each plot. Samples were taken randomly in a diagonal transect from one corner of the paddy to the opposite corner. Soil and RWW immatures were washed from the roots of the plants through a 0.84 mm screen and immature RWW's were counted by flotation. The plants were measured and weighed. Grain yield data was obtained by hand harvesting all plants in two, 1 m^2 quadrants per plot, 20 weeks following seeding.

In 1985, four treatments were randomized in a complete block design in each of 3 blocks (each block contained 4 paddies, one paddy for each treatment). Each paddy was maintained with a separate H_2O management.

Triflumuron (25% WP) was applied at 0.28 kg (AI)/ha as single adpplications at 4 and 7 days post mean rice emergence and as a double application at 4 and 10 days post mean rice emergence. Water was held in the paddies for 4 days after each application before a continuous flow was started again.

Ten soil samples were removed from each plot at 21, 37, and 43 days after the first application. Samples were processed for RWW immatures as described earlier. Paddies were harvested by a small field combine 24 weeks after seeding for grain yield.

The 1986 experimental design was identical to that used in 1985. All BPU treatments were applied at 5 days post mean rice emergence. Triflumuron (4F) was applied at 0.28, and 0.42 kg (AI)/ha as single applications. Diflubenzuron (25% WP, Uniroyal, Middleburn CT) was applied at 0.28 kg (AI)/ha as a single application. Paddy water was held for 7 days after each treatment before a continuous flow was started again.

Ten soil core samples were removed from each plot at 25 and 40 days post application and processed for RWW immatures as described previously. Paddies were harvested by a small field combine 20 weeks following seeding for grain yield.

All data were transformed by taking the square root of X + 0.5 (because of heterogeneity of variances for some of the means) and analyzed by two-way ANOVA. Duncan's (1955) multiple range test was used to separate treatment means.

RESULTS

During 1982, the most effective rate for either BPU was a single application at 0.42 kg (AI)/ha, applied 7 days post rice emergence (Table 1). Percent reduction of RWW immatures compared with the control was 69.8% for triflumuron and 74.7% for diflubenzuron. RWW counts from the triflumuron treatment at a single application of 0.14 kg (AI)/ha were not significantly different from the control. A double application of either BPU did not provide a significant advantage in reducing the number of RWW immatures. Significantly greater yeilds ($P = 0.02$) of rice grain were found for all treatments over the control. Although not significantly different from the other treatments, both BPU's at 0.42 kg (AI)/ha produced higher yields. Plant growth characters measured were not significantly different from the control but a consistent pattern in less growth for the control was noted for most characteristics except for triflumuron at 0.14 kg (AI)/ha.

Maximum oviposition in rice occurs during the first 2 weeks after rice emerges through the water. Because our applications ranged from 4 to 10 days after rice emergence we decided to sample RWW immatures 3 times over a 22 day period to give us some indication of the extended control capabilities of triflumuron. In 1985, all 3 treatment regimes significantly ($P = 0.02$) reduced RWW immatures over the control when sampled at 21 and 37 days following the first treatment (Table 2). At 42 days post-application, the number of immatures on plants treated with triflumuron applied at 7 days post-emergence was not significantly different from the control. In comparison to the control, triflumuron applied at 4 days post-emergence caused a greater percent reduction of RWW immatures at 21 days post-application, while triflumuron applied at 4 and 10 days post-emergence had a greater percent reduction at 37 and 42 days post-application.

Table 1. Control of rice water weevil immatures with diflubenzuron and triflumuron, 1982. Biggs, CA.

Treatment*	Rate kg (AI)/ha	No. applications	Avg no. immatures per plant	Percent reduction	Grain yield (g)/m²
Triflumuron	0.14	1	2.65bc†	14.0	629.2a†
Triflumuron	0.42	1	0.93a	69.8	744.1a
Triflumuron	0.14	2	1.18ab	61.7	666.1a
Diflubenzuron	0.42	1	0.78a	74.7	708.5a
Diflubenzuron	0.42	2	1.03a	66.6	698.6a
Control	—		3.08c	—	470.6b

* Treatments applied at 7 days post mean rice emergence and sampled 25 days post-application.
† Means in columns followed by a different letter are significantly different (P = 0.05; Duncan's [1955] multiple range test).

Table 2. Control of rice water weevil immatures with triflumuron, 1985. Biggs, CA.

	Number days sampled post application						
	21 days		37 days		43 days		
No. days post application*	Avg no. immatures per plant	Percent reduction	Avg no. immatures per plant	Percent reduction	Avg no. immatures per plant	Percent reduction	Grain yield (g)/m²
4 days	0.06a†	93.5	0.83a†	82.2	0.66a*	65.3	861.2
7 days	0.13a	86.0	1.60a	65.7	1.96b	—	868.0
4 & 10 days	0.13a	86.0	0.10a	97.9	0.10a	94.7	898.0
Control	0.93b	—	4.66b	—	1.90b	—	904.1

* Triflumuron applied at 0.28 kg (AI)/ha.
† Means in columns followed by a different letter are significantly different (P = 0.05); Duncan's [1955] multiple range test).

Plant growth characteristics for triflumuron applied at 4 and 10 days post-emergence when sampled 21 days post-application were generally higher than the control, although no significant differences were observed for any treatment regime. All growth characters at 37 days post-application for triflumuron applied at 7 days, and 4 and 10 days post-emergence were significantly different (P = 0.03) from the control, indicating increased plant growth. At 43 days post-application these two treatments again showed a pattern for increased growth but were not significantly different. Grain yield data showed no significant difference among any treatments.

During 1986, both BPU's were applied at 5 days post mean rice emergence. All insecticide treatments for the two sampling periods caused a significant reduction (P = 0.05) in RWW immatures (Table 3). Both BPU's applied at 0.28 kg (AI)/ha caused significantly greater weevil population reduction than triflumuron at 0.42 kg (AI)/ha at 25 days post-application. Triflumuron at 0.42 kg (AI)/ha however, caused an increased reduction of weevil immatures over the control from 36.5% on day 25 to 69.5% on day 40. Grain yields for triflumuron at 0.42 kg (AI)/ha were also significantly (P = 0.03) greater (20.6%) from the control. Yields for plots treated with both BPU's at 0.28 kg (AI)/ha were not significantly different from the control, but showed a 6.2% and 11.2% increase, respectively.

Plant growth characteristics showed a trend for less growth in the controls. The only significant difference (P = 0.01), however, was for average root length at 40 days post-application for plants treated with either BPU at 0.28 kg (AI)/ha.

Table 3. Control of rice water weevil immatures with diflubenzuron and triflumuron, 1986. Biggs, CA.

		Number days sampled post application				
		25 days		40 days		
Treatment*	Rate kg (AI)/ha	Avg no. immatures per plant	Percent reduction	Avg no. immatures per plant	Percent reduction	Grain yield (g)/m²
Triflumuron	0.28	2.27a†	72.6	1.97a†	64.6	627.7b†
Triflumuron	0.42	5.27b	36.5	1.70a	69.5	742.0a
Diflubenzuron	0.28	2.83a	65.9	2.43a	56.4	663.0ab
Control	—	8.30c	—	5.57b	—	589.0b

* Treatments applied at 5 days post mean rice emergence.

† Means in columns followed by a different letter are significantly different (P = 0.05; Duncan's [1955] multiple range test).

DISCUSSION

In 1985, the adult RWW infestation was lower than in 1986, since RWW immature counts were almost twice as low as 1986. The lack of significant differences in yield in 1985 may have been due to the fact that paddies were hand seeded in 1985 and the rice stand was not as uniform as the 1986 rice stand which was machine seeded. Apparently the lower RWW immature population level did not reduce yields or the non-uniform stand resulted in highly variable yields.

Both BPU's at 0.42 kg (AI)/ha gave adequate weevil control but in general this rate was not significantly different from the 0.28 kg (AI)/ha rate. Therefore, the most economical rate for RWW control in California rice fields appears to be 0.28 kg (AI)/ha.

Since adult RWW's begin to feed and oviposit on rice as soon as it emerges through the water, timing the application of these BPU's is critical. If applied too soon there is not enough leaf tissue present for sufficient chemical deposit. If applied to late viable eggs will have already been oviposited and adequate control will not be achieved. Our results indicate that applying either BPU at 4 or 5 days post mean rice plant emergence can provide good RWW control.

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FACTORS AFFECTING THE SUSCEPTIBILITY OF THE CAT FLEA, *CTENOCEPHALIDES FELIS* BOUCHE, TO CHLORPYRIFOS[1]

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Abstract: Anesthetizing adult cat fleas, *Ctenocephalides felis* Bouche, with CO_2 or by cooling (2-9°C) for periods ranging between 5 to 90 min did not cause significant mortality within the first 24 h after exposure. However, when similarly exposed fleas were exposed to chlorpyrifos there was a direct correlation with the length of exposure to the anesthetizing agents and the susceptibility of the fleas to this chemical. The age of the adult flea also affected its susceptibility to the chemical especially those greater than 48 h old. Handling and transporting of the immature fleas in the pupal stage did not affect their susceptibility to chlorpyrifos in the adult stage.

Key Words: Insecta, cat flea, Siphonaptera, Pulicidae, chlorpyrifos.

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The World Health Organization (W.H.O.) standard laboratory evaluation of insecticides against adult fleas involves exposing the insects to filter paper pieces treated with insecticides either for a fixed period of time using serial concentrations of the chemical or for a fixed concentration of the chemical and different periods of time (WHO 1981). The source of fleas and methods of handling are variables that may affect the results of experiments with insecticides. Some researchers use fleas from their own colonies, others use fleas which have been commercially purchased and transported long distances (El-Gazzar et al. 1986), and still others use fleas collected from domestic animals (Schwinghammer et al. 1985). Various methods of collecting and handling fleas have been reported. Schwinghammer et al. (1985) anesthetized fleas with CO_2, El-Gazzar et al. (1986) immobilized fleas by cooling before handling, and Sustriayu et al. (1980) handled them directly without any treatment. Also, Rust et al. (1980) is one of few researchers to report the age of the adult fleas which were used in pesticide evaluations.

Our goal was to investigate the effect of anesthetization of adults, adult age after emergence, and transportation of fleas on the subsequent sensitivity of cat flea, *Ctenocephalides felis* Bouche, adults to chlorpyrifos. Chlorpyrifos was selected since it was the most effective of 9 commercial insecticides tested (El-Gazzar et al. 1986) and is one of the most widely used chemicals for flea control.

MATERIALS AND METHODS

Mixed sexes of adult cat fleas from a colony maintained with previously published methods (El-Gazzar et al. 1986) at the USDA-ARS Insects Affecting

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Man and Animals Laboratory in Gainesville, Fla. were used in this study. Adult fleas were stimulated to emerge daily into 8-l utility jars by lightly breathing on 4-7 day-old cocoons. Moisture was provided with 25 ml cotton-stoppered, water-filled vials placed in the jars. The fleas were removed from the jars with a vacuum needle, and ten were placed in each test tube (2.3 cm diameter by 20 cm high). All fleas, except those used for the age test, were less than 24 h old. For each replicate within an experiment, fleas were randomly removed from the same jar so that sex ratios and age would not vary.

To study the effect of anesthetizing fleas with carbon dioxide or cooling, fleas were exposed in test tubes to a stream of CO_2 (58 ml/min.) or cooled on a cold-table (min. - max. = 2-9°C) for periods ranging between 5 and 90 min. After exposure, the fleas were tansferred from test tubes to utility jars and held for 1 h to recover. The fleas were then returned to test tubes (10 fleas/tube) and exposed for 24 h to Whatman no. 1 filter paper pieces (7.5 cm^2) treated with 0.8 $\mu g/cm^2$ of chlorpyrifos. The technical active ingredient dissolved in reagent grade acetone was placed on the filter paper using a pipette. Flea mortality was recorded after 24 h of exposure. Treatments were replicated 30 times for CO_2 and 40 times for cold, and the untreated controls consisted of flea exposure to filter paper strips treated only with acetone.

The effect of flea age on chlorpyrifos toxicity was evaluated by exposing fleas of known age to insecticide residues. Flea cocoons (4-7 day-old) were placed in 8-l glass utility jars, and emergence was stimulated by lightly breathing on the cocoons. The cocoons were then removed, and the adult fleas were held at $26 \pm 2°C$ and $50 \pm 2\%$ RH until reaching the desired age. Fleas were removed from holding jars at 3 h and at daily intervals from 1-7 d and exposed to filter paper treated with 0.8 $\mu g/cm^2$ of chlorpyrifos, as previously described. Treatments were replicated five times and included filter paper treated with acetone to determine control mortality. Mortality at 24 h was recorded.

To study the effect of transportation on flea pupae, 24-48 hour-old flea cocoons were driven to the airport, transported by a plane for ca. 4 h, then driven by car back to the laboratory. The cocoons were in transit ca. 24 h from initial packing to return to the laboratory. Adult fleas that emerged from the transported cocoons were collected in test tubes. Filter paper (7.5 cm^2) treated with 6 serial dilutions of chlorpyrifos (residues ranging from 0.1-2.0 $\mu g/cm^2$) was inserted into the test tubes with the adult fleas. Treatments were replicated 10 times. Mortality was recorded at 24 h and compared with mortality from insects that had been held under laboratory conditions.

Percentage mortalities were corrected by Abbott's formula (1925); means were separated with the Waller-Duncan procedure ($P > 0.05$; SAS Institute 1985) using arcsine transformed data. Lethal concentrations were estimated by probit analysis (Finney 1971).

RESULTS AND DISCUSSION

Exposing fleas to CO_2 gas or cooling 2-9°C (using a cold table) for periods up to 90 min did not accect percentage survival of untreated fleas (not exposed to chlorpyrifos) during the following 24 h (Table 1). When the anesthetized fleas were exposed to chlorpyrifos-treated filter papers for 24 h, there was a significant increase in mortality. For the CO_2-anesthetization treatment, only the mortalities

of fleas anesthetized for 60 and 90 min were significantly higher than the non-anesthetized fleas. Cold-anesthetization resulted in significantly higher mortality after treatment with chlorpyrifos. No significant differences in mortality between the CO_2- and cold-anesthetized fleas after exposure to chlorpyrifos was observed.

Table 1. Effect of anesthetizing adult cat fleas with CO_2 or cooling on their susceptibility to chlorpyrifos (n = 300-400).

Time of exposure	Mean % mortality* ± SE			
	CO_2†		Cold table‡	
(min)	Untreated	Chlorpyrifos	Untreated	Chlorpyrifos
0	0.00a	48.77a ± 4.47	0.00a	53.20a ± 3.31
5	1.33a	55.13ab ± 4.93	—	—
15	0.00a	60.50ab ± 3.79	0.67a	63.59b ± 3.53
30	0.67a	56.69ab ± 4.20	2.67a	62.67b ± 3.83
45	0.67a	57.38ab ± 3.33	1.33a	66.97b ± 3.52
60	1.50a	69.04b ± 3.80	0.67a	64.00b ± 4.14
90	1.67a	67.38b ± 3.62	2.19a	66.00b ± 3.85

* Means within a column followed by the same letter are not significantly different ($P > 0.05$; Waller-Duncan procedure [SAS Institute 1985]).
† Exposed in test tubes to CO_2 at a flow rate of 58 ml/min.
‡ Exposed on a cold table at 2-9°C.

Table 2 shows the effect of flea age after emergence on flea mortalities before and after exposure to chlorpyrifos. The data indicate that mortality of untreated fleas (not exposed to chlorpyrifos) increased significantly 3 days after emergence as the age of the flea increased from 3-4 or 5 days, and from 5-6 or 7 days. Flea mortalities after exposure to chlorpyrifos were significantly higher for 2-7 day-old fleas, compared to the mortalities of 3 h or 1 day-old fleas.

Table 2. Chlorpyrifos-induced mortality as affected by age of exposed cat fleas (n = 400).

Age (d)	% mortality* ± S.E.	
	Control	Chlorpyrifos-treated†
0.125 (3 h)	0.67a ± 0.67	43.31d ± 4.44
1	0.67a ± 0.67	44.14d ± 4.15
2	2.67a ± 1.18	61.08e ± 4.24
3	2.67a ± 1.53	59.76e ± 5.00
4	21.80b ± 4.29	66.80ef ± 4.13
5	24.13b ± 4.76	70.35ef ± 4.01
6	32.40c ± 3.71	66.05ef ± 4.74
7	35.00c ± 3.64	66.05f ± 3.91

* Means followed by the same letter are not significantly different ($P > 0.05$; Waller-Duncan procedure [SAS Institute 1985]).
† 24 h exposure to filter paper pieces treated with 0.8 µg chlorpyrifos/cm².

Transport of flea cocoons did not significantly affect the susceptibility of the emerged adults to chlorpyrifos. The LC_{50} values were 0.66 (n = 600, 95% C.I. = 0.46-0.93, slope = 1.22) and 0.79 μg/cm² (n = 600, 95% C.I. = 0.72-0.87, slope = 1.62) for untransported and transported adults, respectively, and they did not significantly differ, as determined by overlap of the 95% C.I.

Laboratory practices in handling and selecting fleas for laboratory experiments can stress fleas, and although this stress is not observed as decreased survival under normal conditions, use of stressed fleas can result in biased data. Anesthetizing adult fleas with CO_2 gas or cooling for periods ranging between 5 and 90 min seemed to affect adult cat fleas adversely. This effect was seen as increaased mortality after exposure to chlorpyrifos. Since only the 60 and 90 min CO_2 treatments produced significant mortality increases, shorter exposures to CO_2 gas, up to 45 min, or to a cold table (2-9°C) up to 90 min could still be used as reliable techniques for handling fleas for testing. Age and starvation was found to be a critical factor that affected the mortality of both unexposed and exposed fleas to chlorpyrifos. Mortality significantly increased with age. These results suggest that young fleas (< 48 h) should be used for laboratory toxicant evaluations. Also based on our results, transport of flea cocoons from commercial suppliers should not affect adult flea susceptibility to chlorpyrifos.

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PESTICIDAL ACTIVITY OF SUBSTITUTED BENZOSELENADIAZOLES[1]

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Abstract: Twenty-two derivatives of 2,1,3-benzoselenadiazole, substituted on the benzene ring with one or more methyl, halogen, or other groups have been prepared. When tested in a laboratory assay vs. *Panagrellus redivivus*, four of six compounds tested were lethal below 10 ppm, comparable to the best commercial nematicides. In insecticidal tests, some compounds were more active than carbaryl in housefly larvae, Indian meal moth larvae, and Indian meal moth reproduction assays, approximately as active as carbaryl against first instar yellow fever mosquito and confused flour beetle larvae, and much more active in a confused flour beetle reproduction assay. Unsubstituted benzoselenadiazole prevented *Tribolium confusum* reproduction at 10-30 ppm, making it the most active compound we have seen in this assay.

Key Words: Insecticide, acaricide, nematicide, selenium, piazselenole.

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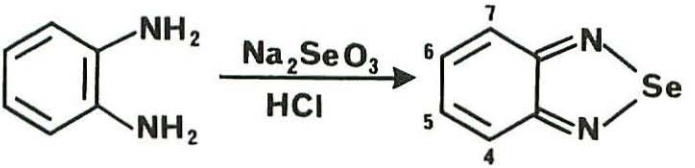
While the reaction of o-phenylenediamine with selenous acid to give benzoselenadiazoles has been known since the last century (Hinsberg 1889a, b), the derivatives have not been widely used for anything but microdetermination of



2,1,3-benzoselenadiazole
Piazselenole

selenium. (For two recent papers see Shimoishi (1977) and Dilli and Sutikno (1984)). A search in Chemical Abstracts revealed use as photographic antifoggants (Brown and Cheer 1971), analogous to benzotriazole (Kodak anti-fog #1, among other trade names), and additives to copper plating baths (DuRose 1969 and Andoniant et al. 1980). Unsubstituted benzoselenadiazole has been found to be

[1] This article reports the results of research only. Mention of a pesticide does not constitute an endorsement or a recommendation for its use by USDA, nor does it imply registration under FIFRA, as amended. Mention of a trademark or proprietary product does not constitute an endorsement or a recommendation for its use by the USDA. Accepted for publication 20 May 1988.

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moderately bacteriostatic against *Mycobacterium tuberculosis in vitro* (Takeda et al. 1952), and has been tested against Ehrlich ascites carcinoma (Takeda et al. 1955), but we could find no other reports of pesticidal efficacy of these materials. There are apparently no good mammalian toxicity data, although Hinsberg (1889a) reported that 400 mg of 5-methylbenzoselenadiazole killed 'a small dog' in a few hours. The route of administration was not given, but this represents fairly high toxicity.

Since some substituted benzoselenadiazoles prepared in a search for new selenium reagents (Wolf et al. 1988) showed encouraging biological activity in our insecticide and nematicide test systems, we decided to examine this class of compounds more thoroughly. This paper reports the insecticidal and nematicidal activity of these compounds.

MATERIALS AND METHODS

The benzoselenadiazoles were prepared by analogy with published methods (Shimoishi 1977; Dilli and Sutikno 1984). The o-phenylenediamines were purchased (Aldrich) or prepared by hydrogenation of the corresponding o-nitroanilines in acetic acid over palladium/charcoal in a Parr apparatus (25°C, 3 atm). After removal of the acetic acid on a rotary evaporator, the crude amine acetate could be used directly for the next step.

The amine or its acetate salt was dissolved in ca. 4-5 equivalents of dilute HCl, treated with charcoal if necessary, and 1 molar equivalent of sodium selenite was added with vigorous swirling. After standing for 1/2 hr, the voluminous precipitate was removed by filtration, dried and recrystallized from EtOH, $CHCl_3$, or mixtures of these solvents. Vacuum sublimation sometimes improved the color but did not change the melting point. GC/MS analysis (Finnigan 4500) was used to confirm structures of the desired products. All compounds were $\geq$ 99% pure by gas chromatography (Packard 421 instrument, 2 m × 4 mm glass column, 3% OV-1 on 100/120 mesh Chromosorb W-HP). Melting points (uncorrected) are listed in Table 1. Carbaryl was extracted from a commercial wettable powder formulation and crystallized to constant melting point. Methyl parathion (MP) was synthesized from dimethyl chlorothiophosphate (Aldrich) and the sodium salt of p-nitrophenol, followed by recrystallization from methanol.

The 'tea bag' test to assess activity against scabies mites (*Psoroptes cuniculi*, the rabbit ear mite, was used in this case) was described by Kochansky and Wright (1985) and Wright and Riner (1979). Groups of 20-25 adult mites and late instar nymphs in porous 'tea bags' were dipped for 30s in aqueous emulsions (concentrate = 25% test chemical, 65% xylene, 10% Triton® X-100) at various dilutions. Mortality was averaged over 3 bags. The check (a corresponding dilution of solvent/emulsifier only) gave mortality of 10-15%.

The nematicidal assay vs. *Panagrellus redivivus* was as described by Feldmesser et al. (1983). Approximately 400 nematodes per replicate, in all developmental stages, were exposed to a water/quartz sand/toxicant emulsion prepared using acetone, water, Tween® 20, and Triton® X-100 as emulsifiers. At least three replicates were run, and check mortality was < 10% (typically 3-4%).

Table 1. Physical properties of substituted benzoselenadiazoles.

Compound	Substituents	m.p., °C	m.p. (lit.), °C
1	H_4	71- 74	73* 73- 74†
2	4-Me	107-111	
3	5-Me	66- 70	70* 72- 73†
4	4,5-Me_2	120-124	
5	5,6-Me_2	143-145	
6	4,6-Me_2	154-157	
7	Me_4	154-155	
8	5-Cl	117-119	119* 121†
9	5,6-Cl_2	162-163.5	163* 163-164†
10	4,6-Cl_2	205-206.5	
11	Cl_4	241-244	
12	4,6-Br_2	215-218	217-218†
13	5-CO_2Me	147-151	
14	5-NO_2	220-223	222* 222-223†
15	5-CF_3	87- 88.5	91*
16	5-OCH_3	106-109	109-110†
17	5-OEt	99-100	102†
18	5-Bz	104-107	
19	5-NH_2	142-146	
20	5,6-Benzo	dec.>200	265 dec.*
21	4,5;6,7-dibenzo	208-211	
22	4-NO_2-6-CF_3	217-220	

* Dilli and Sutikno 1984.
† Shimoishi 1977.

The tests on house fly larvae (*Musca domestica* (L.)), confused flour beetle (*Tribolium confusum* Jaquelin duVal) larvae and adult reproduction, and yellow fever mosquito (*Aedes aegypti* L.) larvae were run in duplicate as described by Robbins et al. (1970). Briefly, for the house fly test, 75 eggs were reared aseptically on 3.15 g of sterile, semi-defined diet to pupariation and adult emergence. The test was run at 30°C, 55% RH, and in the dark. Control mortality ranged from 5-10%. In the *Tribolium* larval test, 40 newly emerged larvae were reared on 2 g of treated diet (white flour, whole wheat flour, brewers yeast, 4:4:1) to adulthood at 30° in the dark. Control mortality ranged from 0-10%. For the *Tribolium* reproduction test, newly-emerged adults (25 each sex) were reared on treated diet (1 g) for 10 days at 30°C, 55% RH, in the dark, then transferred each week for two weeks to 15 g of untreated diet. The number of progeny was determined for each 1-week period. For controls, adult mortality was 0-5% and each female produced 40-50 offspring. In the yellow fever mosquito larval test, 20 larvae of the first of fourth instar were reared in 100 ml distilled water at 25°, continuous light, and fed micropulverized dog meal. Test compounds are added in a maximum of 200 µl of MeOH. Control mortality is 0-25% (first instar) or 0-10% (4th instar) and successful adult emergence is the feature measured.

The Indianmeal moth (*Plodia interpunctella* (Hübner)) larval and reproduction tests were run as one continuous test. The larval part was as used in a bioassay of chitin synthesis inhibitor activity as described by Cohen and Marks (1979). Fifty eggs per duplicate test were reared in the dark on 5 g of treated diet at 30°, 55%

RH. On the tenth day, corrugated cardboard rolls were added for pupation. Pupae were counted just prior to adult emergence for a measure of larval toxicity. For the reproductive test, the same pupae were allowed to emerge, and the total egg production was measured, as well as the egg hatch. When egg laying had ceased, the adults were frozen and sexed. Control mortality in each part of the test is from 0-20%, and each female produces 60-140 eggs, depending largely on the male/female ratio.

The *Manduca sexta* larval test was as described by Robbins et al. (1975), Svoboda and Robbins (1967), and Svoboda et al. (1967). Five larvae per duplicate test were reared at 20°, continuous light, on artificial diet treated with the candidate material. We particularly looked for symptoms indicating interference with JH- or ecdysteroid-mediated processes such as precocious pupation, larval-pupal intermediates, molting difficulty, etc. To test for interference with steroid metabolism, the steroids of treated larvae were analyzed by the method of Svoboda et al. (1986). Control mortality was essentially zero (only occasionally did a control animal fail to pupate successfully) and successful pupation was the end point of this test.

All organisms tested were from established laboratory colonies and had not been selected for resistance to pesticides.

RESULTS

The results of the screening tests on insects are given in Table 2 with results of selected compounds vs. *Manduca sexta* in Table 3. None of the compounds was very active against mosquito larvae, with the best being about threefold less active than carbaryl against 4th instars. Other compounds were more active than carbaryl against this insect, however; methyl parathion was 100-fold more active against first instar larvae, and some of the chitin-synthesis inhibitors were more active yet, with LC_{75} below 1 ppb (Cohen and Marks 1979).

In the house fly larval test, fifteen compounds had activity equal to or greater than carbaryl, with the 5-ethoxy compound ca 1/10 as active as methyl parathion. A similar situation was observed with Indian meal moth. Very few materials in these tests were *less* active than carbaryl.

Most of the compounds were as active as carbaryl against *Tribolium* larvae, albeit less active than MP. In the *Tribolium* reproduction test, however, two compounds were outstanding (Table 2, last column). The unsubstituted benzoselenadiazole prevented reproduction at 10-30 ppm, and the 5-methoxy derivative prevented reproduction at 30-100 ppm. MP required 1000-3000 ppm and carbaryl did not prevent reproduction at concentrations as high as 10,000 ppm.

Six derivatives tested in the diet of *Manduca* larvae caused extensive mortality in the low ppm range (Table 3). No symptoms were observed suggesting disruption of JH or ecdysone metabolism. Gas chromatographic examination of the non-saponifiable lipids of *Manduca* larvae fed 1 ppm concentrations of the derivatives in Table 3 showed no apparent effect on sterol metabolism. Exact cause of death is unknown.

5-Chloro and 5-methoxy benzoselenadiazole killed *Panagrellus* at 2.5-5 ppm in the nematicide screen (Table 4). This represents very high activity, comparing favorably with aldicarb and carbofuran which are lethal at ca. 5 ppm in this assay.

Table 2. Biological activity of substituted benzoselenadiazoles (ppm).

Substituents	YFML 1	YFML 4	HFLT	IMML	IMMR	TCLT	TCRT
H_4	3-10	3-10	10- 30	100-300		100- 300	10- 30
4-Me	1- 3	3-10	30-100	100- 300			
5-Me	1- 3	3-10	10- 30	100- 300	30- 100	100- 300	1000- 3000
4,5-Me_2	1- 3	1- 3	30-100	100- 300			
5,6-Me_2	0.3-1	1- 3	10- 30	100- 300	30- 100	100- 300	300- 1000
4,6-Me_2	1- 3	1- 3	10- 30	30- 100			
Me_4	0.3-1	1- 3		30- 100	30- 100		> 10000
5-Cl	1- 3	3-10	10- 30	100- 300	100- 300	100- 300	1000- 3000
5,6-Cl_2	0.3-1	3-10	10- 30	100- 300	30- 100	100- 300	3000-10000
4,6-Cl_2	1- 3	3-10		100- 300	100- 300	1000-3000	> 10000
Cl_4	> 10	> 10		300-1000	100- 300	> 3000	> 10000
4,6-Br_2	3-10	3-10	100-300	> 300	> 300	100- 300	> 10000
5-CO_2Me	3-10	3-10	100-300	> 300	> 300	3000	> 10000
5-NO_2	3-10	3-10	10- 30	30- 100	30- 100		3000-10000
5-CF_3	3-10	3-10	30-100	100- 300	100- 300	100- 300	3000-10000
5-OCH_3	1- 3	3-10	30-100	100- 300	100- 300	100- 300	30- 100
5-OEt	1- 3	3-10	3- 10	30- 100	30- 100		3000-10000
5-Bz	3-10	3-10	10- 30	100- 300	100- 300	100- 300	> 10000
5-NH_2	0.3-1	3-10	30-100	100- 300	100- 300	100- 300	
5,6-Benzo	0.3-1	0.3-1	> 300	100- 300	100- 300	1000-3000	> 10000
4,5;6,7-Dibenzo	> 10	3-10	100-300	> 300	100- 300	1000-3000	3000-10000
4-NO_2-6-CF_3	3-10	3-10	10- 30	300-1000	300-1000	1000-3000	> 10000
Carbaryl	0.1-0.3	0.003-0.01	30-100	100-300	100-300	100-300	> 10000
Methyl parathion	0.003-0.01	0.003-0.01	0.3-1.0	0.3-1.0	0.3-1.0	> 1	1000-3000

Key to tests: YFML 1,4 Yellow Fever Mosquito (*Aedes aegypti*) larval test, first or 4th instar. HFLT House fly (*Musca domestica*) larval test. IMML, IMMR Indianmeal moth larval or reproduction tests. TCLT *Tribolium confusum* larval. TCRT *T. confusum* reproduction, respectively. Criteria of activity given in Materials and Methods.

Table 3. Activities of selected benzoselenadiazoles vs. *Manduca sexta* larvae.

Substituents	Effct	Concentration, ppm
H_4	100% larval mortality	2-4
5-Me	>90% " "	4-5
5,6-Me_2	100% " "	1-2
5-Cl	100% " "	1-2
5,6-Cl_2	63% " "	5
5-NO_2	84% " "	4

Tests against scabies mites (*Psoroptes cuniculi*) showed very low activity (Table 5), but the small differences in activity over two order of magnitude in concentration suggested that solubility may have been a problem, i.e. that a nearly saturated solution was being tested in each case.

DISCUSSION

The screening tests and especially the criteria used to define activity in those tests were rather severe. These tests were designed to provide the lowest concentrations necessary to kill 100% of test organisms or completely prevent

reproduction in replicated tests, but were not designed to collect statistically analyzable data as for probit analysis for LD_{50}, LC_{50}, etc. For insecticide and nematicide tests, the upper concentration gave 100% kill, while the lower concentration of the range did not (usually activity was $\geq$ 70%). Non-100% lethality was corrected by Abbott's formula (Abbott, 1925).

Table 4. Toxicities of selected benzoselenadiazoles vs. *Panagrellus redivivus*.

Substituents	Minimum LC_{100}
H_4	10-20
5-Me	5-10
5,6-Me_2	20-40
5-Cl	2.5- 5
5-CF_3	5-10
5-OMe	2.5- 5

Subject to these restrictions, selected compounds of this series showed impressive insecticidal activity against *Tribolium confusum*, *Musca domestica*, *Plodia interpunctella*, and *Manduca sexta*, and as nematicides against *Panagrellus redivivus* but had very poor activity against *Aedes aegypti* and *Psoroptes cuniculi*. There seemed to be no consistent trend of activity vs. degree or position of substitution.

Table 5. Activity of selected benzoselenadiazoles vs. *Psoroptes cuniculi*.

	% Mortality at Concentration		
Substituents	0.1%	0.01%	0.001%
H_4	6	0	0
5-CH_3	7	0	0
5-CF_3	26	0	0
5,6-Me_2	15	0	0
5-Cl	39	10	1
5,6-Cl_2	4	1	15
5-CO_2Me	46	45	21
5-OEt	94	10	13

Some of these compounds show appreciable volatility (the monomethyl isomers in particular have pronounced odors reminiscent of quinoline and lepidine). Because most assays involved treatments where insects were in intimate contact with vapor-containing air as well as the diet containing the chemicals, it is possible that some of the activity was the result of fumigant action. We have collected no data bearing directly on this possibility, nor on oil/water partition coefficients, mammalian toxicity, environmental persistance, or any of the other things necessary for further consideration of the practical application of these compounds.

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COMPARATIVE TOXICITY OF SEVEN INSECTICIDES TO ADULT *SPALANGIA CAMERONI* PERKINS[1]

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Abstract: The toxicity of seven insecticides used for control of muscoid flies on dairies was evaluated against adult male and female *Spalangia cameroni* Perkins, an important biological control agent of fly pests. The toxicity of the insecticides decreased in the order of dimethoate > crotoxyphos > permethrin ≃ dichlorvos ≃ Pyrenone > fenvalerate. Male *S. cameroni* were more sensitive than females to crotoxyphos, permethrin, and fenvalerate. Females were more sensitive than males to dichlorvos, while there was no significant difference between the LC_{50}s of dimethoate and Pyrenone for each sex. The tetrachlorvinphos bioassay revealed a distinct plateau at ca. 55% mortality, suggesting that resistant individuals were present in this population.

Comparison of *S. cameroni* and *Musca domestica* L. toxicity data (μg/vial) revealed that fenvalerate, permethrin, and dimethoate were more toxic to *M. domestica*, dichlorvos and Pyrenone were more toxic to *S. cameroni*, and crotoxyphos was equally toxic to both species. The significance of these findings to successful control of fly pests on dairies is discussed.

Key Words: Insecta, parasitoid, pyrethroid, organophosphate, resistance.

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The house fly, *Musca domestica* L. and stable fly, *Stomoxys calcitrans* (L.), are major pests in and around dairy farms in the USA. Lowered milk production, reduced feed conversion efficiency, exposure to disease-causing agents, blood loss, and public health and nuisance concerns all result from house fly and stable fly activity. These pests cause direct annual losses (losses plus control costs) estimated at over $120 million nationwide (Anon. 1979). Present control techniques rely principally on pesticide use, which has many potentially serious drawbacks including destruction of nontarget biological control agents, development of insecticide resistance in the target fly population, harmful levels of insecticide residues, and environmental pollution. For these reasons, considerable interest has been generated in development of integrated pest management (IPM) programs for flies on dairy farms, with emphasis on biological control using hymenopterous parasitoids. For IPM to be successful, the pesticides used should be highly selective (i.e., toxic to the pest and non-toxic to the beneficial parasitoids). However, little is currently known about the toxicity of insecticides commonly used for fly control on dairy farms to the beneficial biological control agents.

Indigenous hymenopterous pupal parasitoids (Family Pteromalidae) are important biological control agents of house flies and stable flies on dairy farms (Peterson and Meyer 1983; Smith and Rutz 1985). A previous study comparing the selectivity of seven insecticides between *M. domestica* and *Urolepis rufipes* (Ashmead) revealed

[1] HYMENOPTERA: Pteromalidae, Accepted for publicated 20 May 1988.

that fenvalerate and crotoxyphos were more toxic to the fly than the parasitoid; Pyrenone and dichlorvos were more toxic to the parasitoid, while permethrin, dimethoate and tetrachlorvinphos showed similar toxicity to both (Scott and Rutz 1988). However *U. rufipes* is only one of several important muscoid fly biological control agents. Because similar data are needed on other parasitoids we determined the toxicity of seven commonly used insecticides against *Spalangia cameroni* Perkins, an important parasitoid of house flies and stable flies.

MATERIALS AND METHODS

Spalangia cameroni was collected from dairies in and around Tompkins, Schuyler, and Cayuga counties of New York in 1985. Parasitoids were collected by using the sentinel pupae parasitoid monitoring method (Rutz and Axtell 1979) and have been colonized in the lab since being collected.

Insecticides tested were: crotoxyphos (Ciodrin) [dimethyl (*E*)-1-methyl-2-(1-phenylethoxycarbonyl)vinyl phosphate], dichlorvos (Vapona) [2,2-dichloro-vinyl dimethyl phosphate], dimethoate (Cygon) [*O,O*-dimethyl *S*-methylcarbomoyl-methyl phophorodithioate], fenvalerate [(*RS*)-∝-cyano-3-phenoxybenzyl (*RS*-2-(4-chlorophenyl)-3-methylbutyrate], permethrin [3—phenoxybenzyl(1RS)-*cis-trans*-3-)2,2dichlorovinyl)-2,2-dimethylcyclopropanecarboxylate, Pyrenone [pyrethrins + piperonyl butoxide], and tetrachlorvinphos (Rabon) [(*Z*)-2-chloro-1-(2,4,5-trichlorophenyl) vinyl dimethyl phosphate]. Sources of the insecticides were given previously (Scott and Rutz 1988). The synergists piperonyl butoxide (PBO > 95%), diethyl maleate (DEM, Aldrich Chem. Co., 97%) and *S,S,S*-triburyl phosphorotrithioate (DEF) were also used.

One- to six-day-old parasitoids were tested by a surface contact assay (Scott and Rutz 1988). This method involved covering the inside surface of a 37 cm^2 glass jar with insecticide (or insecticide plus synergist), aspirating the parasitoids, and transferring 20 parasitoids into the jar. The dosage of the synergists was 100 µg/jar for DEM and 10 µg/jar for PBO and DEF. These doses gave no control mortality. Each jar was covered with a wire mesh top and the parasitoids were given a small drop of 10% sucrose on the lid as a food source. Males and females were tested separately. Susceptibility was assessed after holding the parasitoids at constant temperature (28°C) for 48 hours (LD 15:9). Twenty parasitoids were tested at each dose using at least four doses giving between 0% and 100% mortality. Each test was replicated a minimum of five times. Results were analyzed by the method of Finney (1952). Weights of *S. cameroni* were determined using freshly killed specimens. Batches of 20 male or female parasitoids (n = 30) were weighed.

RESULTS

Toxicity of the seven insecticides to *S. cameroni* decreased in the order of dimethoate > crotoxyphos > permethrin ≃ dichlorvos ≃ Pyrenone fenvalerate. Overall, there was a 126-fold reduction in toxicity from dimethoate to fenvalerate based on female LC_{50} values. There was substantial variation in the toxicity of compounds within classes of insecticides; however, the pyrethroids were generally less toxic than the organophosphates to *S. cameroni*. Male *S. cameroni* were significantly more sensitive than females to fenvalerate, permethrin, and crotoxyphos ($P \leq 0.05$). For all insecticides except tetrachlorvinphos, the data points fit the log dose-probit line ($P \leq 0.05$, chi-squared test; $P \leq 0.10$ for dichlorvos) and there was no

indication of any plateaus. A plot of log dose tetrachlorvinphos vs. mortality (probit scale) shows the existence of a plateau that extends over a 16-fold dose range (Fig. 1) for both sexes. This indicated the existence of two distinct populations relative to their susceptibility to tetrachlorvinphos. The most likely explanation for this phenomenon was that 45% of the population was resistant to tetrachlorvinphos due to selection in the field. The similar response of males and females suggested that the resistance was not affected by the chromosomal ploidy in these parasitoids.

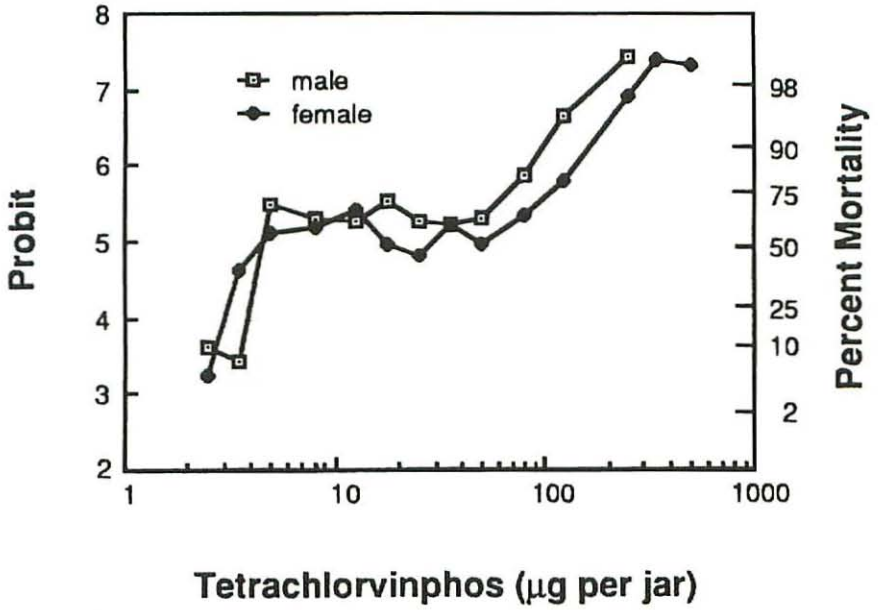

Fig. 1. Toxicity of tetrachlorvinphos to male and female *S. cameroni*.

In an effort to determine the mechanism(s) responsible for the resistance to tetrachlorvinphos, we examined the effect of three synergists: piperonyl butoxide (PBO), DEF and DEM, inhibitors of oxidative, hydrolytic and glutathion S-transferase mediated metabolism, respectively. In these tests the effect of the synergist was evaluated against both the susceptible (i.e., the approximately 55% of the individuals that die at the lower doses) and resistant (i.e., the approximately 45% of the individuals that die only at the higher doses) populations. DEM did not affect either population (Fig. 2). DEF showed a noticeable but erratic effect. DEF had no effect on the susceptible population, and its effect on the resistant population was inconclusive. PBO had a significant effect on both populations. There was an 8-fold increase in toxicity to the susceptible population, suggesting that mixed-function oxidases (MFOs) are important in the detoxification of tetrachlorvinphos in this species. There was also a significant effect on the resistant population(i.e., greater than seen for the susceptible strain), implying that the resistance may be due in part to elevated MFO activity.

To investigate the basis of the differential susceptibility of males compared to females, we determined the average weight of each sex. Males were found to be

consistently smaller ($P \leq 0.05$) than females with average weights of 0.64 ± 0.02 mg ($\bar{X} \pm$ SEM) and 0.74 ± 0.04, respectively. The generally greater tolerance to the insecticides exhibited by female *S. cameroni* was probably due to their larger size. The most notable exception was dichlorvos, to which females were more sensitive than males.

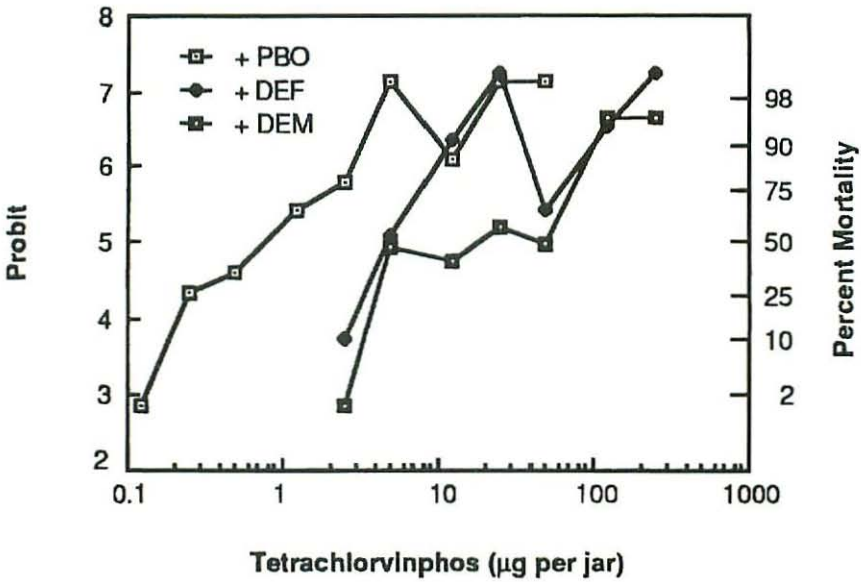

Fig. 2. Toxicity of tetrachlorvinphos to female *S. cameroni* in the presence of the synergists piperonyl butoxide (PBO), DEF or diethyl maleate (DEM).

Comparison of the parasitoid LC_{50}-fly LC_{50} ratio (Table 1) indicated that fenvalerate was much less toxic (41-fold) to the parasitoid than to *M. domestica* followed by permethrin (17-fold) and dimethoate (2-fold). Crotoxyphos and Pyrenone showed little selectivity while dichlorvos was more toxic to the parasitoids, having a parasitoid-fly ratio of 0.14. It must be noted that the susceptibility strain of *M. domestica* used for these comparisons may not necessarily be representative of *M. domestica* populations on dairies that are often resistant to many pesticides (Meyer et al. 1987).

DISCUSSION

Our results indicate that fenvalerate, permethrin and dimethoate were more toxic to *M. domestica* than to *S. cameroni*. It is likely that these insecticides would, therefore, offer a greater degree of control of fly populations due to their reduced toxicity to *S. cameroni*. It should be realized that selective pesticides are only one of the alternatives for developing a successful IPM strategy. Selectivity may also be obtained by proper timing, application techniques, and other factors such as resistance. For example, if resistance to a given insecticide developed in the parasitoids, and not in the target flies, use of that insecticide would greatly improve the eficiency of an IPM strategy. It appears resistance to tetrachlorvinphos has already developed to some extent in this strain of *S. cameroni*. It is noteworthy

that the resistance has been maintained in this colony for over one year without any selection pressure, suggesting that the resistance mechanism(s) does not have a strong reproductive disadvantage. Our results suggest that a high level of resistance could be selected for in *S. cameroni*, and the resistant parasitoids released to supplement control of muscoid flies in an IPM system for daily farms. A similar system has been proposed for a predatory mite in apple orchards (Roush and Hoy 1981).

Comparison of the toxicity between *S. cameroni* and *M. domestica* after correcting for differences in weight (LC_{50}/mg body weight; $\bar{X} \pm$ SEM weight of female house flies was 11.9 mg $\pm$ 0.4, Scott and Rutz 1988) revealed that *S. cameroni* was more tolerant to all insecticides tested (range: 2.3-670-fold, especially permethrin (280-fold) and fenvalerate (670-fold). This implies that *M. domestica* is probably a better (i.e., more sensitive) animal for toxicity studies on pyrethroids or organophosphates. However, correcting toxicity data for differences in body weights may have little relevance to field situations in this case. Comparison of toxicity data between *S. cameroni* and *U. rufipes* (Scott and Rutz 1988) shows that dimethoate, permethrin, Pyrenone and fenvalerate were more toxic to *U. rufipes*. LC_{50} values for dichlorvos and crotoxyphos were similar between both species.

We now have data on two of the most common hymenopterous pupal parasitoids of flies on dairies. Fenvalerate is the only compound showing the desired characteristic of being highly toxic to *M. domestica* and much less toxic to *S. cameroni* and *U. rufipes*. However, more information is needed on the toxicity of these commonly used insecticides to other parasitoid species before adequate data will be available for whole-scale incorporation into an IPM program.

Table 1. Toxicity of seven insecticides to male and female *Spalangia cameroni* and female *Musca domestica*.

		S. cameroni					*M. domestica*‡	
			95% Confidence Intervals					
Compound	Sex	LC_{50}*	Lower Limit	Upper Limit	Slope (S.E.)	n	LC_{50}	Ratio
Tetrachlorvinphos	male	†	—	—	—	1580		
	female	†	—	—	—	2400	0.90	—
Dichlorvos	male	3.1	2.9	3.4	2.8 (0.2)	900		
	female	1.5	1.4	1.6	2.8 (0.2)	1380	10.4	0.14
Crotoxyphos	male	1.1	1.0	1.2	5.6 (0.1)	680		
	female	1.8	1.7	1.9	8.3 (0.6)	560	1.7	0.94
Dimethoate	male	0.12	0.11	0.13	5.0 (0.5)	500		
	female	0.14	0.13	0.15	6.5 (0.5)	600	0.07	2.0
Permethrin	male	1.5	1.4	1.7	2.5 (0.2)	1040		
	female	5.5	5.2	5.8	5.2 (0.4)	680	0.32	17
Pyrenone	male	5.0	4.7	5.4	4.2 (0.3)	700		
	female	5.0	4.7	5.3	5.4 (0.6)	440	5.8	0.86
Fenvalerate	male	11.3	10.2	12.4	2.8 (0.2)	700		
	female	17.6	16.4	18.9	3.5 (0.3)	820	0.42	42

* In units of μg/jar.
† Data did not fit a straight line.
‡ From Scott and Rutz (1988).

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FIELD EVALUATION OF SYNERGIZED PERMETHRIN FOR CONTROL OF PERMETHRIN-RESISTANT HOUSE FLIES[1] ON SOUTHERN CALIFORNIA DAIRIES

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Abstract: Field applications of permethrin wettable powder (WP) and permethrin WP plus piperonyl butoxide (PB) (1:5) were evaluated for control of permethrin-resistant house flies on dairies at two different areas in Southern California. Multiple applications of permethrin WP or permethrin WP plus PB between June and October 1986 did not influence significantly the levels of resistance to permethrin, as determined by laboratory bioassays on house flies collected just before (June) and soon after (November) the treatment period. The addition of PB to permethrin for the June and November bioassays of field populations strongly increased permethrin toxicity, indicating that a major portion of the resistance was due to mixed-function oxidases. However, the mixture of permethrin plus PB did not provide significant improvement in control over that achieved by permethrin alone. The limitations of current PB formulations for use with insecticides with long residual stability is discussed.

Key Words: *Musca domestica*, permethrin, dairies, piperonyl butoxide, resistance.

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Dairy production is California's largest single agricultural industry, valued over $2 billion in 1986 (California Department of Food and Agriculture 1987). Nearly 25% (232,500) of the state's dairy cows are located in Chino Basin, an area of ca. 80 km^2 in Southern California. The general area surrounding the Chino Basin supports one of the fastest-growing human populations in the United States (U.S. Census Bureau 1985).

All dairies in the Chino Basin practice confinement-type management, which leads to tremendous manure accumulations and ideal conditions for development of house fly, *Musca domestica* L., infestations. Control of adult house flies on many of the dairies is performed by licensed pest control operators (PCO's) and usually consists of weekly or biweekly applications of permethrin to animal shelters, storage buildings, and other fly resting sites. In general, this control program has been in effect since the late 1970's when permethrin was registered for fly control in California.

After several years of commercial use, PCO's in the Chino Basin began reporting failures of permethrin to achieve adequate control of house flies. Meyer et al. (1987) documented high levels of permethrin resistance in several house fly populations sampled from dairies in the area. During that study, it was also found through laboratory bioassays that the addition of piperonyl butoxide (PB) to the permethrin (5:1) increased the toxicity of the latter by as much as 34-fold. The practical implication was that the increased toxicity of synergized permethrin might re-establish control, thus prolonging the effective use of permethrin against resistant flies (MacDonald et al. 1985).

Formulating synergists with registered insecticides poses some logistical concerns (Raffa and Priester 1985) such as increased control cost, government registration,

[1] DIPTERA: Muscidae. Accepted for publication 26 May 1988.

and formulation problems. However, successes of permethrin and PB mixtures for controlling resistant pests have been reported (Ghidiu and Silcox 1984; MacDonald et al. 1983). The following study was therefore designed to evaluate permethrin and synergized permethrin for control of permethrin-resistant house fly populations in Southern California.

MATERIALS AND METHODS

Six dairies were selected for this study: three were located in the Chino Basin and had been documented as having relatively high resistance levels to permethrin (Meyer et al. 1987). The other three were near the community of Sun City, ca. 60 km from the Chino Basin. Two of the dairies in each area received periodic applications of either permethrin wettable powder (WP) or synergized permethrin WP, the third dairy served as a control. House fly control on each unsprayed dairy was limited to applications of granular fly baits formulated with sugar and 1.0% methomyl. All permethrin and permethrin/PB treatments were applied by a PCO at the labeled rate of 0.125% (AI). The relative size of each dairy and the frequency of each treatment are shown in Table 1.

Table 1. Relative herd size and treatment interval for cooperating dairies.

Dairy	Treatment	Treatment interval	Herd size
Chino Basin			
J1	Permethrin	21 (6)*	720
PC	Permethrin + PB	7 (22)	1,400
CD	Control	—	650
Sun City			
MP	Permethrin	21 (6)	350
AB	Permethrin + PB	21 (7)	1,150
DJ	Control	—	980

* Number of applications of each treatment from June through October 1986.

Treatments were initiated on each facility during mid-June 1986 and continued periodically until late October. Adult house fly samples were collected from each dairy before (11 June 1986) and after (3 November 1986) the treatments had been applied. Each sample, consisting of several hundred flies, was colonized in the laboratory by rearing the larvae on standard CSMA (Chemical Specialties Manufacturers Association) media at 27°C and 50 to 60% RH. Flies were fed sucrose-water from a saturated dental wick placed in the holding container.

Resistance levels to permethrin alone or in combination with PB (permethrin: PB, 1:5) were determined by topical application of a 1-µl-drop of the insecticide solution in acetone to F_1 adult females (ca. 3 d old) under CO_2 anesthesia according to standard methods (Georghiou and Bowen 1966). All tests were replicated five times on different days, with each test including at least five different doses of permethrin or synergized permethrin; 20 flies/replication were used. Bioassays were also performed on a susceptible NAIDM (National Assoc. of Industrial Disinfectants Manufacturers) strain of house flies. Treated flies were held in ventilated containers at room temperature (24°C) and examined for mortality after 24 h. Each container of flies received a dental wick saturated with a 20% sucrose solution. The data were corrected for control mortality by Abbott's formula (1925) and were subjected to probit analysis on a programmed computer according to the method of Finney 1971; resistance ratios (RR) (LD_{95}

field strain/LD_{95} susceptible strain) and synergism ratios (SR) (LD_{95} unsynergized/LD_{95} synergized) were calculated. All LD_{95} values are expressed as µg/female fly.

Fly population densities on each dairy were measured on a weekly basis with a baited jug trap (Burg and Axtell 1984). Three traps, baited with True Grit Blue fly bait, were placed on each dairy near areas where flies congregated (calf pens, manure mounds, commodity pits). Fresh bait was added to each trap every 14 days to maintain its attractiveness. A three-point running mean was utilized to reduce the variation in weekly trap catch data.

RESULTS

Results of tests on permethrin and synergized permethrin are shown in Tables 2 and 3, respectively. Resistance ratios for the collections made in June (pretreatment) were very similar, ranging from 17.3- to 30.6-fold (Table 2). Results from November (posttreatment) bioassays were also similar, but resistance levels showed a surprising decline at five of six dairies despite multiple (6-22) applications of permethrin (Tables 1 and 2); none of the changes were statistically significant ($\alpha = 0.05$). The slopes of the probit regression lines from the June and November bioassays were also quite similar, suggesting little change in the extent of heterogeneity in the response of the population toward permethrin. The inclusion of PB to the bioassay mixture considerably increased permethrin toxicity to each population, suggesting that an important portion of the resistance was due to mixed-function oxidases (Table 3) (Meyer et al. 1987; MacDonald et al. 1985). The SR values ranged from 67.1 (PC, synergized permethrin) to 243.6 (DJ, control) in the June assay and from 75.8 (CD, control) to 218.1 (MP, permethrin) in the November assay. The SR decreased in house fly populations sampled at control dairies between June and November, while increasing at all of the treated dairies except J1 (Table 3). The decrease in the SR at J1 was relatively small, with values of 112.2 and 95.5 for the June and November bioassays, respectively (Table 3).

Table 2. Resistance levels of six field strains of house flies, bioassayed with permethrin before (June) and after (November) various treatments.

Dairy	Treatment	Sample date	LD_{95}*	95% Fiducial limits Lower	Upper	Slope (SEM)	RR†
NAIDM‡	—	June	0.066	0.053	0.080	3.26 (0.32)	—
		Nov.	0.072	0.062	0.082	3.85 (0.25)	—
DJ	Control	June	1.218	0.624	4.894	1.91 (0.27)	18.5
		Nov.	0.869	0.650	1.162	1.86 (0.12)	12.1
MP	Permethrin	June	1.475	0.779	6.202	1.83 (0.28)	22.3
		Nov.	1.527	0.861	4.386	1.76 (0.23)	21.2
AB	Permethrin	June	1.390	0.916	2.109	1.80 (0.18)	21.1
	+ PB	Nov.	0.906	0.479	3.389	1.97 (0.29)	12.6
CD	Control	June	1.471	0.939	2.303	1.73 (0.18)	22.3
		Nov.	1.138	0.807	1.603	1.79 (0.13)	15.8
J1	Permethrin	June	2.021	0.862	3.545	2.36 (0.51)	30.6
		Nov.	1.050	0.824	1.338	1.92 (0.11)	14.6
PC	Permethrin	June	1.143	0.619	4.145	2.14 (0.30)	17.3
	+ PB	Nov.	1.531	0.840	4.747	1.89 (0.23)	21.3

* µg/female fly.
† RR, resistance ratio, LD_{95} field strain/LD_{95} susceptible strain.
‡ Susceptible laboratory strain of house flies.

House fly population densities at each dairy in the Chino Basin and Sun City are shown in Fig. 1 A and B, respectively. Results from Chino Basin showed the

CD (control) dairy generally to have greater numbers of flies per trap than either J1 (permethrin) or PC (synergized permethrin) (Fig. 1A). The addition of PB to the permethrin had no obvious impact on fly densities on the PC dairy; mean weekly trap counts were actually lower on the J1 dairy over the duration of the study.

Table 3. Resistance levels of six field strains of house flies, bioassayed with permethrin plus PB before (June) and after (November) various treatments.

Dairy	Treatment	Sample date	n	LD_{95}*	95% FL Lower	95% FL Upper	Slope (SEM)	RR†	SR‡
NAIDM§	—	June	340	0.002	0.002	0.003	6.66 (0.65)	—	33.0
		Nov.	600	0.006	0.005	0.007	3.46 (0.29)	—	12.0
DJ	Control	June	520	0.005	0.004	0.006	3.95 (0.32)	2.5	243.6
		Nov.	800	0.005	0.004	0.006	3.63 (0.21)	0.8	173.8
MP	Permethrin	June	400	0.012	0.006	0.792	3.41 (0.61)	6.0	122.9
		Nov.	1060	0.007	0.005	0.011	2.56 (0.24)	1.2	218.1
AB	Permethrin	June	480	0.017	0.013	0.023	2.42 (0.19)	8.5	81.7
	+ PB	Nov.	800	0.006	0.005	0.007	2.49 (0.17)	1.0	151.0
CD	Control	June	500	0.009	0.008	0.012	3.21 (0.24)	4.5	163.3
		Nov.	420	0.015	0.011	0.019	2.82 (0.25)	2.5	75.8
J1	Permethrin	June	460	0.018	0.014	0.024	2.75 (0.25)	9.0	112.2
		Nov.	680	0.011	0.008	0.020	2.98 (0.35)	1.8	95.5
PC	Permethrin	June	400	0.017	0.014	0.021	3.48 (0.31)	6.5	67.1
	+ PB	Nov.	700	0.013	0.009	0.020	3.31 (0.39)	2.2	117.7

* µg/female fly.

† RR, resistance ratio, LD_{95} field strain/LD_{95} susceptible strain.

‡ SR, synergism ratio, LD_{95} unsynergized/LD_{95} synergized (see Table 2 for unsynergized LD_{95} values).

§ Susceptible laboratory strain of house flies.

Results from Sun City dairies were different from those in Chino Basin (Fig. 1B). The MP (permethrin) dairy had relatively low numbers of flies, compared with the AB (synergized permethrin) and DJ (control) dairies. The addition of PB had no clear effect on the fly population at the AB dairy.

DISCUSSION

Laboratory bioassays with permethrin alone (Table 2) showed no drastic increase in the resistance level. In fact, the resistance level dropped at five of the test sites. These decreases may be due to variability in some undetermined aspect of the experimental procedure, since assays were conducted 6 mo apart. The differences could also have been induced by the influx of susceptible flies into the treated populations prior to sampling. Mullens and Meyer (1987) showed house fly population densities to be greatest on Southern California dairies between July and September, declining to low levels by November. The heavy concentration of dairies in each community could have provided an influx of susceptible flies between July and September, with the resulting November population being more susceptible to permethrin.

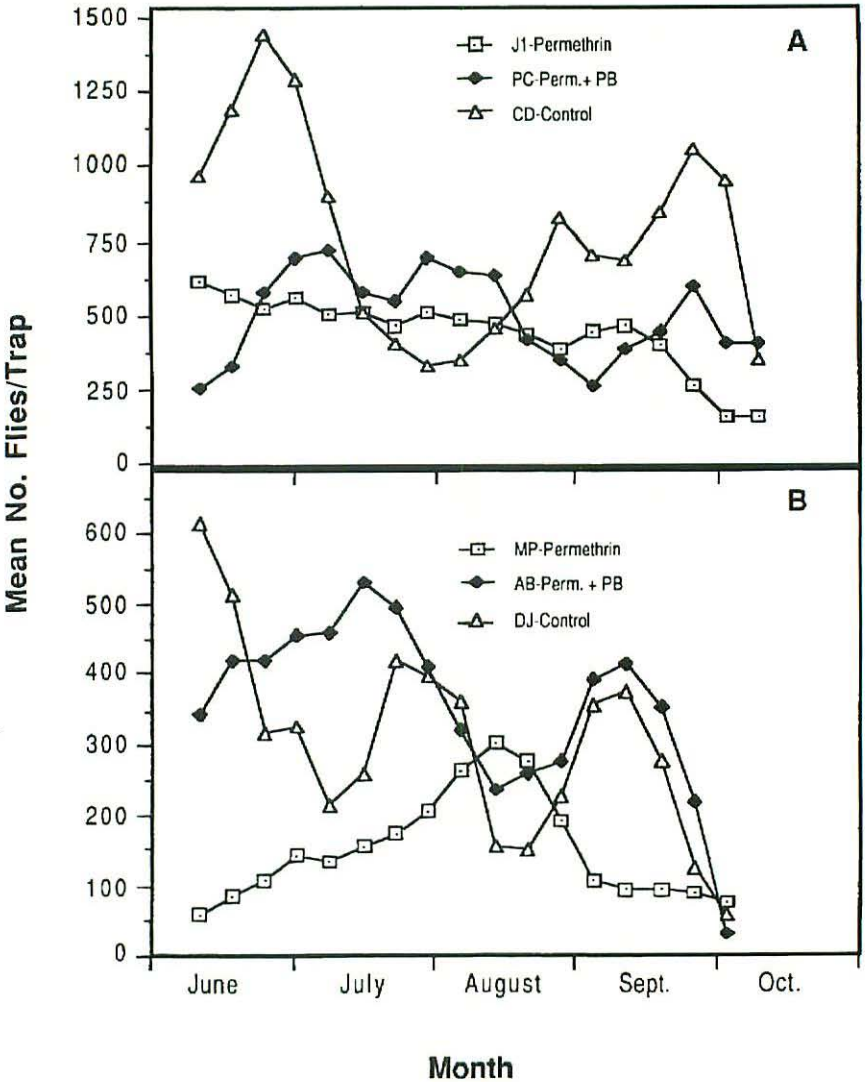

Fig. 1. A. Mean number of adult house flies sampled each week with baited jug traps (three traps per dairy) on three dairies in the Chino Basin of Southern California.
B. Mean number of adult house flies sampled each week with baited jug traps (three traps per dairy) on three dairies near Sun City, California.

The increased SR values between June and November at the treated dairies (except J1), and decreased values at the control dairies, indicates that the periodic applications of permethrin and synergized permethrin were selecting for higher levels of mixed-function oxidases in the populations. A comparison of SR values determined for the PC and J1 dairies in November 1984 (Meyer et al. 1987) and November 1986 (Table 3) shows a 3- to 5-fold increase, again reaffirming the effect

of permethrin applications on selection for higher levels of mixed-function oxidases. In spite of the increased levels of mixed-function oxidases detected in the house fly populations from the dairies bioassayed in November, the RR values decreased, indicating that sufficient levels of PB were present in the bioassay mixture to adequately synergize the permethrin. Although the same level of PB was utilized in the field applications as in the laboratory bioassays, the effect of the PB was not detectable through measurements of adult house fly population density (Fig. 1). The level of house fly control at the AB dairy was generally poorer than that obtained at the MP dairy, where only permethrin was utilized (Fig. 1B). The PC (synergized permethrin) and J1 (permethrin) dairies had comparable levels of fly control (Fig. 1A).

Synergizing permethrin WP with PB provided no enhanced control of permethrin-resistant house flies. The high levels of synergism demonstrated in the laboratory bioassays of the field strain of house fly (Table 3) indicate that some field-related factors were modifying the effectiveness of PB. These factors could include the poor photostability of PB or its incompatibility with a long-lived residual treatment. Some enhanced mortality may have occurred immediately after application and not have been represented in the weekly assessment of the fly population density.

The current resistance problem is a direct result of a more severe problem — that being the lack of effective integrated fly control strategies. The size, intensity, and concentration of dairies in Southern California almost precludes the reliance on manure management for fly control. Biological control of house flies on dairies in this area has not been evaluted, and it is unlikely that releases of parasitic wasps could be accomplished at rates sufficient to impact fly populations. Also, integrated pest management programs would have to be implemented on an areawide basis, requiring the cooperation of several hundred individual dairies.

ACKNOWLEDGMENT

We thank Marilyn Hawley, Hieu Tran, and Tracy Cyr for their assistance on the project. This research was supported by a grant from the Western Region Pesticide Impact Assessment Program.

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Journal of Agricultural Entomology

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ERRATUM

The following reference was inadvertently omitted from the index on the front cover of the Journal of Agricultural Entomology, Volume 5, Number 3, July 1988:

COLEMAN, RUSSELL E, and REID R. GERHARDT — Frequency of regurgitation by laboratory-reared face flies fed trypticase-soy broth 185

The editor regrets any inconvenience caused by this omission.

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MANAGEMENT OF FIRST GENERATION EUROPEAN CORN BORER, *OSTRINIA NUBILALIS*,[1] IN EARLY SEASON, FRESH MARKET SWEET CORN IN CONNECTICUT

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Abstract: Acceptable yields for fresh market (96.4% of ears free of damage) were obtained with early (May) planted hybrid sweet corn that was subjected to attack by first generation European corn borer (ECB), *Ostrinia nubilalis* (Hubner), without insecticide applications. This occurred even though young corn plants were heavily infested during the whorl and tassel stages and initially exhibited substantial amounts of feeding damage (up to 36% of the plants) that exceeded current recommended economic thresholds. Early instar larvae that infest young plants appear to die from natural causes and do not survive to invade the late developing ears, thus suggesting that treatments could be eliminated with no adverse effect on yields. Unacceptable yields with 37.0% and 19.0% of the ears damaged, were obtained with mid-(June) and late-(July) season plantings respectively, indicating that some form of insecticide treatment is needed to control feeding damage caused by second generation ECB populations.

Key Words: European corn borer, *Ostrinia nubilalis*, first generation management, sweet corn, Connecticut.

J. Agric. Entomol 5(3): 153-159 (July 1988)

The European corn borer (ECB), *Ostrinia nubilalis* (Hubner) is a major economic pest of sweet corn, *Zea mays* L. in the northeastern United States. In Connecticut, the ECB has two generations a year. Individuals overwinter as fifth instars in old corn stalks and wild host plants that remain from the previous season. Larvae pupate in mid-May and adults emerge in June. Females of the first generation typically oviposit on early plantings of mid-whorl stage corn. Larval borers actively feed in the leaves, tassels, and stalks and pupate in late-July. Adults of the second generation emerge shortly thereafter, and oviposit from early August through mid-September on mid- and late-season cultivars that are often planted successionally. Larvae of this brood attack all parts of the corn plant and characteristically bore into the ears making them unmarketable. Larvae enter diapause within the plant in November and emerge the following spring to continue the cycle.

Because the ECB has such a low economic injury level on fresh market sweet corn, current management procedures for larval control rely heavily on the use of conventional chemical insecticides. Treatments are usually initiated when ca. 15% of the corn plants in a field show active leaf feeding damage and applications may be made every four to five days until harvest (Bouton & Nicklow 1986). This spray schedule will usually involve two to four applications for early- and mid-season cultivars that mainly support first generation ECB and as many as twelve applications for late-season plantings that are attacked by the second generation.

[1] LEPIDOPTERA: Pyralidae. Accepted for publication 31 May 1988.

Recent studies conducted for five successive years (1981-1985) (Andreadis 1984, 1986, 1987), have repeatedly shown a high degree of natural mortality among first generation ECB larvae in Connecticut. This mortality, which has in part been attributed to infection by the microsporidian parasite, *Nosema pyrausta* (Paillot), occurs during larval development and can reduce initial ECB densities by as much as 80% by the time of pupation and adult emergence, which usually coincide with harvest. As a result, many larvae than inhabit immature plants and initially cause damage to the leaves and stalks, appear to die from natural causes before they can migrate to and feed in the later developing ears.

These observations suggest that it may be possible to obtain acceptable yields with early planted sweet corn that is subjected to attack by first generation ECB without insecticide applications. The objective of this study was to assess this possibility by monitoring ECB feeding damage in untreated fields of successionally-planted sweet corn and obtaining quantitative fresh market yield data.

MATERIALS AND METHODS

This study was conducted from May through October, 1986 in fields of commercial hybrid sweet corn, *Zea mays* L. located at the Connecticut Agricultural Experiment Station's research farm in Hamden, Connecticut. Three planting dates were selected: 12 May, 11 June, and 10 July. These dates were based on prior observations of natural ECB oviposition (Andreadis 1984, 1986, 1987) so that the first planting would support first generation ECB populations, the second planting would support both first and second generations, and the third planting would support second generation ECB only. Plots were arranged in a complete randomized block design with six replicates for each planting date. Individual plots were eight rows wide by 90 meters long.

Plants were inspected weekly for ECB activity and studies began with the first signs of larval feeding. Each week, 50 plants were randomly selected from every plot and assayed for fresh feeding damage. The number of plants showing damage and the location of the damage on the plant (leaf, whorl, tassel, stalk, ear) were recorded. Live EBC larvae were also extracted on the same sample dates from several damaged plants in each plot and their developmental stage(s) was recorded. Previous studies (Andreadis 1984, 1986, 1987) had shown these procedures to be reliable for assessing ECB phenology and population trends in each generation. Growth stages of the corn plant were monitored weekly and classified by the criteria of Beard & Turner (1942). These data were collected for each planting until harvest.

RESULTS

First generation ECB feeding activity was initially detected in early-May planted plots on 26 June (Table 1). Corn plants were in the early- to mid-whorl stage (tassel completely enclosed by whorl) and they measured 40-45 cm in height. Feeding damage was restricted to the leaves and an average of 21.0% (n = 300) of the plants in the six replicated plots, exhibited some form of leaf damage. This exceeded the economic threshold of 15%. Similar levels of feeding damage were detected the following week (ave. = 26.0% of the plants), but as plants began to tassel (11 July), a significant increase in ECB feeding activity was observed (ave.

= 36.0% of the plants). Thereafter, as plants and borers developed to maturity, a decreasing amount of fresh feeding damage was recorded and this trend continued until ECB pupation, which coincided with harvest (12 August).

First generation ECB feeding damage was not detected in the second mid-June plantings until 11 July (Table 1). All plants were in the early- to mid-whorl stage and were only 22-35 cm in height. Initial feeding damage did exceed the economic threshold but was less than half of the amount recorded in the first early-May plantings that were sampled on the same date (17% vs 36% of the sampled plants). Minimal feeding activity was observed in subsequent weeks and substantial feeding damage was not observed until mid- to late-August, when plants were subjected to attack from the second ECB generation, as indicated by the presence of first instar larvae. This activity, however, was coincident with the onset of silking and ear formation and an average of more than 50% of the plants were damaged by the time of harvest (3 September).

ECB feeding activity was detected in the third late-June planting (10 July) on 30 July, just three weeks after planting (Table 1). Most corn plants were in the early-whorl stage and they ranged from 19-26 cm in height. Feeding damage was initially confined to an average of only 10.0% of the plants, but this increased steadily with each successive week. This increase occurred in all replicated plots until harvest and was attributed to attack by individuals of the second ECB generation, as indicated by the presence of early instars in the newly-infested corn plants. As in the second mid-June planting, more than 50% of the plants showed active feeding damage by harvest on 9 October.

Although moderate stalk damage (ave. of 32.3% of the plants) was recorded for the early-May plantings at harvest (Table 2), ear damage was minimal; 98.9% to 92.7% of the ears examined from the six replicated plots were free of damage and thus considered marketable. ECB densities in the stalks and ears were also quite low (ave. of 6.7 and 1.1 / 50 plants, respectively) and the number of damaged plants that harbored live ECBs averaged only 34.2%. These findings were consistent with the weekly declines in fresh feeding damage observed during late July and early August (Table 1), suggesting a high degree of mortality among first generation ECB larvae.

Substantial feeding damage was sustained in ears harvested from the second mid-June planting on 3 September; an average of 37.0% of ears had kernel damage (range = 27.5% to 48.6%) (Table 2). ECB densities and feeding damage in the stalks were also quite extensive and in contrast to the early-May plantings, almost all (ave. = 92.9%) corn plants exhibiting some form of feeding damage, harbored live second generation borers.

Stalk and ear damage were also quite extensive in the late-July plantings that were not harvested until 9 October (ave. = 59.7% of plants and 19.0% of ears) (Table 2). However, while ECB density and damage in the stalks were comparable to the second mid-June planting, ECB densities and damage in the ears were significantly lower. The number of damaged plants that were inhabited by live ECB larvae at harvest was also slightly but not significantly lower (82.1% vs 92.9%).

DISCUSSION

This study demonstrates that acceptable yields can be obtained in early planted sweet corn that is subjected to attack by first generation ECB without the

use of chemical insecticides. This occurs even though young corn plants are heavily infested by ECB larvae during the whorl and tassel stages and initially exhibit substantial amounts of feeding damage in excess of accepted economic thresholds. It appears that young larvae which inhabit the immature plants, do not survive to invade and damage the late developing ears. The weekly declines in fresh feeding damage, low numbers of ECB larvae inhabiting plants at harvest, and small proportion of damaged plants with live borers all indicate a high degree of juvenile mortality. The phenological data of ECB development further demonstrates that the few larvae that do cause ear damage originate from eggs laid during the whorl stage and that the small amounts of ear damage obtained with the early planting cannot be attributed to larval pupation prior to ear formation. This corroberates previous studies (Andreadis 1984, 1986, 1987) that have repeatedly shown large declines in first generation ECB populations as a result of natural causes.

Under current control recommendations for fresh market sweet corn (Bouton & Nicklow 1986), insecticide applications would have been initiated in all plots planted in early May on the 26 June, when an average of 21% of the plants exhibited fresh feeding damage. Two to four treatments would then have been made, depending on the degree of additional feeding damage. My results show that these treatments could be eliminated without adversely affecting yields, since 3-5% ear damage is generally considered acceptable even with chemical treatments. Simlar conclusions with successionally planted sweet corn have been reached in a recent study done in western Massachusetts, where Ferro and Fletcher-Howell (1985) reported that no more than one or two treatments were needed to effectively manage first generation ECB larvae.

These same conclusions do not apply to mid- and late-season plantings. Mid-season sweet corn planted in June was subjected to attack by larvae of both generations and sustained much greater ear damage. This damage occurred in all plots and appeared to be due to the simultaneous occurrence of silking stage corn and the emergence of second generation ECB populations. These findings indicate that treatment would be necesary in order to achieve satisfactory yields. However, results obtained in the weekly assays for fresh feeding damage also suggest that treatments would not be necessary against the first generation in July and that applications probably could be withheld until mid-August, when second generation larvae initially began to feed.

Heavy feeding damage by second generation ECB populations was also sustained in late-season (July) plantings that were harvested in October. Although ear damage was somewhat less than that for the mid-June plantings, yields were equally unacceptable and repetitive applications would undoubtedly be required in these plantings as well. In this instance, however, treatments probably would have to begin as early as the first week in August when most plants were in the early-whorl stage and be continued weekly until harvest.

Table 1. Weekly ECB feeding damage in early-(May), mid-(June), and late-(July) season plantings of hybrid sweet corn at Hamden, Connecticut 1986.

	May 12			June 11			July 10		
Sample date	Corn plant stage*	ECB stage†	Mean ± SE % plants damaged‡	Corn plant stage*	ECB stage†	Mean ± SE % plants damaged‡	Corn plant stage*	ECB stage†	Mean ± SE % plants damaged‡
June 26	W	L_1	21.0 ± 2.9ab						
July 3	W	L_2	26.0 ± 4.0a						
July 11	T	L_2,L_3	36.0 ± 3.6d	W	L_1,L_2	17.0 ± 4.9a			
July 17	T	L_3	28.0 ± 4.3a	W	L_2,L_3	8.7 ± 1.6b			
July 23	S	L_3,L_4	12.4 ± 3.4c	W	L_3,L_4	4.3 ± 2.4c			
July 30	S	L_4,L_5	10.0 ± 2.3c	W	L_4,L_5	6.3 ± 1.2bc	W	L_1	10.0 ± 2.0a
Aug. 7	S	L_5P	14.0 ± 1.0bc	T	L_2P	4.0 ± 0.9c	W	L_1	16.0 ± 4.3b
Aug. 14				S	PL_1	8.7 ± 1.6b	W	L_2	17.0 ± 2.7bc
Aug. 21				S	L_2	40.0 ± 5.4d	W	L_2	22.3 ± 4.8cd
Aug. 29				S	L_2	52.0 ± 2.0e	W	L_2,L_3	19.7 ± 3.2bc
Sept. 5							W	L_2,L_3	27.3 ± 4.3de
Sept. 10							T	L_2,L_3	28.0 ± 4.9de
Sept. 17							S	L_2-L_4	33.6 ± 8.2e
Sept. 26							S	L_3,L_4	51.0 ± 5.7f
Oct. 3							S	L_3,L_4	52.0 ± 4.6f

* W = whorl, T = tasseling, S = silking.

† L_1 - L_5 = larval instars, P = pupae.

‡ Means within columns which are followed by the same letter are not significantly different ($P > 0.05$) as determined by chi-square contigency table analysis.

Table 2. ECB densities and feeding damage at harvest in early-(May), mid-(June), and late-(July) season plantings of hybrid sweet corn at Hamden, Connecticut 1986.

Planting date	Harvest date	Mean ± SE no. ECB/50*		Mean ± SE % damaged†		Mean ± SE % damaged plants with live ECB†
		Stalks	Ears	Stalks	Ears	
May 12	August 12	6.7 ± 2.3a	1.1 ± 1.6b	3.6 ± 1.0a	32.3 ± 5.3a	34.2 ± 6.6a
June 11	September 3	63.8 ± 6.5b	19.0 ± 1.6b	37.0 ± 3.3b	73.0 ± 4.8b	92.9 ± 1.1b
July 10	October 9	43.8 ± 7.8b	9.0 ± 2.0c	19.0 ± 2.1c	59.7 ± 2.1c	82.1 ± 7.4b

* Means within columns followed by the same letter are not significantly different ($P > 0.05$) as determined by Duncan's (1955) multiple range test.

† Percentages within columns followed by the same letter are not significantly different ($P > 0.05$) as determined by chi-square contigency table analysis.

ACKNOWLEDGMENT

I thank Paul Moore and Darlene Stasiak for their technical assistance in the field.

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ERRATUM

In the Journal of Agricultural Entomology, Volume 5, Number 3, July 1988, there are two errors in Table 1 on page 157 under "June 11 -- ECB stage†" the correct entry for Aug. 7 should have been "L_5P" and the correct entry for Aug. 14 should have been "P,L_1." The following is the corrected table in its entirety:

Table 1. Weekly ECB feeding damage in early-(May), mid-(June), and late-(July) season plantings of hybrid sweet corn at Hamden, Connecticut 1986.

	May 12			June 11			July 10		
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July 3	W	L_2	26.0 ± 4.0a						
July 11	T	L_2,L_3	36.0 ± 3.6d	W	L_1,L_2	17.0 ± 4.9a			
July 17	T	L_3	28.0 ± 4.3a	W	L_2,L_3	8.7 ± 1.6b			
July 23	S	L_3,L_4	12.4 ± 3.4c	W	L_3,L_4	4.3 ± 2.4c			
July 30	S	L_4,L_5	10.0 ± 2.3c	W	L_4,L_5	6.3 ± 1.2bc	W	L_1	10.0 ± 2.0a
Aug. 7	S	L_5P	14.0 ± 1.0bc	T	L_5,P	4.0 ± 0.9c	W	L_1	16.0 ± 4.3b
Aug. 14				S	P,L_1	8.7 ± 1.6b	W	L_2	17.0 ± 2.7bc
Aug. 21				S	L_2	40.0 ± 5.4d	W	L_2	22.3 ± 4.8cd
Aug. 29				S	L_2	52.0 ± 2.0e	W	L_2,L_3	19.7 ± 3.2bc
Sept. 5							W	L_2,L_3	27.3 ± 4.3de
Sept. 10							T	L_2,L_3	28.0 ± 4.9de
Sept. 17							S	L_2-L_4	33.6 ± 8.2e
Sept. 26							S	L_3,L_4	51.0 ± 5.7f
Oct. 3							S	L_3,L_4	52.0 ± 4.6f

* W = whorl, T = tasseling, S = silking.

† L_1 - L_5 = larval instars, P = pupae.

‡ Means within columns which are followed by the same letter are not significantly different ($P > 0.05$) as determined by chi-square contigency table analysis.

ERRATUM

In the Journal of Agricultural Entomology, Volume 5, Number 3, July 1988, the data in Table 2 on page 158 were inadvertently reversed in the two columns under "Means ± SE % damaged†"; under "Ears" in the column "Mean ± SE no. ECB/50*," the first entry should be "1.1 ± 1.6a." The following is the corrected table in its entirety:

Table 2. ECB densities and feeding damage at harvest in early-(May), mid-(June), and late-(July) season plantings of hybrid sweet corn at Hamden, Connecticut 1986.

		Mean ± SE no. ECB/50*		Mean ± SE % damaged†		
Planting date	Harvest date	Stalks	Ears	Stalks	Ears	Mean ± SE % damaged plants with live ECB†
May 12	August 12	6.7 ± 2.3a	1.1 ± 1.6a	32.3 ± 5.3a	3.6 ± 1.0a	34.2 ± 6.6a
June 11	September 3	63.8 ± 6.5b	19.0 ± 1.6b	73.0 ± 4.8b	37.0 ± 3.3b	92.9 ± 1.1b
July 10	October 9	43.8 ± 7.8b	9.0 ± 2.0c	59.7 ± 2.1c	19.0 ± 2.1c	82.1 ± 7.4b

* Means within columns followed by the same letter are not significantly different ($P > 0.05$) as determined by Duncan's (1955) multiple range test.
† Percentages within columns followed by the same letter are not significantly different ($P > 0.05$) as determined by chi-square contigency table analysis.

NOTE: These are not author's errors. The author had marked these changes in the final proof, and the correction was made on the final galley; however, when the final typeset copy of the sideways-tablular material was produced, the old disk was inadvertently utilized. The editor regrets any inconvenience caused by these errors.

CABBAGE APHID, *BREVICORYNE BRASSICAE (L.)*,[1] CONTROL IN BRUSSELS SPROUTS IN RELATION TO CROP DEVELOPMENT

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Abstract: *Brevicoryne brassicae* (L.) infestations in brussels sprouts heads were characterized seasonally in relation to sprout development from Santa Cruz Co., California field plantings during 1984 and 1985. Plants received 0 to 9 insecticide sprays applied at 2 week intervals in 1984. Plants received 0 to 5 insecticide sprays applied at 3 week intervals in 1985.

Number of *Brevicoryne brassicae* contaminated sprouts at harvest were significantly greater ($P < 0.05$) on plants sprayed 0, 1 or 2 times than on plants sprayed 3 or more times in 1984. A significant ($P < 0.01$) relationship between proportion infested sprouts and number of insecticide applications was observed in both years. The period of initial sprout infestation appears to coincide with lateral bud development which occurs rapidly after apical stalk removal (topping). A significant difference ($P < 0.05$) in sprout plant weight between treatments was found in 1984, with lower weight for plants receiving more sprays.

Key Words: Cabbage aphid, *Brevicoryne brassicae*, brussels sprouts, damage, development, distribution.

J. Agric. Entomol. 5(3): 161-167 (July 1988)

Approximately 2200 ha of brussels sprouts *Brassicae oleracea gemmifera* are grown in California, with the majority being located on coastal plateaus along the Pacific Ocean from Monterey Bay to Half Moon Bay south of San Francisco. Aphids are considered the most serious pests of brussels sprouts in this area (Picket et al. 1983) because of quality standards imposed on local growers by packers who permit at most 10% of the sprouts at harvest to be infested with this insect for market. The cabbage aphid, *Brevicoryne brassicae* (L.), is particularly important because of the difficulty in controlling this species or removing it from the produce once it has entered individual sprouts. Fear of aphid contamination and to a lesser degree yield loss has resulted in a situation where insecticides are routinely applied for aphid control from early season to harvest.

Wilson, et al. (1983) developed a sampling program for both *B. brassicae* and the green peach aphid, *Myzus persicae* (Sulzer), which incorporated provisional control action thresholds based on those developed for broccoli by Trumble, et al. (1982). Pickel and Bigelow (1983) demonstrated that the program could reduce insecticide treatments as much as 50% when compared to conventional preventative spray programs.

In California, brussels sprouts are transplanted into fields in late spring. The stalks are allowed to grow until late summer when the apical portion of the stalk is removed (topping), forcing the lateral bud nodes along the stalk to mature into harvestable sprouts. The specific objective of this research focused on characterizing

[1] HOMOPTERA: Aphididae. Accepted for publication 2 June 1988.

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the infestation of the developing sprouts by *B. brassicae* spatially on the plant and seasonally in relation to sprout development in order to determine the potential for delaying or omitting early season aphid treatments.

METHODS

Small Plots

Brussels sprouts were transplanted onto twelve 30m long beds at the Sheriff's Rehabilitation Center in Watsonville, Santa Cruz Co., CA on 31 May, 1984. The plants were topped on 7 September, and harvested on 2 November.

Approximately five field collected *B. brassicae* were transferred to each plant after the plant became established to insure uniform infestation. Plant growth was monitored weekly by counting the number of nodes present on each of 10 plants randomly selected from the field. Plants from each treatment were examined for the presence of aphids to insure that all test plants were infested before initial treatments were applied. Treatments consisted of 0 to 9 applications of oxydemeton-methyl applied to ten successive plants in a row with four replications of each treatment placed at random in the field. The insecticide was applied with a backpack sprayer at a rate of 0.092 kg (A.I.)/hectare. Two untreated plants were left between each plot as a buffer to prevent insecticide drift. Plants receiving 9 applications were first treated 33 days after transplanting (3 July) with the remainder of the sprays being applied on two week intervals until two weeks before harvest (19 October). Plants treated fewer times received oxydemeton-methyl at two week intervals beginning successively one spray period later.

At harvest, leaves were removed from each plant and the remaining stalk of sprouts cut and weighed. Ten plants were sampled from each treatment replicate. Every 4th sprout was removed from each plant so that a total of 20 sprouts per plant could be examined for the presence of *B. brassicae*. Plant weight and proportion of aphid infested sprouts were compared between treatments by multiway analysis of variance and Duncan's (1951) multiple range test. Binomial data were transformed using an arc sine conversion prior to analysis.

Large plots

In 1985, the results of the 1984 study were tested on a 2 ha commercial field transplanted on 4 June at Wilder Ranch State Park north of Santa Cruz Co., CA. The plants were topped on 16 September, and the field harvested on 28 October.

The entire field was divided into 6 blocks of 8 row buffers on each end. The rows were ca. 130m long. Treatments consisted of 0 to 5 applications of insecticides applied with a tractor driven ground rig and 9 row wide boom. The 8 row buffers were sprayed at the growers' discretion approximately every 2 to 3 weeks. Plots receiving 1 to 4 treatments were sprayed with a mix of oxydemeton-methyl at a rate of 0.092 kg (A.I.)/hectare and mevinphos at a rate of 0.180 kg (A.I.)/hectare on each date. The plot receiving 5 treatments was side dressed with disulfoton at a rate of 0.023 kg (A.I.)/hectare for the first treatment and sprayed with the mixture of oxydemeton-methyl and mevinphos for the subsequent four treatments. Plants receiving five applications were first treated on 15 July. Additional sprays were applied on 7 August, 29 August, 20 September, and 10 October. Plants receiving fewer treatments were first sprayed on those dates, but successively later during the season. *Brevicoryne brassicae* populations were monitored weekly in untreated

plots using the binomial sampling plan developed by Wilson et al. (1983). In this method, each plant is examined for presence of aphids, but total number of aphids are not counted. It is possible to estimate population density because of the correlation between proportion of plants infested and mean number of aphids per plant.

At harvest, 20 plants were removed at random from the middle rows of each treatment. The leaves were removed from each plant and the remaining stalk of sprouts weighed. Twenty sprouts were removed at random from the full length of each stalk, cut open, and examined for the presence of *B. brassicae* to determine proportion infestation and within-plant distribution of aphids.

RESULTS

Aphid infestation

A significant relationship ($P < 0.01$) was observed between the mean proportion of cabbage aphid infested sprouts at harvest and the number of insecticide applications in both 1984 ($Y = 75.865\text{-}10^{0.168x}$) and 1985 ($Y = 46.739\text{-}10^{0.175x}$) (Fig. 1). *Brevicoryne brassicae* infestations on plants receiving 3 or more applications in 1984 were not significantly different ($P > 0.05$; $n = 4$) from one another while those receiving 0, 1, or 2 sprays were significantly different ($P < 0.05$; $n = 4$) from those receiving more than 2 sprays and from one another. The same trend occured in 1985 on the commercial plots. In both years, this corresponds to the period after topping in terms of sprout formation (Fig. 2).

In 1984, plants were artificially infested to insure uniform infestation before initial treatment. The proportion of aphid infested plants remained above 0.50 in the untreated plants the remainder of the season.

In 1985, the proportion of *B. brassicae* infested plants in untreated plots remained above 0.50 for most of the season (Fig. 3).

Brussels sprouts weight

There was a significant difference ($F = 2.02$; $P < 0.05$; $DF = 9$) in the weight of sprout plants treated with different numbers of insecticide applications in 1984 (Table 1). In general, plants receiving more sprays tended to weigh less that plants with fewer sprays. Plants sprayed 4 to 9 times weighed on average 18% less than plants sprayed 0 to 3 times. No relationship between plant weight and number of insecticide applications was observed in 1985.

Within-plant distribution

A greater proportion of *B. brassicae* infested sprouts tended to be found mear the apex of the plant than the base at harvest (Fig. 4). Analysis of variance, however, showed no significant differences ($P > 0.05$) by sprout location. It is our observation that sprouts nearer the apex tend to grow larger than those lower on the plant, and that leaf infestations appear to be greater on the more apical leaves throughout the season.

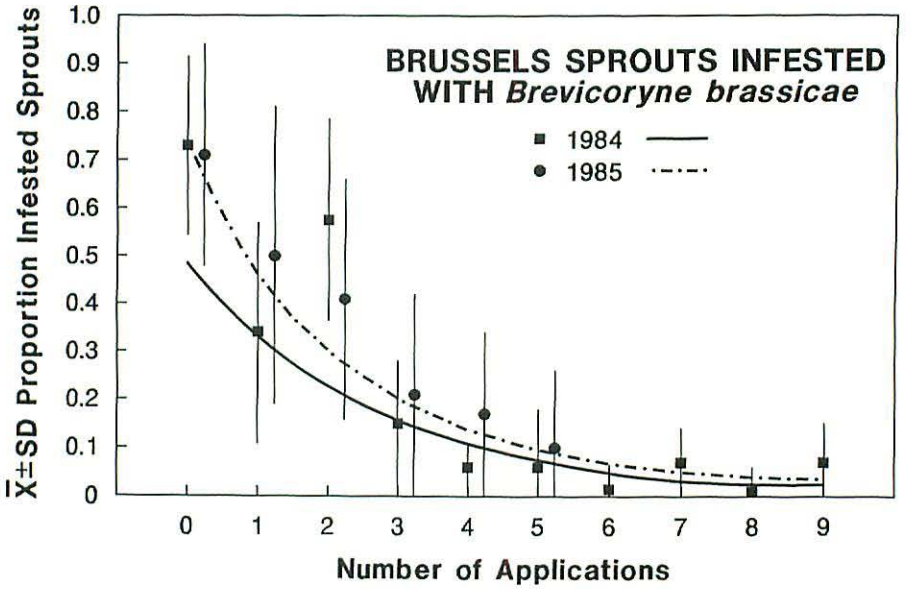

Fig. 1. Mean (± SD) proportion of cabbage aphid infested sprouts at harvest in 1984 and 1985 for plants receiving different numbers of insecticide applications.

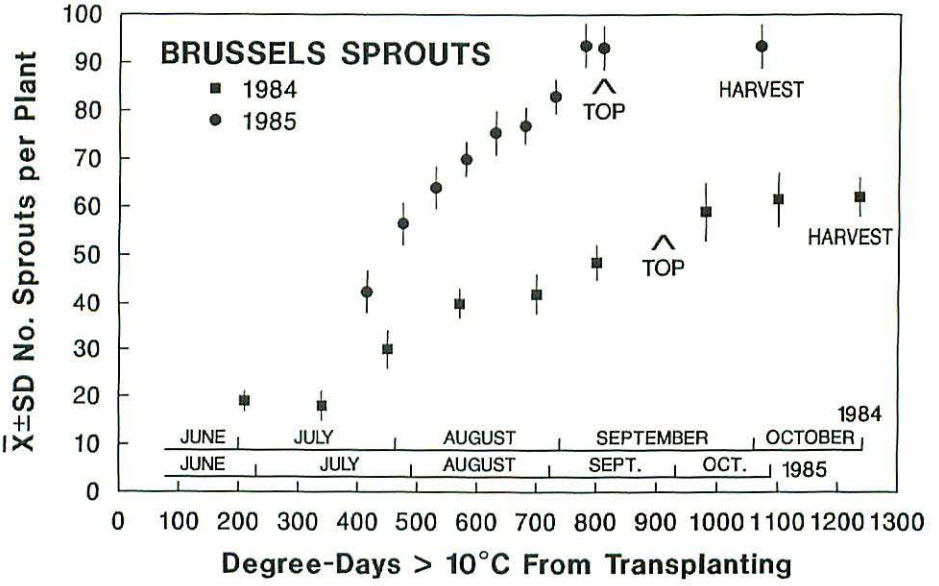

Fig. 2. Development of brussels sprouts plants in relation to degree-days greater than 10°C (Baskerville and Emin 1969) and calendar time in 1984 and 1985.

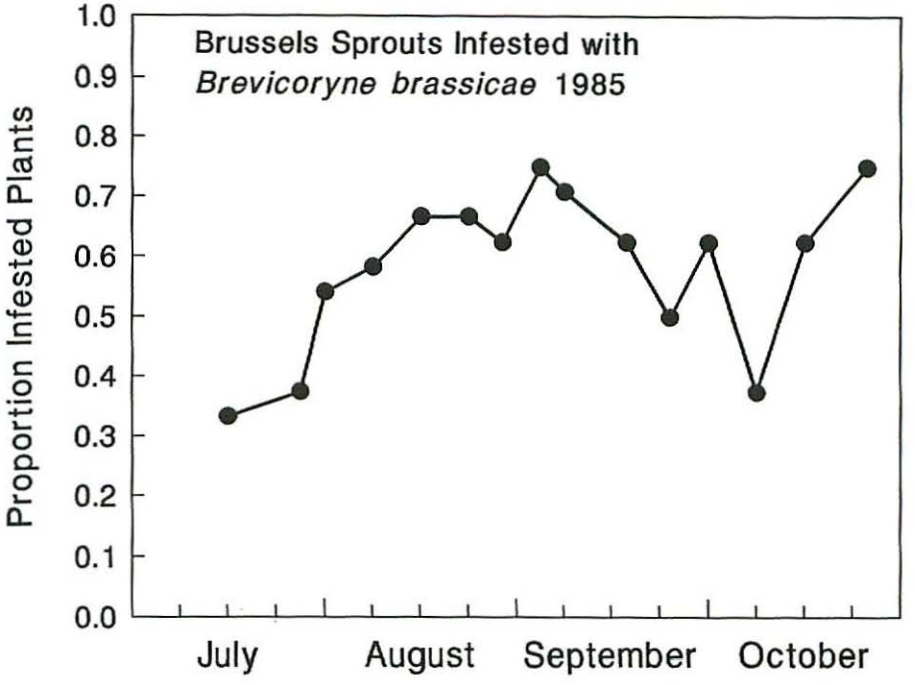

Fig. 3. Proportion of *Brevicoryne brassicae* infested plants in untreated plots during 1985.

Table 1. Weight (grams) of brussels sprouts plants treated with different numbers of insecticide sprays in 1984.

# of Sprays	$\bar{X} \pm SD$
0	2100.0 ± 429.3 AB*
1	1909.6 ± 520.4 ABC
2	1959.5 ± 203.8 ABC
3	2186.3 ± 520.2 A
4	1614.8 ± 416.4 BC
5	1873.3 ± 221.2 ABC
6	1592.1 ± 341.8 C
7	1891.4 ± 653.5 ABC
8	1651.1 ± 541.9 BC
9	1596.6 ± 605.8 C

* Means followed by the same letter are not significantly different (P = 0.05; Duncan's [1951] multiple range test).

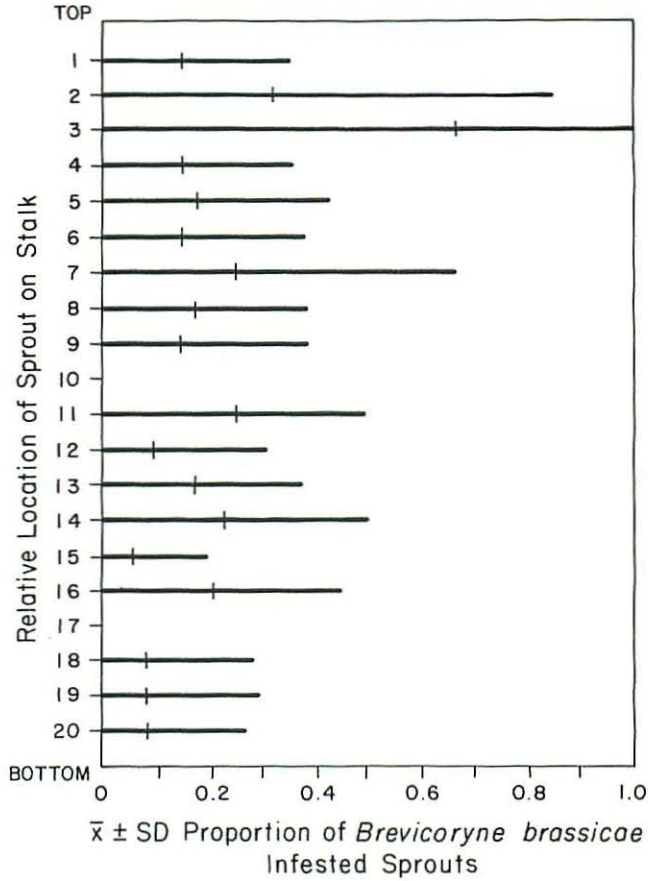

Fig. 4. Proportion of *Brevicoryne brassicae* infested sprouts at harvest in relation to sprout location on the plant.

DISCUSSION

The results of our study suggest that insecticide applications applied after topping are necessary to reduce *B. brassicae* infestations in sprouts at harvest, and that at least one application prior to topping may also be necessary to reduce aphid abundance to manageable levels. The importance of keeping aphid populations low after topping is likely due to sprout formation. Individual sprouts enlarge as the lateral buds are forced to develop after topping. We assume that prior to topping, sprouts are too compact to permit aphid entry. Once aphids are within the sprout heads, it appears that oxydemeton-methyl, and presumably most insecticides with contact action, will not sufficiently reduce infestations to meet quality standards.

Church and Strickland (1954) working in England stated that approximately 95% of *B. brassicae* infesting a brussels sprouts plant in late summer were on the leaves. Hafez (1961) in the Netherlands confirmed that aphids occur primarily on

leaves earlier in the season, but noted that in autumn a considerable fraction of the population were found on the sprouts. This is consistent with our observation that when not excluded by treatment in the period of sprout head maturation, *B. brassicae* do colonize the sprouts.

In 1984, a reduction in plant weight appears to correspond to a greater number of oxydemeton-methyl applications. Sances et al. (1981) has shown similar weight reductions in other crops following repeated insecticide treatment. They speculate that excessive insecticide use disrupts normal physiological processes of the plant. The phenomenon was not observed in 1985. However, the maximum number of insecticide applications were fewer with no plant receiving more than 4 oxydemeton-methyl treatments.

CONCLUSION

It is necesary to initiate control actions if *B. brassicae* are present when the sprouts become hostable after topping or when sprout heads begin to develop and mature. It may be necessary to initiate control actions prior to topping to reduce aphid infestations to manageable levels for the period of sprout enlargement. Multiple insecticide sprays applied prior to this time reduce the abundance of aphids, but do not by themselves appear to significantly improve yield or quality.

ACKNOWLEDGMENT

This study was funded in part by the California State Parks Department, the California General Services Administration, the Association of Monterey Bay Area Governments, and the Santa Cruz County Farm Bureau.

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RESPONSE OF LEAF PHOTOSYNTHESIS AND WATER RELATIONS OF IMPATIENS AND PEACH TO THRIPS[1] INJURY

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Abstract: The effect of injury by a leaf-feeding thrips, *Echinothrips americanus* Morgan, on leaf photosynthesis and water relations of impatiens, *Impatiens wallerana* Hook f., and peach, *Prunus persica* L. Batsch, was investigated. These plants differ in growth characteristics and leaf morphology. Thrips injury reduced leaf photosynthetic rates, but increased stomatal conductance of impatiens. Impatiens leaf water potential was not affected by thrips injury. *Echinothrips americanus* adults fed primarily on the upper leaf surface of peach. Feedinginjury reduced leaf photosynthesis, but had no effect on stomatal conductance and transpiration rates of peach leaves. These findings suggest possible mechanisms by which thrips injury reduces leaf photosynthesis.

Key Words: Trhips, *Echinothrips americanus*, photosynthesis, stomatal conductance, water relations, impatiens, peach.

J. Agric. Entomol. 5(3): 169-177 (July 1988)

Injury by leaf-feeding thrips causes direct leaf damage and has been found to reduce growth and yield of a number of plants (Lewis 1973). Thrips possess highly modified, asymmetrical mouthparts consisting of a single mandibular stylet and pairedm axillary stylets surrounded by a mouthcone, thus allowing thrips to pierce and suck fluids from plant cells. In a histological study with wheat, Chisholm and Lewis (1984) found that *Limothrips cerealium* (Halliday) completely removed the contents of mesophyll cells while feeding which resulted in the collapse of surrounding epidermal cells. More extensive injury completely disrupted leaf cell structure causing desiccation of mesophyll and epidermal cells. They further postulated that this feeding behavior was representative of injury by terebrantian thrips.

Although geeding behavior and the resulting injury of leaf-feeding thrips have been studied (Chisholm and Doncaster 1982; Chisholm and Lewis 1984; Heming 1978), the impact of injury by leaf-feeding thrips on host leaf photosynthesis and water relations has not been investigated. *Echinothrips americanus* Morgan is a polyphagous, leaf-feeding thrips with a host range than includes many broadleaved foliage and woody ornamental plant species. This terebrantian thrips has been recognized recently as a pest of numerous ornamental and greenhouse plants. We investigated the impact of leaf feeding by *E. americanus* on leaf photosynthesis and water relations of two plant species that vary in growth characteristics and leaf morphology. The two plants selected for study were impatiens, *Impatiens*

[1] THYSANOPTERA: Thripidae. Accapted for publication 4 January 1988.

wallerana Hook f., and peach, *Prunus persica* L. Batsch. Both species are dicotyledons and possess a leaf structure with distinct spongy mesophyll and palisade layers. Impatiens, however, is an annual, foliage plant with succulent leaves, whereas peach is a perennial, woody species with relatively firm leaves.

MATERIALS AND METHODS

Echinothrips americanus were obtained from a stock colony that had been maintained in the greenhouse for 1 year. The stock colony was reared on Irish shamrock, *Oxalis acetosella* L.

Impatiens Study

Impatiens were seeded in pots containing a potting mix (2:2:2:1 by volume of peat, pine bark, perlite, sand) and were grown in the greenhouse for 2 months. Pots were watered daily, and a complete nutrient solution (200 ppm of N:P:K) was supplied twice per week. Four randomly selected plants of uniform size were placed in screen cages within the greenhouse. Four treatments were established by infesting cages with 0, 10, 20, and 40 adult *E. americanus* per plant. Thrips were allowed to feed and reproduce for 25 days. At that time, leaves of similar size, age, and position within the canopy were selected and categorized by damage severity. A scale of damage severity based on the percentage of discolored leaf area caused by feeding punctures was used, where (0) = no visible damage, (1) = 1 to 33%, (2) = 34 to 67%, and (3) = 68 to 100%. Leaves were randomly selected and rated for damage by one person. Selected continued until 15 leaves in each damage category were chosen. Final thrips density was determined for each leaf by counting immature and adult thrips under a 2X magnification.

Leaf photosynthesis and stomatal conductance measurements were made in a growth chamber under conditions of 25.5°C, 46 to 56% RH, and photosynthetically-active-radiation levels of 55 $\mu E\ m^{-2}\ s^{-1}$. Measurements were made with LI-6000 (LI-COR, Lincoln, NE) closed gas-exchange photosynthesis system equipped with a 1.0-liter leaf chamber. Ten measurements were taken at 5-sec intervals over a 1-min period to calculate mean photosynthetic rates and stomatal conductance. Each leaf was excised, and leaf water potential was measured using a Scholander pressure bomb (Scholander et al. 1965). Leaf area of each leaf also was measured using a LI-3000 (LI-COR, Llincoln, NE) leaf area meter. Photosynthesis, stomatal conductance, and leaf water potential values (y) were regressed against final thrips density (x) using the linear model $y = a + bx$ (SAS Institute 1985). Plant physiological measurements also were analyzed by damage rating category using an analysis of variance and Duncan's (1955) multiple range test (Steel and Torrie 1960).

Peach Study

'Lovell' peach seedlings were established in 2.5-liter pots containing a 2:1 soil:sand mixture. Seedlings were grown in the greenhouse for six months before selecting ten uniformly-sized plants for experimentation. All plants were watered throughout the study to maintain soil water potential near field capacity. A complete nutrient solution (200 ppm of N:P:K plus soluble trace element (fertilizer) was supplied weekly to each plant.

Six leaves of similar size, age, and height from the soil surface were selected on each plant. The center portion of five leaves was covered with a thrips cage that consisted of a 5 × 5-cm piece of foam (0.3 cm thick) with the center 3 × 3 cm area removed. A piece of foam with the center removed was placed on the upper and lower leaf surfaces and were covered with plexiglass to form a cage around the leaf. The cage was clipped together and attached to a support stake. Cages were wider than the peach leaves, thus thrips could move freely between leaf surfaces. Cages were infested with either 0, 5, 10, 20, or 40 adult *E. americanus*. A sixth leaf served as an uncaged control. Adult thrips fed for seven days after which cages and thrips were removed.

Photosynthesis, stomatal conductance, and transpiration of the leaf section that was caged were measured following cage removal. Measurements were made with an LI-6000 portable, closed gas-exchange photosynthesis system equipped with a 1.0-liter leaf chamber. Measurements were taken as described in the impatiens study in the greenhouse at 27 to 30°C, 50 to 70% RH and photosynthetically-active-radiation levels of 700 to 900 $\mu E\ m^{-2}\ s^{-1}$. The leaf was excised, and leaf area of the caged portion was measured with a LI-3000 portable leaf area meter. Leaf water potential was not measured because thrips injury was confined to only a portion of the leaf.

Leaf damage was rated using the 0 to 30 scale described in the impatiens study. Although thrips mortality within the cages was not determined, it was apparent that differential amounts of feeding injury occurred within the same treatment. Therefore, the amount of feeding activity was estimated by counting the final number of fecal droppings on both surfaces of each leaf. Daily fecal production of an adult *E. americanus* was determined in a companion study that was conducted in a growth chamber using the same environmental conditions as described in the impatiens study. Five adult thrips were placed on an excised peach leaf within each of twelve petri dishes. Petri dishes were lined with moistened filter paper and fecal production was monitored daily for four days. One adult *E. americanus* produced ($\bar{x} \pm$ SE) 11.9 ± 2.7 fecal droppings per day. Fecal dropping counts were used to calculate the number of thrips feeding days (thrips-days) where, thrips-days = the number of fecal droppings per leaf ÷ the mean daily rate of fecal production per adult.

Photosynthetic and transpiration rates and stomatal conductance (y) were regressed on the number of thrips-days (X) using a linear model of $y = a + bx$ (SAS Institute 1985). Plant physiological measurements also were analyzed by initial thrips density and damage rating category using an analysis of variance, with means being separated using Duncan's (1955) multiple range test (Steel and Torrie 1960).

RESULTS

Impatiens Study

Thrips caused extensive damage to impatiens leaves with some leaves in the highest density treatment abscising before the end of the feeding period. Leaf photosynthetic rate declined linearly ($F = 11.15$; $P = 0.0015$; df = 1,58) with increasing final thrips density (Fig. 1). Inclusion of a quadratic term into the model was not significant ($F = 2.50$; $P = 0.12$; df = 1,57). Conversely, stomatal conductance increased linearly ($F = 24.72$; $P < 0.0001$; df = 1,58) with increasingly

greater thrips density (Fig. 2). Leaf water potential was not significantly ($F = 2.74$; $P = 0.10$; $R^2 = 0.05$; df = 1,58) related to thrips density.

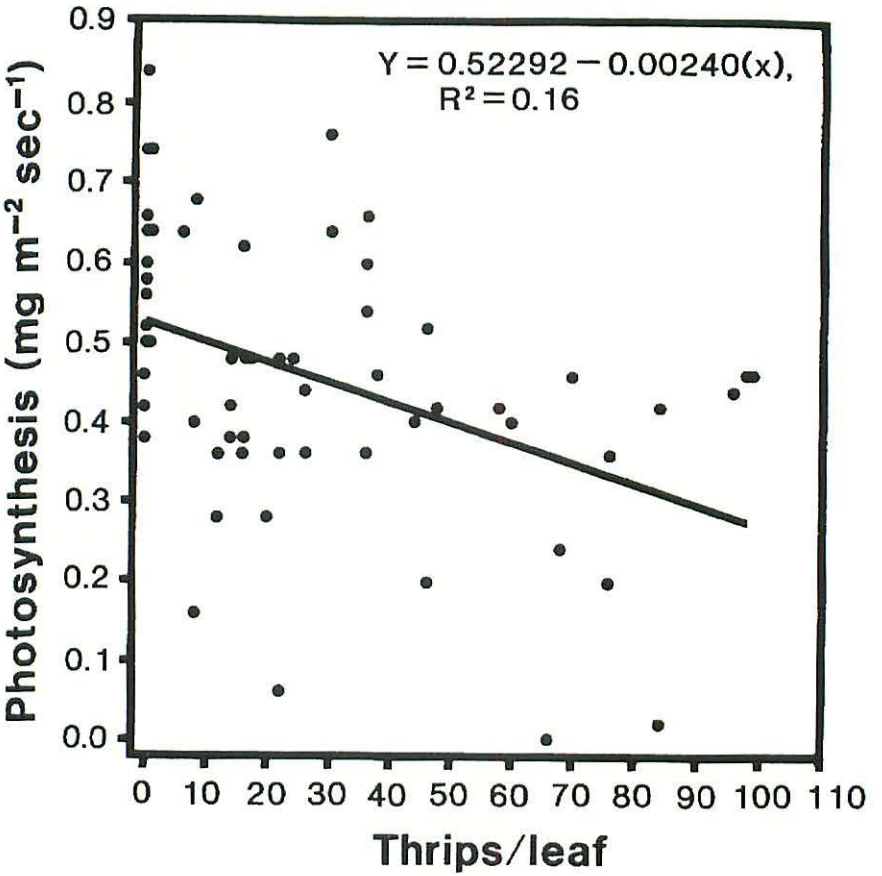

Fig. 1. Response of impatiens leaf photosynthesis to thrips feeding injury as measured by number of thrips per leaf.

Final mean (± SE) thrips densities on leaves with damage ratings of 0, 1, 2, and 3 were 0, 17.0 ± 2.3, 38.3 ± 4.3, and 60.6 ± 7.8 thrips per leaf, respectively. Leaf photosynthesis declined and stomatal conductance increased with increasingly severe damage (Table 1). Leaf water potential was not affected by damage rating ($F = 1.15$; $P = 0.34$, df = 3,56).

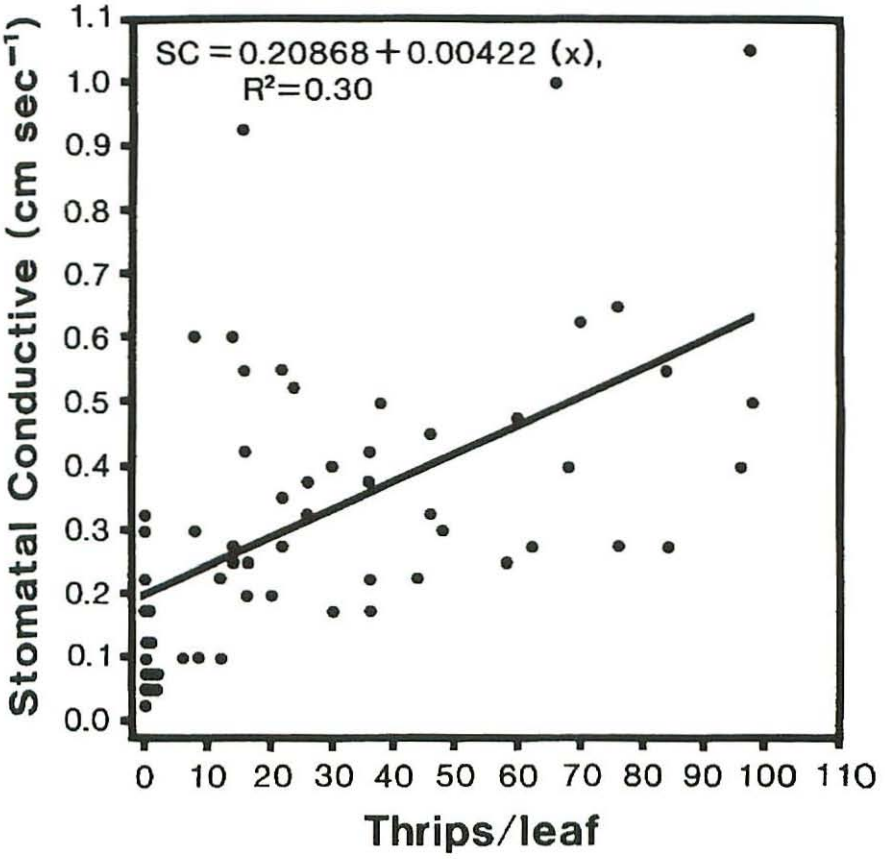

Fig. 2. Response of leaf stomatal conductance in impatiens to thrips feeding injury as measured by number of thrips per leaf.

Peach Study

The cages used in this study did not cause noticeable leaf damage nor did they significantly ($P > 0.05$) affect leaf photosynthesis, transpiration and stomatal conductance. Adult *E. americanus* feeding caused visible injury to peach leaves after seven days. Densities of 0, 5, 10, 20, and 40 thrips per cage cuased an average damage rating of 0.2, 0.3, 0.7, 1.9, and 2.0, respectively. Fecal production and thrips-days increased with increasingly severe damage (Table 2). When averaged across all treatments, 74.6% of fecal droppings occurred on the upper leaf surface which suggests that adults fed primarily on the upper leaf surface. However, we have observed that larvae feed primarily on the lower leaf surface.

Leaf photosynthesis declined with increasingly severe damage levels (Table 2). Leaf photosynthesis also declined linearly with increasing number of thrips-day ($F = 11.69$; $P = 0.0002$; $df = 1,58$) (Fig. 3), with the greatest decline in photosynthesis tending to occur between 0 to 15 thrips-day. Photosynthesis did not decline as

sharply at greater levels of feeding injury. addition of a quadratic term, however, did not significantly ($F = 3.56$; $P = 0.06$; $df = 1,58$) improve the linear model of photosynthesis and thrips-days. Leaf transpiration rates and stomatal conductance were not different between leaf damage ratings (Table 2). Likewise, thrips-days was not linearly related to leaf transpiration ($F = 0.18$; $P = 0.67$; $df = 1,58$) or stomatal conductance ($F = 0.43$; $P = 0.51$; $df = 1,58$).

Table 1. Leaf photosynthesis (PN) and stomatal conductance (SC) of impatiens leaves with various degrees of damage by *E. americanus*.

Damage rating*	PN ($mg/m^2/sec$)	SC (cm/sec)
0	0.59a	0.13a
1	0.47b	0.29b
2	0.46b	0.31b
3	0.31c	0.59c

Means followed by the same letter are not significantly different (P = 0.05, Duncan's (1955) multiple range test).

* Damage rating based on percentage of leaf area discolored by feeding injury, where (0) = no visible discoloration, (1) = 1-33%, (2) = 34-67%, and (3) = 68-100%.

Table 2. Thrips-days, thrips fecal production, and leaf photosynthesis (PN), transpiration (E), and stomatal conductance (SC) of peach leaves with various degrees of damage by *E. americanus* adults.

Damage* Rating	No. of leaves	Thrips-days	Fecal Spots per leaf	PN ($mg/m^2/sec$)	E ($mg/m^2/sec$)	SC (cm/sec)
0	28	1.9a	22a	0.741a	181.8a	1.51a
1	20	7.7b	92b	0.620b	183.8a	1.30a
2	4	27.1c	323c	0.581b	168.9a	1.20a
3	8	30.9c	368c	0.532b	189.2a	1.30a

Means followed by the same letter are not significantly different (P = 0.05, Duncan's (1955) multiple range test).

* Damage rating based on percentage of leaf area discolored by feeding injury, where (0) = no visible discoloration, (1) = 1-33%, (2) = 34-67%, and (3) = 68-100%.

DISCUSSION

Reduction in leaf photosynthesis as a result of injury by *E. americanus* was demonstrated for both impatiens and peach. Thrips feeding also increased stomatal conductance of impatiens, which implies an increase in leaf transpiration (Nobel 1974), but had no effect on stomatal conductance and transpirationf of peach leaves.

Leaf injury by thrips is superficially similar to leaf injury caused by spider mites (Chisholm and Lewis 1984; Sances et al. 1979b). Spider mite injury has been found to reduce leaf photosynthesis, conductance, and transpiration of strawberries (Sances et al. 1979a, b), peppermint (DeAnglis et al. 1982, 1983), almonds (Andrews and LaPre 1979), and peach (Mizell et al. 1986) with the greatest reductions occurring at low infestation levels. The initial reduction in photosynthesis and transpiration seems to be caused by stomatal closure which inhibits the exchange of carbon dioxide and water (Sances et al. 1979a; DeAnglis et al. 1983). Stomatal

closure, which is measured as a decrease in stomatal conductance, occurs in response to mite-induced water stress caused by mechanical disruption of the leaf epidermis and cuticle. This injury, however, enhanced cuticular transpiration in peppermint (DeAnglis et al. 1982). Additional reductions in photosynthesis occur as a result of mechanical damage of mesophyll cells (Sances et al. 1979a).

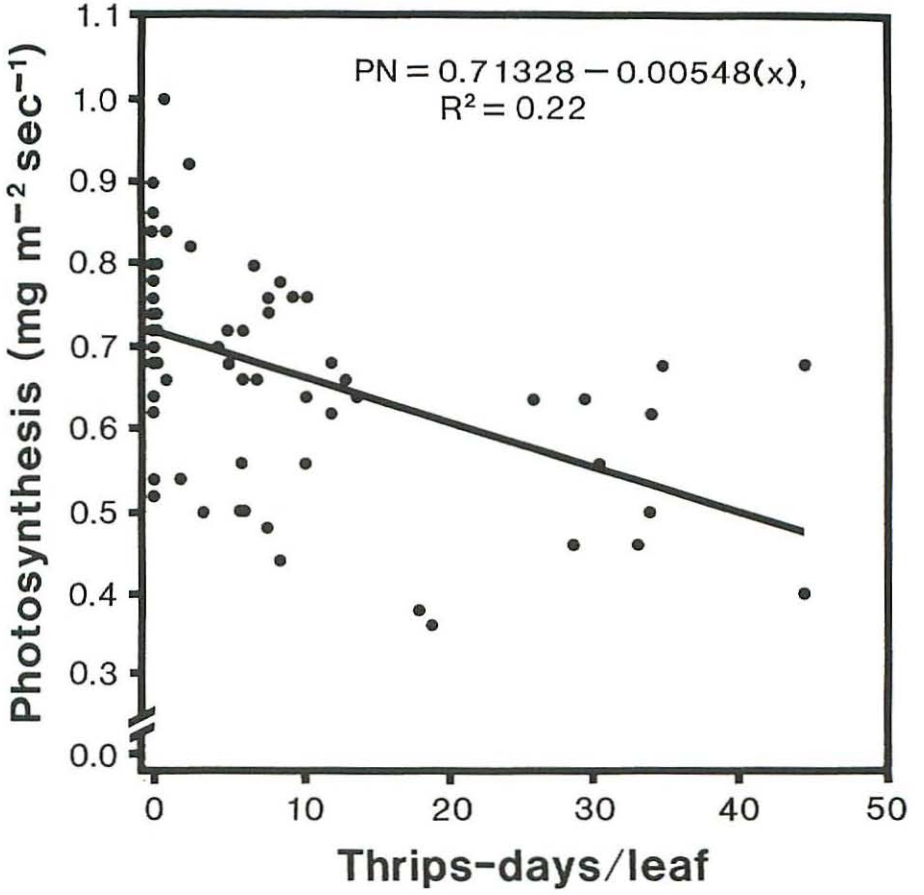

Fig. 3. Response of peach leaf photosynthesis to thrips feeding injury as measured by thrips-days.

Thrips injury reduced the leaf photosynthetic rate of impatiens and peach leaves in a manner similar to the reduction reported for spider mite injury. The mechanism of stomatal closure inhibiting gas exchange is not supported by our data in that stomatal conductance increased in impatiens, and stomatal conductance and transpiration were not affected by thrips injury in peach. It is possible that mechanical disruption of chlorophyll-containing mesophyll cells by thrips is more severe than with spider mites and that this damage is the primary mechanism whereby thrips reduce leaf photosynthesis. The lack of effect of thrips injury on

leaf water potential in impatiens suggest that leaves were not under moisture stress. However, the impatiens measurements were made under conditions of very low evaporatie demand. Consquently, stomata may have remained open and the increase in leaf conductance could reflect cuticular gas exchange as a result of epidermal damage. The lack of change in stomatal conductance and transpiration in peach leaves may be due, in part, to adult *E. americanus* feeding mostly on the upper leaf surface, whereas peach stomata occur entirely on the lower leaf surface (Mizell et al. 1986). Damage to mesophyll cells near the upper leaf surface could have reduced photosynthesis without extensively disrupting water movement through the leaf and stomatal function on the lower leaf surface. Results of our study suggest that this reduction probably is caused by mechanical damage of mesophyll cells, but more detailed information will be needed to determine conclusively the mechanism of photosynthesis reduction by thrips feeding. The reduction in leaf photosynthesis by thrips undoubtedly would lower photosynthate production by infested plants which eventually oculd be expected to reduce plant growth and productivity.

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EFFECT OF FIRST-GENERATION EUROPEAN CORN BORER[1] ON YIELD AND PLANT HEIGHT OF POPCORN[2]

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Abstract: A study was conducted to evaluate damage to popcorn by first-generation European corn borer (ECB), *Ostrinia nubilalis* (Hübner). Plants were artificially infested with 1, 2, 4, 8, or 16 ECB egg masses/plant at the mid-whorl stage of plant development. Numbers of egg masses, numbers of larvae, cm of lesions in the leaf midrib, cm of stalk cavities, and visual ratings were all correlated with yield losses and reduction in plant height. Trend comparisons showed the relationship between measurements of damage and yield losses and height reduction was non-linear with the greatest losses per unit of damage occurring at the lower levels of infestation.

Key Words: *Ostrinia nubilalis*, popcorn.

J. Agric. Entomol. 5(3): 179-183 (July 1988)

The European corn borer (ECB), *Ostrinia nubilalis* (Hübner) has been studied extensively as a pest of dent corn and to some extent as a pest of sweet corn. Few studies have been made of this insect as a pest of popcorn. Thomas et al. (1960) evaluated popcorn genotypes for resistance to leaf feeding by first-generation ECB; some differences were found but none of the genotypes were considered resistant. Jarvis and Guthrie (1980) evaluated popcorn plant introductions for resistance to sheath-collar feeding by second-generation ECB; all were susceptible. Jarvis et al. (1986) showed that the ECB causes significant yield losses in popcorn hybrids when plants were infested at midwhorl (the time of infestation by first-generation ECB by as few as an average of 0.5 egg masses per plant.

The only method used for evaluating damage to popcorn by first-generation ECB was that of Jarvis et al. (1986) who found a significant relationship between numbers of egg masses used for infestation and yield losses. The relationship between various measurements of first-generation ECB damage and yield losses in popcorn are not known. Several measurements, however, have been used for evaluating damage to dent corn by first-generation ECB. Everett et al. (1958) found an inverse relationship between numbers of leaf lesions and yield but concluded that the best index was the number of cavities or larvae per plant. Kwolek & Brindley (1959) showed that the number of cavities was a more reliable indicator of yield loss than the number of larvae. Jarvis et al. (1961) found that both number of cavities and leaf lesions were more reliable than number of larvae in estimating yield loss due to first-generation ECB. Chiang and Holdaway (1965) demonstrated that both height and yield of dent corn were reduced by a first-generation ECB infestation, and that there was a significant correlation between reduction in height and reduction in yield.

[1] LEPIDOPTERA: Pyralidae. Accepted for publication 16 June 1988.

[2] Joint contribution of Corn Insects Research Unit, United States Department of Agriculture, Agricultural Research Service, and Journal Paper No. J-12667 of the Iowa Agriculture and Home Economics Experiment Station, Ames 50011, and Ankeny 50021. Project 2513.

The purpose of this study was to evaluate damage by first-generation ECB to popcorn and to compare the reliability of several measurements of plant infestations for estimating yield loss and damage.

MATERIALS AND METHODS

Research plots were planted and managed for maximum popcorn yields (early planting, proper fertilization, herbicides before planting, and late cultivation for maximum weed control). The hybrid Iopop 12 was used for this study because it is susceptible to first-generation ECB and because it has been one of the most widely grown popcorn hybrids in the Midwest. Planting dates were 11 May 1984 and 26 April 1985 at the Iowa State University Research Farm, Ankeny, Iowa. Plots were planted with a 6-row conventional planter at the rate of 56,000 seeds per ha. All plots were thinned to 43,055 plants per ha. when plants were in the 3-to 6-leaf stage.

A randomized complete block experimental deisgn with 5 replications was used. Treatments consisted of 6 levels of ECB infestation: None (treated with a wettable powder formulation of the insecticide carbaryl (Sevin) at the rate of 3.4 kg of actual insecticide per ha.), 1, 2, 4, 8, and 16 ECB egg masses per plant (each mass = ca. 30 eggs). Natural infestation by first-generation ECB was very low and had no significant affect on popcorn plants. Egg masses were applied when plants were in the midwhorl stage of plant development. All plots were treated with insecticides (carbaryl) later in the season to prevent infestation by second-generation ECB.

Each replication consisted of a single row of popcorn. Within each row, each of the 6 ECB infestation levels was randomly assigned within the row. Each plot (various levels of ECB infestation) contained 40 plants spaced ca. 25 cm apart with at least 150 cm between plots. Rows were 76 cm apart with 1 guard row between rows containing plots to prevent larval migration between plots.

A 9-class rating scale (class 1 = no damage, class 9 = extensive damage to leaf tissue) as described by Guthrie et al. (1960) was used to evaluate leaf-feeding damage. Visual ratings were made on a plot basis 21 days after egg hatch. After ratings were made, 20 plants from each plot were dissected. ECB damage was determined by counting lesions in leaf midribs and measuring their accumulative length in cm, counting cavities in the stalk, and measuring their accumulated length in cm, and by recording numbers of ECB larvae per plant. After anthesis, when plants had reached their full height, 20 plants in each plot were measured from the ground to the top of the tassel.

Ears of popcorn were harvested by hand from the 20 remaining plants in each plot after field drying to ca. 20% moisture. Ears from each plot were placed in bags, hung in a drying building, and later shelled with a mechanical sheller. Yields were converted to 13.5% moisture and expressed in terms of quintals per ha.

The data from individual plants were averaged to obtain a mean for each treatment in each replication. The data for the 2 years were combined for an analysis of variance; means were separated by least significant differences. Regression equations were computed to determine the relationship between the independent variable (number of ECB egg masses, cm of leaf lesions, cm of stalk cavities, numbers of larvae, and visual ratings), and yields. Regression equations were also computed to determine the relationship between these independent variables and plant height, and between plant height and yields.

RESULTS AND DISCUSSION

Data on plant damage, yields, and plant height of popcorn infested by first-generation ECB are summarized in Table 1. Centimeter of leaf lesions, cm of stalk cavities, numbers of larvae and visual ratings were all greater at the higher levels of infestation, with the greatest difference among levels of infestation usually occurring in plants that were infested from 0 to 1 egg mass/plant. Yield was reduced by nearly 6 Q/ha and height was reduced by nearly 9 cm when plants were infested with only 1 egg mass/plant. Both yields and height were further reduced by higher levels of infestation.

Table 1. Yields, plant height, and damage to Iopop 12 popcorn hybrid by first-generation European corn borer.

No. ECB egg masses/ plant	Cm leaf lesions/ plant	Cm cavities/ plant	No. larvae/ plant	Visual rating	Yield Q/ha	Height (cm)
0	0.1	0.4	0.1	1.2	43.1	251.8
1	2.2	1.5	1.7	3.4	37.3	243.1
2	3.7	2.2	3.0	5.1	35.3	240.9
4	4.0	2.2	3.4	5.1	34.5	234.4
8	4.5	2.7	3.8	5.7	34.6	231.6
16	6.0	2.8	3.9	6.3	32.8	226.7
LSD-05	0.7	0.7	0.7	0.8	3.5	3.0

To evaluate the five measurements of first-generation ECB damage, regression equation (linear, quadratic, and cubic) were computed using the indices of ECB damage and infestation level as the independent variables. When regression equations were significant, the coefficient of determination was calculated. This is a proportion of the total sums of squares of the dependent variable (yield) that can be attributed to by variation in the independent variable (damage measurements). Trend comparisons and coefficients of determination of the relationships between the five measurements of first-generation ECB damage and yield and plant height are shown in Table 2. A highly significant amount of variation in yields among treatments can be accounted for by variation in the various damage measurements.

The significance of the quadratic and cubic trends indicates that yield losses and reduction in plant height per egg mass or per unit of damage were greater at the lower levels of infestation. There was also a significant relationship between plant height and yield with the taller plants having higher yields. Thus, infestation by the ECB when popcorn plants are in the mid-whorl stage of development can result in both reduced plant height and lower yields.

The coefficients of determination (Table 2), were about the same for all damage categories as well as larvae or egg masses. Thus, all of the variables (number of larvae, cm of lesions in leaf midribs, cm of cavities in stalks, and amount of damage to leaf tissue) can be used to estimate yield losses and damage to popcorn infested by first-generation ECB.

Damage measurements other than larval leaf feeding cannot be used to determine if chemical control measures are needed because by the time damage is

noticeable, chemical control would no longer be effective in preventing yield losses. However, egg mass counts or early larval leaf feeding could both be used to determine if chemical control should be applied. Popcorn is a high-value crop on a per-hectare basis and chemical control would be practical if potentially damaging ECB populations are present. Thus, popcorn growers should closely monitor ECB populations throughout the season.

Table 2. Trend comparisons and coefficients of determination between indicies of first-generation ECB infestation and yields, and between these indicies of first-generation ECB infestation and yields, and between these indicies and plant height.

	Trend comparisons*			Coefficients of determination		
	Linear	Quadratic	Cubic	Linear	Quadratic	Cubic
$\bar{X}$ no. of (cm) of leaf lesions per plant and yield	***	**	NS	.45	.49	NS
$\bar{X}$ no. (cm) of cavities per plant and yield	***	*	NS	.39	.43	NS
$\bar{X}$ no. of larvae per plant and yield	***	**	NS	.43	.47	NS
$\bar{X}$ no. of egg masses per plant and yield	***	***	***	.22	.31	.42
$\bar{X}$ visual rating and yield	***	***	NS	.33	.41	NS
Plant height and yield	***	NS	NS	.30	NS	NS
$\bar{X}$ no. (cm) of leaf lesions per plant and plant height	***	**	NS	.55	.59	NS
$\bar{X}$ no. (cm) of cavities per plant and plant height	***	NS	NS	.48	NS	NS
$\bar{X}$ no. of larvae per plant and plant height	***	**	NS	.39	.44	NS
$\bar{X}$ no. of egg masses per plant and plant height	***	***	***	.62	.74	.78
$\bar{X}$ visual rating and plant height	***	NS	NS	.49	NS	NS

* NS not significant, *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$.

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NOTE

FREQUENCY OF REGURGITATION BY LABORATORY-REARED FACE FLIES[1] FED TRYPTICASE-SOY BROTH

Key Words: *Musca autumnalis*, face fly, regurgitation.

J. Agric. Entomol. 5(3): 185-187 (July 1988)

The face fly, *Musca autumnalis* DeGeer, has been implicated as a vector of the bacterium *Moraxella bovis* Hauduroy, the causative agent of infectious bovine keratoconjunctivitis (Gerhardt et al., 1982, J. Amer. Vet. Med. Assoc. 180: 156-159; Arends et al., 1984, J. Econ. Entomol. 77: 394-398). Glass and Gerhardt (1984, J. Econ. Entomol. 77: 399-401) suggested that regurgitation of ingested lacrimal secretions containing viable *M. bovis* was a possible mechanism of transmission of this pathogen by face flies. This study was designed to determine the frequency of regurgitation in laboratory-reared face flies.

Newly emerged female face flies were placed in 450 ml cardboard containers with screened lids. Water and sucrose were provided ad libitum until the flies were 5 days old, after which water was provided. All flies were reared under a 16:8 (L:D) photoperiod, with temperature and relative humidity (RH) maintained at 24°C and 75%, respectively.

Trypticase-soy broth (TSB) has been shown to be a readily ingested source of protein for face flies (Glass et al., J. Econ. Entomol. 75: 545-546). Six-day-old flies which had been sucrose starved for 24 hours were placed individually in 45×13 mm glass vials. Each vial contained 10µl of a 1.2, 3.0, or 4.8% TSB solution (w/v), or 5, 10, or 15 µl of a 4.8% TSB solution. As a control, flies were offered 5, 10, or 15 µl of water. Each test was replicated 10 times. During each replication, 10 flies were presented with each TSB solution.

Following the introduction of a fly into a vial, the presence or absence of regurgitation droplets was recorded once every 10 minutes. Regurgitation droplets exuded from the food canal and were held at the tip of the proboscis for varying intervals prior to being sucked in. These droplets, which had a mean diameter of 1.1-1.3 mm (Coleman & Gerhardt, unpublished), were visible without magnification. The number of flies regurgitating/10 flies was recorded during each 10 minute period. Observations were made between 1000 and 1500 (EST) under fluorescent lights. Experiments were terminated when none of the flies in an experimental group regurgitated for three consecutive observation periods.

We observed a definite relationship between the amount of TSB presented to the flies and the length of time the flies regurgitated (Fig. 1). None of the flies which had been offered only water regurgitated. Few flies which had been offered only 5 µl of TSB regurgitated during any observation period (Fig. 1). All regurgitation by these flies occurred within the initial 70 min. Those flies offered 10 or 15 µl of TSB regurgitated for as long as 200 and 230 min, respectively. Increasing concentration of TSB solution had a similar effect on regurgitation (Fig. 2). Flies offered 1.2% TSB regurgitation for only 20 min, while flies offered 3.0 and 4.8% TSB regurgitated for 70 and 200 min, respectively.

[1] (DIPTERA: Muscidae). Accepted for publication 25 June 1988.

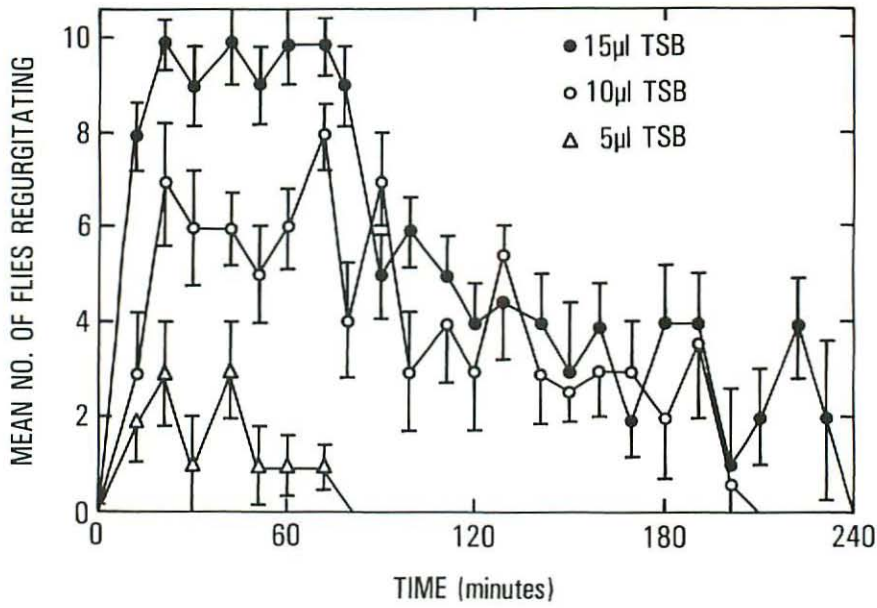

Fig. 1. Effect of the amount of trypticase-soy broth (TSB) on the incidence of regurgitation by laboratory-reared *Musca autumnalis* (SEM).

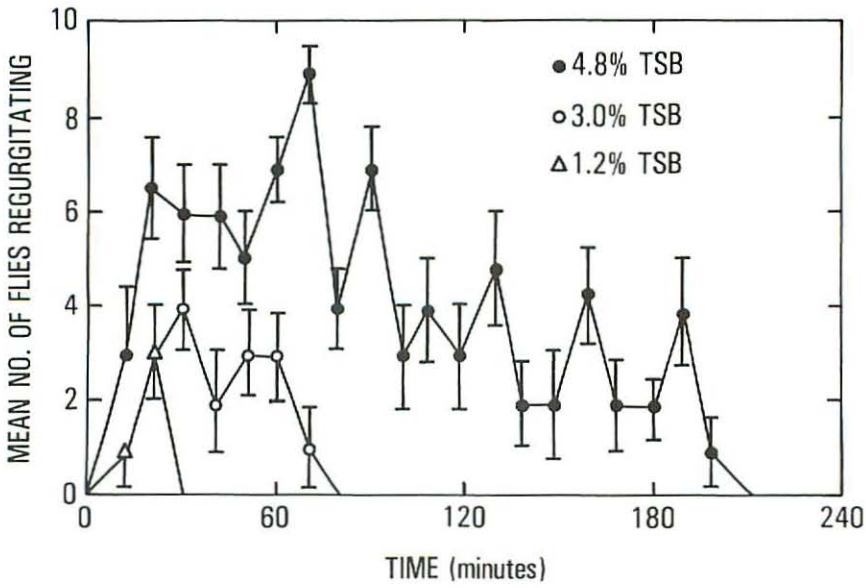

Fig. 2. Effect of the concentration of trypticase-soy broth (TSB) on the incidence of regurgitation by laboratory-reared *Musca autumnalis* (SEM).

These results indicate that regurgitation is most frequent when face flies; a) ingest large volumes of TSB, or b) when they ingest a highly concentrated TSB solution. Both of these factors affected the incidence of regurgitation; however, the relative importance of the amount and concentration of ingested solution (TSB) on regurgitation is difficult to determine. We have found that the volume of TSB ingested by face flies is related to the concentration of the substance (Coleman & Gerhardt, unpublished data). Flies offered 4.8% TSB solutions consumed ca. 12-15 µl of the substance, whereas flies offered less concentrated TSB ingested lesser amounts. This is in contrast to the reports by Van Geem & Broce (1986, Ann. Entomol. Soc. Am. 79: 1-6) who found that face flies readily fed to repletion on three concentrations of powdered egg in water. These differences may have resulted from the different food sources used in each study.

Whether the ingestion of oral and nasal secretions of cattle by face flies will elicit regurgitation under field conditions is unknown. These secretions are presumably quite different than TSB, and, as shown by our data, regurgitation is markedly affected by the nature of the ingested solution. Additional research is needed to determine the importance of regurgitation by face flies under field conditions and the epidemiological significance of this process towards transmission of *M. bovis*.

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WHITE GARDEN SNAIL (*THEBA PISANA* MUELLER[1]): EFFICACY OF SELECTED BAIT AND SPRAYABLE MOLLUSCICIDES

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Abstract: Laboratory and field cage tests were conducted to measure the toxicities of some currently available molluscicides against White garden snail (*Theba pisana* Mueller) (WGS). Of 19 commercial bait formulations tested, Deadline Bullets, a 4% metaldehyde bait at 1.79 kg AI/ha, and Slug-Geta, a 2% methiocarb bait at 0.98 kg AI/ha, were the most effective. WGS mortality in Deadline Bullets and Slug-Geta treated cages ranged from 17.1 to 82.3% and 9.3 to 81.2% higher, respectively, than in cages treated with the other 17 bait formulations. Results of laboratory testing of four sprayable molluscicide formulations were highly variable. However, results suggested that the three carbamate formulations tested, Zectran 2 EC, Mesurol 75 WP, and Lance 480 g/L, at 2.24 and 4.48 kg AI/ha, killed significantly higher numbers of WGS than did Slug N Snail Special spray, a 50% metaldehyde spray at 4.7 kg AI/ha. Additional field testing of the Deadline Bullets and Slug-Geta formulations against established WGS populations were conducted. Results revealed that pretreatment populations were reduced an average of 95 and 82% with two applications (applied at 14 or 21 day intervals) of Deadline Bullets of Slug-Geta, respectively.

Key Words: White garden snail, *Theba pisana*, molluscicides, chemical control.

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The White garden snail (WGS), *Theba pisana* (Mueller), a native of Sicily, is distributed throughout the Mediterranean, as well as Australia, Atlantic Islands, southern Africa, and western Europe (Anonymous 1982). It is considered an extremely injurious pest to ornamental flowers and shrubs and a wide variety of vegetables, fruits, and citrus in most of the areas of its distribution. In Israel, it was considered the most economically important of pest snails (Harpez and Oseri 1961). In France it was reported as a pest of seed alfalfa (Cairaschi and Lecomte 1973). In Australia, the snail is a significant pest of cereals (Baker 1986). WGS is also an intermediate host of the lungworm, *Muellerius capillaries* (Mueller), and several other nematodes that can be significant parasites of sheep and cattle (Baker 1986).

The WGS was first discovered in California in 1918 in La Jolla (Basinger 1927). Over the next 20 years, infestations were found in several other counties of southern California. All known infestations were declared eradicated in 1949 after eight years of negative finds in the infested counties (Gammon 1949). In 1985, the WGS was again detected in California. It was found established in five isolated locations in San Diego County. Its current presence in California poses a serious threat to the ornamental and citrus industries of southern California as well as other segments of California agriculture.

[1] STYLOMMATOPHORA: Helicidae. Accepted for publication 17 July 1988.

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Chemical control of WGS, in areas where it has become an established pest, typically employ metaldehyde or methiocarb (Mesurol®) or combinations of these chemicals with other molluscicides in a myriad of bait formulations or sprays (Godan 1983). However, there are not acceptable tolerance levels of metaldehyde on any crop intended for human food or for feeding poultry, dairy, or meat animals; therefore, it has limited use as a spray (Del Rivero and Roca 1969).

The degree of control of snails in the field has varied. Metaldehyde treatments applied when dry conditions are prevalent are usually more successful than when damp, moist conditions are present, at which time snails are most likely to be active. Godan (1983) and others have concluded that the principal effect of metaldehyde is thorough stimulation of the mucous glands which cause excessive sliming leading to death by dehydration. Therefore, under wet conditions many snails are able to avoid dehydration and negate the toxic effects of metaldehyde in a few days. Mesurol, on the other hand, appears to be most effective when moist conditions prevail (Godan 1983).

The purpose of this work was to select an effective molluscicide from commercially available formulations that could be implemented in a control or eradication program in San Diego County of California.

MATERIALS AND METHODS

Laboratory Tests

All candidate chemicals were initially tested in the laboratory and included: Bug-Geta 3.25% metaldehyde bait, Bug-Geta Plus 2% metaldehyde + 5% carbaryl bait, Slug-Geta 2% methiocarb bait (Chevron Chemical Co., Richmond, CA), Slug-N-Snail 3% metaldehyde bait, Slug-N-Snail Special Spray 50% metaldehyde (Cooke Laboratory Products, Commerce, CA), Slug and Snail 2.75% metaldehyde bait, Slug and Snail Bait M 2% methiocarb bait (J. R. Simplot Co., Lathrop, CA), Deadline Bullets 4% metalydehyde bait, Deadline 40 4% metaldehyde bait, Deadline 2% methiocarb + 2% metaldehyde bait (Pace National Corp., Kirkland, WA), Metaldehyde 4% bait, Bait 344 2% methiocarb bait, Metaldehyde 1% + 4% carbaryl bait, Lan Bait 1% methomyl (Soil Serv Inc., Salinas, CA), Lance 6% cloethocarb bait, Lance 480 g/L cloethocarb spray (BASF, Fairfield, NJ), Mesurol 75 WP methiocarb spray (Mobay Chemical Co., Kansas City, MO), Zectran 2% EC mexacarbate spray, Larvin 3% and Larvin 4% thiodicarb baits, UCES 1% and UCES 2% aldoxycarb baits (Union Carbide Co., Triangle Park, NC).

Test protocol employed glass terrariums (30.5 cm long × 20.5 cm wide and 30.5 cm deep) fitted with screen tops to prevent escapes. In an effort to simulate natural conditions, the bottom surface (620 cm^2) of each terrarium was divided by a 2.54 cm wood divider running the width of the terrarium and separated an area (413 cm^2) for spreading bait formulations or introducing treated plant materials and an area (ca 207 cm^2) for moist washed sand 2.54 cm deep. The moist sand provided humidity necessary to keep the snails active and also served as a refuge where snails could rest when not actively feeding. These techniques were similar to those described by Parrella et al. 1985 and Crowell 1977.

Each candidate molluscicide was applied to the treatment area at rates proportioned to label recommendations (Table 1). After treatment, 10 mature snails (four and one-half whorls) held from 24-48 h without food were placed centrally on the sandy area of each terrarium. Snails used in all tests were field collected from a population located in Santee, CA. There were five replications of each treatment. A control group was maintained in a simlar fashion. Controls were provided a 15 cm^2 leaf of lettuce, *Lactuca*, placed in the treatment area in tests comparing sprayable formulations. Controls were not fed in bait comparison tests. Observations of mortality, 5 and 12 days after treatment, entailed using a sharp probe to solicit muscular contraction. Lack of contraction indicated death.

Table 1. Efficacy of laboratory tests using commercial bait formulations against the white garden snail (five replicate treatments of 10 snails each).

Toxicant	No. Snails Exposed	kg AI/ha	% Mortality after 12 days*
Metaldehydes			
Bug Geta	50	1.59	32.0b
Slug and Snail	50	1.67	38.0b
Slug N Snail	50	1.10	30.0b
Deadline Bullets	50	1.79	70.0a
Deadline 40	50	1.54	46.0ab
Soil Serv Metaldehyde	50	2.24	44.0ab
Control	50	—	4.0c
Carbamates			
Slug Geta	50	0.98	64.0a
Slug and Snail Bait M	50	0.98	58.0ab
Bait 344	50	0.98	32.0bc
Zectran	50	0.67	54.0ab
Larvin 3%	50	1.45	16.0cd
Larvin 4%	50	1.96	16.0cd
UCES 1%	50	0.45	18.0cd
UCES 2%	50	0.90	12.0de
Lan Bait	50	0.56	16.0d
Lance	50	2.24	28.0cd
Control	50	—	0 e
Metaldehyde/carbamate combinations			
Soil Serv Metaldehyde + Carbaryl	50	2.80	18.0ab
Bug Geta Plus	50	1.70	40.0a
Deadline Bullets M and M	50	1.79	36.0a
Control	50	—	4.0b

* Means within test classes followed by the same letter are not significantly different ($P > 0.05$; Duncan's [1955] multiple range test).

Field Cage Tests

The most promising candidates from our laboratory screening tests were field tested in screen cages where the primary ground cover consisted of clover, *Trifolium*, with patches of bermuda grass, *Cynodon*, with a sandy soil type. The screen cages were 1 m^2 with 0.3 m high sides and arranged in a randomized block design with five replications including an untreated control. A rock salt barrier glued around the top of each cage prevented escape (Crowell 1977). Most treatments were applied at recommended field rates (Table 1). However, several sprayable formulations were applied at several rates. Twenty-five adult snails held for 48 h under moist conditions without food were topically marked with a fluorescent and introduced into each cage approximately 1 h before treatment. Treatment efficacy was determined by counting dead snails at 7 and 14 days after treatment. When treatments were applied twice, at 7- or 14-day intervals, additional mortality checks were made at 21 and 28 days after initial treatment.

The evening before treatment, each cage was irrigated with approximately 37.8 liters of water. No additional irrigations were made. Bait formulations were broadcast evenly through each cage on the damp soil. Sprayable formulations were applied

with a Model D-201, R and D Sprayer (R & D Sprayers, Inc. Opelousas, LA) which employed a single 01 flat fan nozzle operating at 30 psi. Pressure was obtained from a 58 × 15 cm CO_2 tank. All sprayable formulations were applied in 467 or 934 liters of water per hectare equivalent. A sheet-metal shield 0.9 × 0.9 m was placed around each cage during spray applications to minimize molluscicide drift between treatments.

Field Tests

Two bait materials from our laboratory and field cage studies were selected for further field tests in the front yard of a heavily infested residence located in Lakeside, CA. The treatment areas of approximately 645 m^2 was divided into eight blocks of approximately 80 m^2 each. The vegetation consisted predominately of two species of *Mesembryanthemum*, with patches of grass, *Cynodon*, and several citrus trees. Granular applications of Deadline Bullets (metaldehyde, 1.7 kg AI/ha) or Slug-Geta (Mesurol, 0.98 kg AI/ha) were broadcast over the plots. There were three replications of each treatment, with two replications for the control (due to limited space), arranged in a completely randomized design. Three m^2 sites in each plot were selected as survey areas. Snail counts were made one day before treatment and 7, 14, 21, 28, and 42 days after the initial treatment. Due to dry conditions and lack of snail activity, sprinkler irrigations were conducted three mornings a week for the duration of the test.

A second field test was initiated on two residential properties located in Oceanside, CA. The first property consisted of 1,828 m^2 on a hillside heavily infested with snails. The vegetation consisted of *Mesembryanthemum*, wild radish, *Raphanus*, anise, *Pimpinella*, and wild clover, *Trifolium*. For survey purposes, the hillside was divided into quadrants in which three 1-m^2 survey sites were set up in each quadrant. Deadline Bullets applied at 1.79 kg AI/ha were broadcast over the entire area with a Whirlybird spreader (Chevron-Ortho) with a 1 meter swath on 18 June and 9 July 1986. A second property of 210 m^2 was treated with Slug-Geta at 0.98 kg AI/ha following a similar protocol for treatment application and survey. The area was a vegetable garden consisting of tomatoes, *Lycopersicon*, two varities of *Cucurbita*, and carrots, *Daucus*.

An analysis of variance, randomized block design was employed to determine significant differences between test treatments. When significant differences were present, Duncan's Multiple Range Test (Duncan 1955) was employed to assign significant ranges for the treatment means. All analyzed data were transformed using a rank transformation as described by Conover and Iman (1981).

RESULTS

Lab Screening Tests

The Deadline Bullet formulation was the most efficacious of the six commercial bait formulations of metalydehyde examined with 70% mortality at 12 days postreatment (Table 1). Deadline Bullets caused significantly higher mortality in exposed WGS than did Bug-Geta, Slug and Snail, or Slug N Snail formulations. Furthermore, although not statistically significant, WGS mortality was 34.2 and 37.2% higher in cages treated with Deadline Bullets than in Deadline 40 or Soil Serv metaldehyde 4% bait treated cages, respectively.

Differences in mortality between the two Deadline formulations may in part be attributed to application techniques. Deadline 40, a heavy paste-like formulation, when applied to the small test area at a rate proportional to the recommended label rate (per ha) resulted in a single droplet ca. 0.85 cm^2 in size. The Deadline Bullet formulation was spread uniformly throughout the arena increasing the probability of contact by the snails.

Of 10 carbamate formulations examined, two formulations employing methiocarb as a toxin (Slug-Geta and Slug and Snail Bait M) and Zectran a 2% mexacarbate bait caused significantly higher mortality in WGS than did the other carbamate formulations examined (Table 1). Mortality in all treatments, except the aldoxycarb 2%, was significantly higher than the control. Mortality comparison of three bait formulations tested which combined metaldehyde with a carbamate did not differ significantly from each other and averaged 31 ± 12% mortality. It appeared that combining the two toxins caused no higher mortality than either toxin applied alone.

The efficacy of four sprayable molluscicides was highly variable and a dose/mortality correlation was not evident with either Mexacarbate 2 EC or Methiocarb 75 WP (Table 2). Generally, the three carbamate materials at the highest doses of 2.24 or 4.48 kg AI/ha killed significantly greater numbers of WGS than did the metaldehyde spray at 4.7 kg AI/ha. Furthermore, both mexacarbate and methiocarb appear more efficacious when used as baits than sprays under the conditions of our laboratory tests (Tables 1 and 2).

Table 2. Efficacy of sprayable molluscicides against white garden snails in laboratory tests (five replicate treatments of 10 snails each).

Toxicant	kg AI/ha	% Mortality after 12 days*
Slug N Snail Special Spray	4.7	10 de
Zectran 2EC†	0.56	16 bcde
	1.12	4 e
	2.24	35.4ab
	4.48	40 a
Mesurol 75 WP‡	0.42	24 abcd
	0.84	12 cde
	1.68	26 abcd
	2.24	10 de
	4.48	32 ab
Lance 480 g/L‡	2.24	28 abc
Control	—	4.0e

* Means followed by the same letter are not significantly different ($P > 0.05$; Duncan's [1955] multiple range tests).

† Applied in 935 liters water/hectare.

‡ Applied in 468 liters water/hectare.

We observed in the above laboratory tests that WGS intoxicated with a molluscicide would lie in an immobilized state from one to several days posttreatment before dying. In numerous cases we also observed WGS recovered from the toxic effect of the chemical before final mortality counts were made.

Field Cages

All baits tested had significantly higher WGS mortality than did the untreated control (Table 3, Test 1). However, Deadline Bullets caused a significantly greater (65%) mortality in WGS than Deadline 40, Slug-Geta, or Slug and Snail Bait M (16 - 38% mortality). In Test 2, cages treated with Deadline Bullets and Lance bait killed 50.4 and 47.0% of test snails, respectively, a significantly greater percentage than Lan Bait or Zectran bait (Table 3, Test 2). All treatments except Zectran bait had mortality rates significantly higher than the untreated control.

Table 3. Efficacy of selected molluscicide baits against white garden snail in small field cages (five replicate treatments of 25 snails each).

Toxicant	kg AI/ha	% Mortality after 14 days*
	Test 1	
Deadline Bullets	1.79	65.0a
Slug and Snail Bait M	0.98	16.0c
Slug Geta	0.98	38.0b
Deadline 40	1.54	29.6b
Control	—	0.8d
	Test 2	
Zectran	0.67	18.4bc
Lan Bait	0.56	30.3b
Deadline Bullets	1.79	50.4a
Lance	2.24	47.0a
Control	—	4.6c

* Means within test classes followed by the same letter are not significantly different ($P > 0.05$; Duncan's [1955] multiple range tests).

There was no statistically significant enhancement of mortality in Zectran 2 EC or Deadline Bullets when two applications were made at 10 day intervals compared to a single application of either treatment (Table 4, Test 1). However, numerically, WGS mortality was higher in cages receiving the two applications. In the second test, mortality assessments indicated that two applications of a Mesurol 75 WP formulation applied at 14 day intervals caused significantly (42%) higher mortality than a single Mesurol 75 WP application (Table 4, Test 2). Furthermore, WGS mortality with two applications of Mesurol 75 WP did not differ significantly from one or two applications of Lance.

Field Tests

Both Slug-Geta and Deadline Bullets reduced pretreatment WGS populations by 93 and 100%, respectively, after 42 days (Table 5, Test 1). However, untreated control plots showed a reduction of 38% during this same period. It was observed that our three weekly sprinkler irrigations to keep WGS from aestivation also caused considerable snail movement in and out of the control plots. This problem was magnified by the small size of our plots.

A second test was conducted by treating two residences which had similar pretreatment population densities of WGS. One property, randomly selected, was treated with Deadline Bullets, the other with Slug-Geta. After 42 days and two applications per treatment at 21 day intervals, pretreatment populations were reduced by 90.4% on the residence treated with Deadline Bullets and 70.6% on the residence treated with Slug-Geta.

Table 4. Efficacy of selected molluscicide formulations applied in a single application of double applications (five replicate treatments of 25 snails each).

Toxicant	No. applications	kg AI/ha	% mortality after indicated number of days* 7	14	21	28
		Test 1				
Zectran 2 EC	1	2.24	39.7a	57.8a	60.0a	—
Zectran 2 EC	2†	2.24	51.7a	61.0a	68.0a	—
Deadline Bullets	1	1.79	47.4a	50.3a	51.0a	—
Deadline Bullets	2†	1.79	56.3a	62.5a	78.0a	—
Control	—	—	0 b	0 b	0.8b	—
		Test 2				
Mesurol 75 WP	1	2.24	19.0bc	21.7a	29.6a	38.8b
Mesurol 75 WP	2‡	2.24	30.8ab	31.3a	48.8a	66.6a
Lance 480 g/L	1	2.24	45.2a	44.4a	48.1a	59.5ab
Lance 480 g/L	2‡	2.24	26.6ab	36.3a	43.6a	57.6ab
Control	—	—	3.4c	4.6b	4.7b	4.2c

* Means within test classes followed by the same letter are not significantly different ($P > 0.05$; Duncan's [1955] multiple range tests).
† The second application was applied 10 days after initial treatment.
‡ The seocnd application was applied 14 days after initial treatment.

Table 5. Efficacy of two applications of mesurol or metalydehyde bait treatments on white garden snail populations located on a residential property in San Diego, California.

Toxicant	No. juvenile and adult snails per m² at indicated days Pre-treatment 0	7	14	21	28	42	% reduction from pre-treatment counts*
Slug-geta†	37.0	5.2	1.2	2.3	2.5	2.6	93.0a
Deadline Bullets†	7.9	2.5	1.2	0.7	0.3	0	100.0a
Control	19.8	9.5	14.3	7.5	5.0	12.2	38.4b

* Percentages in last column followed by the same letter are not significantly different ($P > 0.05$; Duncan's [1955] multiple range tests).
† Second application applied on day 14.

DISCUSSION

Of the 19 commercial bait molluscicides tested, Deadline Bullets, a 4% metaldehyde bait, and Slug-Geta, a 2% methiocarb bait, were the most effective against WGS. Godan (1983) and others have suggested the metaldehyde declines in efficiency at high humidities and is less effective than carbamates such as methiocarb when moist high humidity conditions prevail. However, in our field tests where plots received regular sprinkler irrigations (from 1.5 - 3.55 cm water/week), metaldehyde dispensed in the Deadline Bullet formulation performed as well as or better than methiocarb granular formulation dispensed in the Slug-Geta

formulation. Apparently, the loss of toxicity and attractiveness of metaldehyde baits after exposure to enviornmental conditions such as sunlight, soil pH, and moisture has been eliminated to some degree in this formulation.

Metaldehyde's attractiveness to snails has made it an ideal chemical to combine with other molluscicides in attempts to increase the efficacy of granular bait formulations. However, data obtained in our lab tests showed no increase in efficacy against WGS when metaldehyde was combined with several different carbamates in several different bait formulations. In most cases the carbamate or metaldehyde was more effective in killing WGS alone than when combined.

The results of field trials illustrate the fact that WGS populations can be controlled with currently available molluscicides. However, San Diego County as a whole consists of a diversified environment requiring more than a single treatment strategy to obtain control or possible eradication of WGS populations over a sustained period of treatment.

In residential areas where irrigation is a common practice through the dry summer months, Deadline Bullets or Slug-Geta baits could be used year round. In nonirrigated areas, treatments should be initiated immediately following the first substantial winter rain in an effort to eliminate or reduce populations before they mate and lay eggs. During the activity cycle of WGS, it may be necessary to apply multiple bait or spray applications to effect control measures. Treatment interval and number of applications will depend on prevailing environmental conditions and population densities. When foliar treatments are necessary in nonresidential areas, Mesurol 75 WP applied in 468 liters of water/ha could be used. Zectran 2 EC, another material that was effective against WGS, has since been removed from the market.

In addition to chemical control strategies, Basinger (1927) implemented a successful eradication campaign in La Jolla, CA (in the 1920s), employing the following tactics: 1) burning, 2) hand picking snails in environmentally sensitive areas, and 3) vegetation removal in areas where burning was impractical. It is likely that all these strategies would be necessary to launch a successful control or eradication program against the current WGS infestation. Additional studies on the biology and population dynamics of WGS in California are needed to enhance current treatment methodology.

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SAMPLING METHODS FOR ESTIMATING POPULATION DENSITIES OF PLANTHOPPERS AND PREDATORS IN DIRECT-SEEDED AND TRANSPLANTED RICE[1]

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Abstract: Two sampling methods (a water pan and carbon dioxide sampler) were compared for estimating populatings of brown planthopper, *Nilaparvata lugens* (Stal) (Homoptera: Delphacidae), white-backed planthopper, *Sogatella furcifera* (Horvath) (Homoptera: Delphacidae), a predatory mirid bug, *Cyrtorhinus lividipennis* (Hemiptera: Miridae) and spiders in direct-seeded and transplanted rice in the Philippines. Similar population trends were observed in direct-seeded and transplanted rice using both methods although a significantly higher number of spiders ($p < 0.05$) was sampled using the carbon dioxide sampler, especially in direct-seeded rice. The pan sampler however, required less time and was less expensive than the carbon dioxide sampler.

Key Words: Rice, planthoppers, brown planthopper, *Nilaparvata lugens* white-backed planthopper, *Sogatella furcifera* mirid bug, *Cyrtorhinus lividipennis*, spider, water pan sampler, carbon dioxide sampler.

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A carbon dioxide (CO_2NE) sampler was found useful in estimating densities of rice arthropods (Aquino and Heinrichs 1986; Shepard et al. 1985). To use this device, a cone-shaped enclosure was placed over each rice hill to prevent mobile arthropods from excaping, then carbon dioxide was discharged to the enclosure. After a few minutes, the arthropods were removed from the water's surface with a small screen scoop. A detailed discription of this sampler was published by Aquino and Heinrichs (1986).

The D-vac and FARMCOP are suction type samplers (Dietrick 1961; Carino et al. 1979; Perfect et al. 1983). Comparison of the CO_2NE sampler with FARMCOP revealed that in general, FARMCOP yielded higher means of most arthropods except for spiders (Shepard et al. 1985) but coefficients of variation were approximately the same for the two techniques. The CO_2NE sampler was more practical because it was inexpensive and easier to use.

The above methods are useful for research but not practical for making insect management decisions. Thus, there is a need for a simpler but reliable sampling method in direct-seeded and transplanted rice.

Boards (18 × 25 cm) covered with sticky material have been used in sampling brown planthopper (BPH), *Nilaparvata lugens* (Stal) for pesticide tests in Japan (Nagata and Masuda 1978) and modified for a surveillance program in Malaysia (Ooi 1982). The sampler holds the sticky board horizontally to one side of the rice hill and the hill was struck to dislodge the arthropods onto the sticky surface of the boards. The advantage of this method was that after the insects were collected on the sticky boards, they were carried to

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the laboratory for counting and identification, or samples can be stored in the refrigerator for processing later. The major disadvantages were that the sticky material was messy and expensive.

The objective of this study was to develop an efficient and reliable method for practical use in surveillance programs, and compare the population estimates obtained with those using an absolute sampling device (CO_2NE sampler) for sampling BPH and white-backed planthopper (WBPH), *Sogatella furcifera* (Horvath), and major predators (spiders and the mirid bug, *Cyrtorhinus lividipennis* (Reuter)) in rice.

MATERIALS AND METHODS

This study was conducted in Victoria, Laguna and at the International Rice Research Institute (IRRI) Farm, Los Banos, Laguna, Philippines during the wet season (June to October) 1985.

A yellow plastic pan (measuring 25 cm dia at the top × 15 cm at the bottom and 6.5 cm high) was used to collect the samples. About 100 ml of water was added to the pan along with 3-5 drops of liquid detergent (Teepol®) to reduce the surface tension of the water so that small arthropods would sink. The pan was then placed at the base of the hill and the hill was struck three times to dislodge the arthropods onto the pan.

A comparison between CO_2NE sampler and the pan was made in fields measuring approximately 1,200 m^2 planted to IR1917-3-17 at 25 × 25 cm spacings. In each plot, 32 random samples were taken by each method weekly for 11 weeks.

At IRRI Farm, comparison was made between the two sampling methods in 1,200 m^2 plots which were established by direct-seeding and transplanting. The sampling unit was the hill for transplanted rice and 25 cm^2 area for direct-seeded rice. The area was delineated by using a wire frame which approximated the number of tillers in a hill at seeding rates used in the study.

Arthropods collected were placed in glass vials containing 70% ethyl alcohol and taken to the laboratory for sorting and identification. Average weekly densities were estimated and correlation coefficients and coefficients of variability were compared. Tests of homogeniety of correlation coefficients were conducted and Student's T-test ($p < 0.05$) was used to determine significant differences between means.

RESULTS AND DISCUSSION

The predominant arthropod populations during the season (June to October, 1985) in Victoria, Laguna and at IRRI Farm were BPH, WBPH, *C. lividipennis* and spiders. Because BPH and WBPH occur together in mixed colonies on the rice plant, we combined the counts of the adults of these species. Nymphal forms of both species were categorized as delphacid nymphs. Likewise, all spider species were combined but they were predominantly *Atypena* (*Callitrichia*) *formosana* (Ooi) and *Lycosa pseudoannulata* (Boesenberg and Strand).

Comparison between seasonal mean (± S.E.) numbers of arthropods per hill estimated by water pan and CO_2NE sampling devices in transplanted rice in

Victoria and at IRRI Farm are presented in Figs. 1 and 2. In both locations, the same population trends were observed for BPH + WBPH adults, delphacid nymphs, *C. lividipennis* and spiders. In Victoria, for BPH + WBPH adults, delphacid nymphs and *C. lividipennis* the average number of insects caught per sampling occasion was about he same using CO_2NE and water pan. For spiders however, in almost all occasions, significantly higher population density was estimated using CO_2NE. At IRRI Farm, there were no significant differences between etimates of population densities using both devices for most of the arthropods sampled except for *C. lividipennis*. For this predator, higher numbers were estimated using the water pan when populations were denser (at 43-50 days after transplanting).

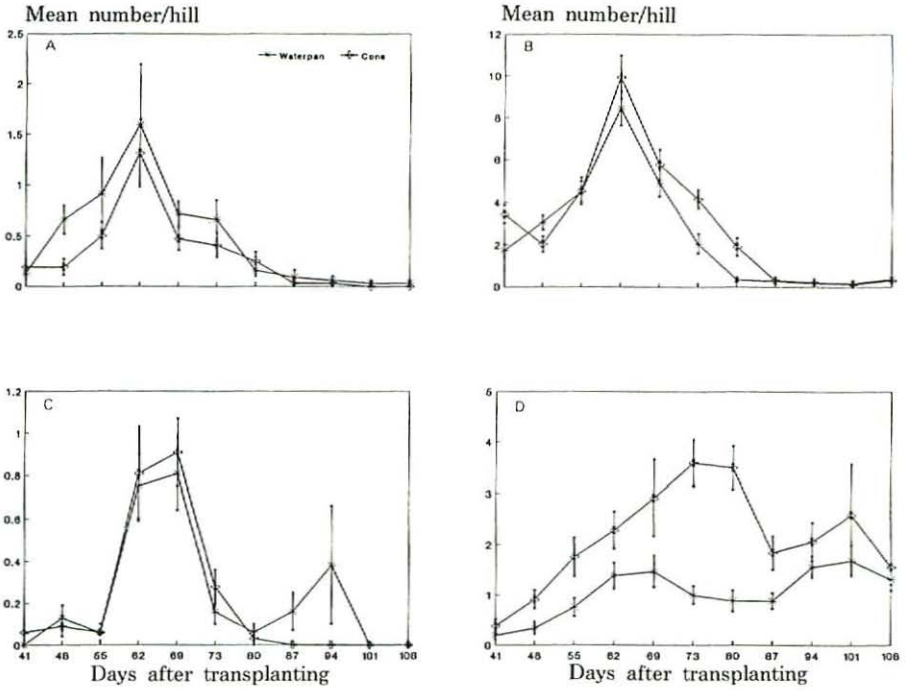

Fig. 1. Seasonal populations of brown- and white-backed planthoppers (BPH + WBPH) adults (A), delphacid nymphs (B), *C. lividipennis* (C) and spiders (D) sampled by water pan and CO_2NE devices in transplanted rice. Victoria, Laguna, Philippines, 1985.

For direct-seeded rice (Fig. 3), the same population trends were observed using both sampling devices. On most occasions, no significant differences between estimates of means were observed for BPH + WBPH adults, delphacid nymphs and *C. lividipennis*. Significantly higher densities of spiders were estimated using CO_2NE sampler.

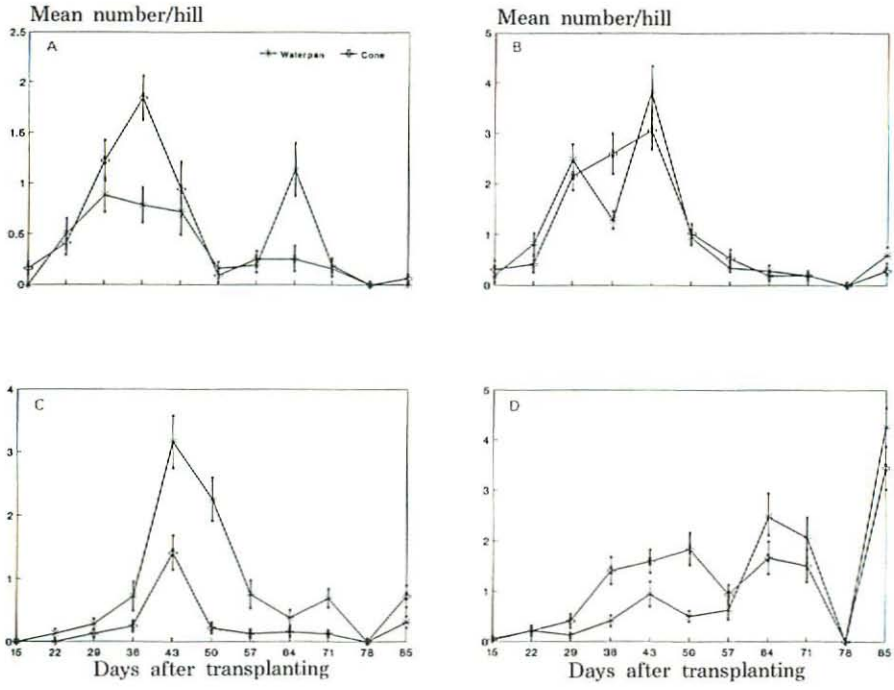

Fig. 2. Seasonal populations of brown- and white-baked planthoppers (BPH + WBPH) adults (A), delphacid nymphs (B), *C. lividipennis* (C) and spiders (D) sampled by water pan and CO_2NE devices in transplanted rice. IRRI Farm, Los Banos, Laguna, Philippines, 1985.

In general, the two techniques were highly correlated (Table 1). At Victoria, the correlation coefficients for BPH + WBPH adults, delphacid nymphs and *C. lividipennis* were similar (91-94) regardless of the sampling devise used as determined by a test of homogeniety. In general, correlation coefficients were lower for spiders and the CO_2NE sampler yielded higher estimates of spider populations. At IRRI Farm, high correlation coefficients were observed for delphacid nymphs, *C. lividipennis*, and spiders while they were relatively lower for BPH + WBPH adults.

Comparisons of coefficient of variability (CV) were similar for all arthropods regardless of the sampling technique used both in Victoria and at IRRI Farm (Table 2). CVs were high at both locations (123-313). This is likely due to the low arthropod population densities during the season.

CO_2NE sampling can only be carried out in flooded fields but sampling by the water pan can be made even in dry fields. Also, while CO_2NE sampling requires that arthropods be transferred to vials and counted later, counting can be carried out more easily in the field using water pan. Considering the ease of use and favorable comparison of population estimates with the CO_2NE sampler, the water pan would be more practical in an insect pest management program for rice.

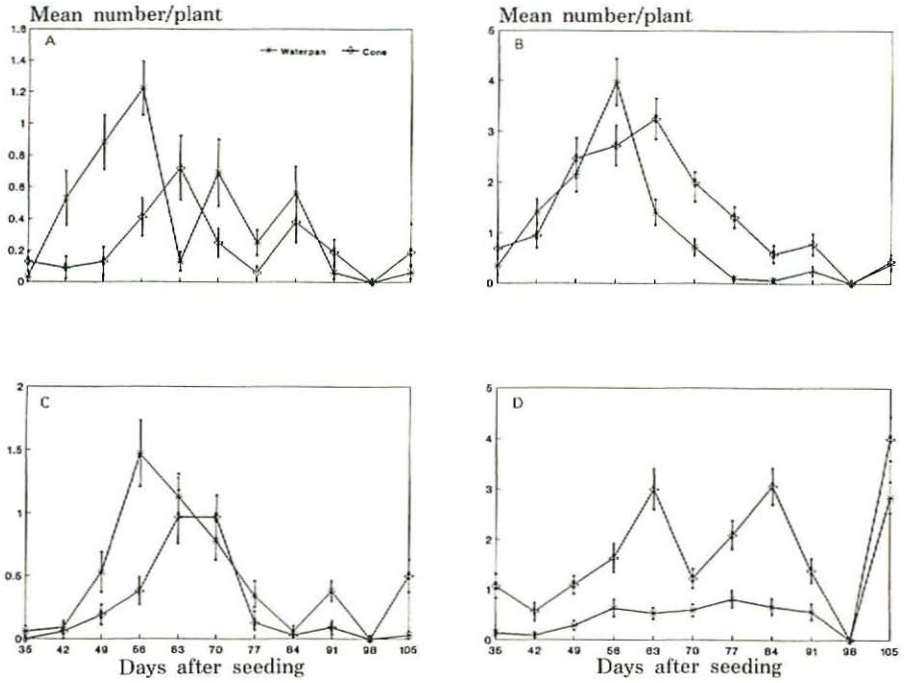

Fig. 3. Seasonal populations of brown- and white-backed planthoppers (BPH + WBPH) adults (A), delphacid nymphs (B), *C. lividipennis* (C) and spiders (D) sampled by water pan and CO_2NE devices in direct-seeded rice. IRRI Farm, Los Banos, Laguna, Philippines, 1985.

Table 1. Correlation coefficients between CO_2NE and water pan sampler. Wet season (June-October), Victoria, Laguna and IRRI Farm, Philippines, 1985.

Arthropods sampled	Victoria	IRRI* Farm
BPH + WBPH adults	0.94	0.65
Delphacid nymphs	0.93	0.90
C. lividipennis	0.91	0.74
Spiders	0.56	0.84

* The International Rice Research Institute.

Table 2. Coefficients of variability of CO_2NE and water pan sampler. Wet Season (June-October), Victoria, Laguna and IRRI Farm, Philippines, 1985.

	Victoria		IRRI* Farm	
Arthropods sampled	Water pan	CO_2NE	Water pan	CO_2NE
BPH + WBPH adults	297	258	213	197
Delphacid nymphs	142	134	168	165
C. lividipennis	313	290	189	261
Spiders	124	136	170	144

* The International Rice Research Institute.

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NOTE

MOVEMENT OF A WEEVIL PARASITOID, *ANISOPTEROMALUS CALANDRAE* (HOWARD)[1], WITHIN A COLUMN OF WHEAT IN RELATION TO HOST LOCATION

Key Words: *Anisopteromalus calandrae*, *Sitophilus oryzae*, dispersal, wheat, biological control.

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The rice weevil, *Sitophilus oryzae* (L.), is one of the most damaging cosmopolitan pests of stored grains, such as wheat, corn and rice. Unlike most stored-produce moth species that infest commodities on or near the surface, *S. oryzae* tends to remain where they are introduced until excess heat causes them to disperse (Birch, L. C., J. Austr. Inst. Agric. Sci. 12: 27-31, 1946). Howe demonstrated that *S. oryzae* moved downward within 132 × 10.2 cm columns of grain under a variety of conditions (Howe, R. W., Bull. Entomol. Res. 42: 125-35, 1951). Thus, the entire bulk may be infested when temperature and moisture conditions are favorable.

Some of the major parasitoids of *S. oryzae* are *Anisopteromalus calandrae* (Howard), *Choetospila elegans* Westwood, *Dibrachys cavus* (Walker) and *Lariophagus distinguendus* (Foerster) (Sinha, R. N. and F. L. Watters, agriculture Canada, 290 pp, 1985). *Anisopteromalus calandrae* is distributed worldwide and is a very common ectoparasitoid of the larvae and pupae of many coleopterous insects associated with stored foods (Ghani, M. A. and H. L. Sweetman, Biologia (Lahore) 1: 115-139, 1955). Laboratory tests have shown that *A. calandrae* has potential for weevil control (Cline, L. D., J. W. Press, and B. R. Flaherty, J. Econ. Entomol. 78: 835-838, 1985) (Press, J. W., L. D. Cline, and B. R. Flaherty, J. Georgia Entomol. Soc. 19: 110-113, 1984). We, therefore, selected *A. calandrae* as our test insect.

Effective suppression of an *S. oryzae* population by *A. calandrae* depends primarily on the parasitoid's ability to locate its host within a grain mass. It was demonstrated in laboratory studies that female *A. calandrae* released in 76.2 cm columns of grain moved in all directions (Ghani, M. A. and H. L. Sweetman, Biologia (Lahore) 1: 115-139, 1955). The objective of our study was to determine the movement of *A. calandrae* within 2.2 m columns of wheat in relation to the presence or absence of hosts above or below the point of introduction or at the point of introduction.

Laboratory cultures of *S. oryzae* (23-d-old) reared on ca. 300 g of soft red winter wheat were used in this study. The cultures contained an average of 1611.48 *S. oryzae* larvae each. Four of these cultures were thoroughly mixed, then divided into four equal lots to insure that a uniform number of larvae would be available for each test condition. These four infested lots were placed individually into wide mouth 0.95 l jars to a depth of ca. 7 cm. Noninfested wheat was added to completely fill the jars. Three 1.9 m × 7.5 cm acrylic cylinders were fitted with jar rings at each end so that the 0.95 l jars containing the *S. oryzae* infested grain could be attached to them. The three cylinders were then filled with noninfested soft red winter wheat. Two of the 0.95 l jars containing the *S. oryzae* infested grain were attached to both ends of the first cylinder. This cylinder was secured perpendicularly to make a column of wheat ca. 2.2 m high. The other two cylinders were set up in the same manner with the exception that the second cylinder had

[1] HYMENOPTERA: Pteromalidae. Accepted for publication 16 May 1988.

the jar of infested wheat attached only to the top of the wheat column while the third cylinder had the jar of infested wheat attached only to the bottom of the wheat column. Jars that contained noninfested wheat were attached to the bottom of the second wheat column and to the top of the third wheat column (Fig. 1). Three 1.5 cm holes had been drilled into each column (ca. 2.5 cm from the top and bottom, respectively, and in the middle) to allow for the introduction of the *A. calandrae*. Rubber stoppers were used to seal these holes. A wire 14 × 14 mesh screen had been inserted into the base of each jar ring at the cylinder ends prior to filling in order to separate the wheat in the jars from the wheat in the cylinders while still allowing free passage by the parasitoids.

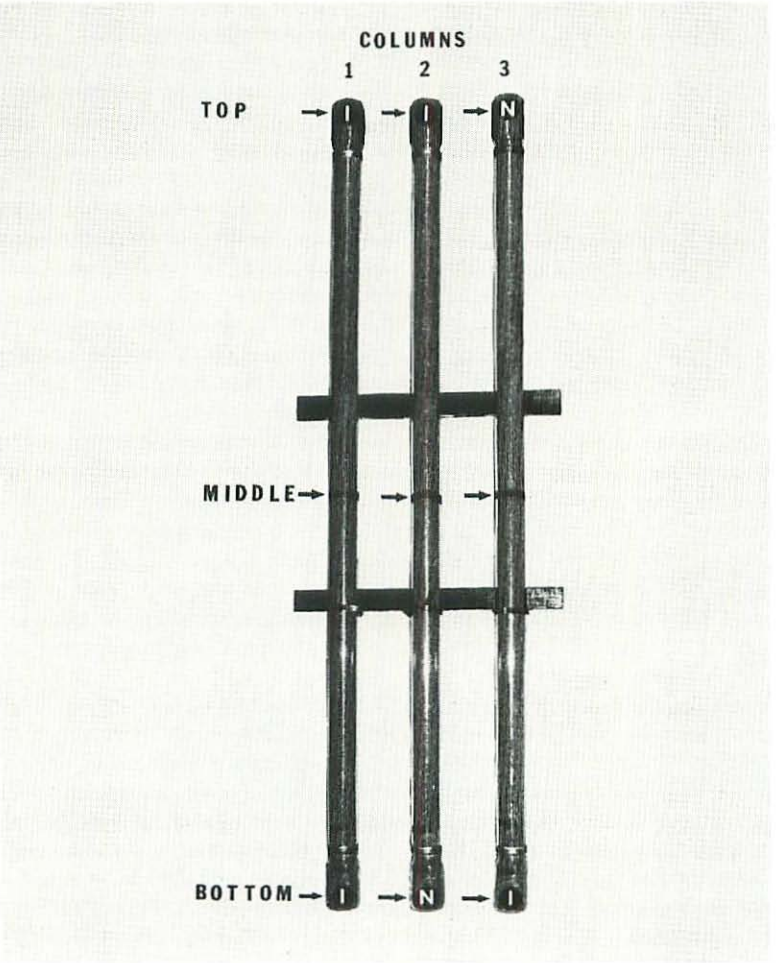

Fig. 1. Columns of wheat (2.2 m) with attached 0.95 l jars containing *S. oryzae* infested wheat (I) or noninfested wheat (N). Arrows indicate points of *A. calandrae* introduction at the top (first test), middle (second test) and bottom (third test).

In the first sequence of tests, ten 24-h-old adult male and ten 24-h-old adult female *A. calandrae* from laboratory cultures were introduced into the top of the three wheat columns. In a second sequence of tests the *A. calandrae* were introduced only into the middle of the three wheat columns. In a third sequence of tests, the *A. calandrae* were introduced only into the bottom of the three wheat columns. Each test sequence was replicated 10 times. Infested grain was removed from each wheat column after 7 d, then incubated for 14 more d, after which the F_1 *A. calandrae* and the P_1 *S. oryzae* adults were tabulated. All tests were conducted at $30 \pm 1°C$ and $60 \pm 5\%$ RH under an alternating 12L:12D cycle. These data were analyzed for significance at the 0.05 probability level using ANOVA and Duncan's multiple range test or by standard error of the means.

When *A. calandrae* was introduced at the top of wheat columns in which *S. oryzae* were present at both top and bottom, ca. 80% of the parasitization occurred at the top compared with ca. 20% at the bottom of the wheat columns (Table 1). Since one *A. calandrae* normally emerges from a single *S. oryzae* host and superparasitism by this wasp is rare when an ample supply of hosts ($\bar{x}$ in infested wheat sites = 1611) is present, it was assumed that each adult *A. calandrae* equalled one parasitized *S oryzae* (Ghani, M. A. and H. L. Sweetman, Biologia (Lahore) 1: 115-139, 1955). When *A. calandrae* were introduced into the top of grain columns where infested wheat occurred only at the top of the columns, ca. 6 times as many *S. oryzae* were parasitized than when the infested wheat occurred only at the bottom of the wheat columns (Table 1). This indicated that, irrespective of the location of the infested wheat, *A. calandrae* females did not penetrate well. The few parasitoids that did penetrate, however, demonstrated that the wheat did not constitute a barrier to the wasps.

Table 1. Mean ($\pm$ SE) number of *Anisopteromalus calandrae* recovered from infested wheat located at the top (7 cm) or bottom (7 cm) of 2.2 m columns of wheat.*†.

Area in Column of Parasitoid Introduction‡	Column 1		Column 2	Column 3
	Top Infested	Bottom Infested	Top Infested	Bottom Infested
Top	865.3 ± 71.0ab	178.4 ± 61.4h	958.4 ± 62.8a	162.8 ± 57.3hi
Middle	570.0 ± 126.2cdef	139.6 ± 56.2hij	610.9 ± 136.6bcde	112.7 ± 52.8hij
Bottom	415.1 ± 83.2efg	832.3 ± 69.2abc	503.4 ± 72.4efg	805.8 ± 74.8abcd

* × 10 replications.

† All means (rows and columns) followed by the same letter(s) are not significantly different ($P > 0.05$; Duncan's multiple range test).

‡ 10 pr (male and female) *A. calandrae*.

Introduction of *A. calandrae* into the middle of the wheat columns provided results similar to those with top introductions. Incidence of parasitism was greater at the top of the column when hosts were available at both the top and bottom. When only the top of the column contained infested wheat, much greater numbers of *S. oryzae* were parasitized than when wheat was infested only at the bottom of the columns.

When parasitoids were introduced at the bottom of the columns and hosts were available at both the top and the bottom, about twice as many *S. oryzae* were parasitized at the bottom near the point of introduction. In the separate grain columns having either the top or bottom infested, the top infestations had fewer weevils parasitized than in the bottom infestations.

Any shortage of hosts was unlikely to be a factor influencing the movement of *A. calandrae* since between 610.9 to 1779.8 *S. oryzae* adults emerged from each infested grain location (Table 2). The lower numbers of emerged *S. oryzae* usually occurred at locations where *A. calandrae* parasitization was the highest.

Table 2. Mean (± SE) *Sitophilus oryzae* recovered after exposure to *Anisopteromalus calandrae* from infested wheat located at the top (7 cm) or bottom (7 cm) of 2.2 m columns of wheat.*

Area in Column of Parasitoid Introduction†	Column 1		Column 2	Column 3
	Top Infested	Bottom Infested	Top Infested	Bottom Infested
Top	801.5 ± 61.5	1463.1 ± 105.1	705.6 ± 93.0	1488.7 ± 113.3
Middle	1003.3 ± 152.8	1604.9 ± 120.0	1211.6 ± 150.8	1779.8 ± 139.1
Bottom	1029.2 ± 157.0	610.4 ± 90.4	856.5 ± 133.0	633.1 ± 88.6

* Each experiment was replicated 10 times.
† 10 pr (male and female) *A. calandrae*.

Our data suggest that *A. calandrae* moved upward within a grain mass. When *A. calandrae* were introduced near the surface, most did not move downward, although the small percentage that did could conceivably produce enough progeny after a period of time for weevil suppression. If hosts were present at the point of *A. calandrae* introduction, substantial numbers remained among the infested wheat kernels to effect adequate parasitization rather than moving upward.

Our data indicated that downward penetration was relatively poor and better suppression of weevils would be likely if introductions of this parasitoid into commercial storages were made at many depths. If *A. calandrae* were released only at the top of a grain mass, substantially larger numbers of the parasitoid would be necessary to obtain adequate suppression since only a few tend to move downward and even these parasitoids may not penetrate to deep infestations. Conversely, if only bottom introductions of the parasitoid were feasible, then better overall rates of parasitization would result since many parasitoids would stop their upward movement when infested grain was encountered while others would continue upward.

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EFFECT OF CHLORPYRIFOS 50W ON FRUIT FINISH AND PACKOUT OF 'GOLDEN DELICIOUS'[1]

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Abstract: A full season spray program of chlorpyrifos compared to azinphosmethyl applied to 'Starkspur Golden Delicious' reduced extra fancy/fancy packout by 54% and reduced crop value because of fruit russeting by $967/ha. Substituting azinphosmethyl for chlorpyrifos during the petal fall, first cover and second cover applications reduced russeting and increased crop value to levels similar to the season long program of azinphosmethyl. Of the three applications, the substitution at first cover accounted for most of the reduction in russeting. Four applications of GA_{4+7}, 7 days apart, significantly improved the extra fancy/fancy packout of chlorpyrifos treated 'Smoothee' fruit.

Key Words: Chlorpyrifos, GA_{4+7}, russeting, economic losses, apples.

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Chlorpyrifos 50W was introduced as a broad spectrum insecticide for use on apple in 1983. Numerous studies have been conducted to evaluate the performance of this organophosphate against both fruit and foliage feeding pests of apple (Forsythe 1987; Hamilton et al. 1986; Hogmire and Crim 1986, 1987; Horsburgh and Cobb 1986; Hull 1987; Reissig et al. 1986). Chlorpyrifos has fit well into apple IPM programs because of its safety to mite predators (Anon. 1985; Howitt and Hays 1986; Hull and Baldwin 1982).

Season-long spray programs of chlorpyrifos have resulted in increased fruit russeting of 'Golden Delicious' (Hamilton et al. 1986; Horsburgh and Cobb 1986; Hull 1987; Weires 1987). Although not statistically significant, daytime applications of chlorpyrifos appeared to cause more russeting of 'Golden Delicious' than applications at night (Hogmire and Crim 1987).

This study was conducted in 1987 to determine: 1) the time during the season when chlorpyrifos was causing fruit russeting of 'Golden Delicious', 2) the economic impact of the russeting, and 3) if the russeting could be reduced.

MATERIALS AND METHODS

Two experiments were conducted at the West Virginia University Plant Science Experiment Farm in Kearneysville, WV. The first experiment was conducted in a block of 23 yr-old 'Starkspur Golden Delicious' on MM 106 rootstock with a tree spacing of 3.0 × 6.1 m. The average tree canopy measured 4.3 m in height and 4 m in width. The block was divided into six unreplicated plots of 0.16 - 0.22 ha in size. The block was relatively uniform in elevation, except for a lower region in the extreme western end which was not used in the study. Chlorpyrifos and azinphosmethyl (standard) were each applied in a full season program beginning at

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petal fall. The remaining four plots consisted of various chlorpyrifos treatment schedules with azinphosmethyl substituted at specific times during the early season (Table 1). Rates of application (g[AI]/ha) were 1120 and 840 for chlorpyrifos and azinphosmethyl, respectively. Treatments were applied as complete sprays (both sides of trees) with a Swanson DA500A airblast sprayer which traveled at 3.86 km/h and delivered 935 liters/ha of finished spray. Dates of application were 6 May [petal fall (PF)], 21 May [first cover (1C)], 5 Jun [second cover (2C)], 19 Jun [third cover (3C)], 3 Jul [fourth cover (4C)], 17 Jul [fifth cover (5C)], 31 Jul [sixth cover (6C)], 13 Aug [seventh cover (7C)], and 27 Aug [eighth cover (8C)]. Methomyl was applied across all plots at petal fall and seventh cover for the control of white apple leafhopper and spotted tentiform leafminer. Fungicides applied as tank mixes to all treatments were metiram (PF-4C), Dikar (5C-7C), benomyl (8C) and captan (8C).

All fruit were harvested on 21 Sep from each of 5 single-tree replications which were selected at random from the center row(s) of each plot. Treatments were evaluated by examining a 100 fruit sample which was selected blindly from the entire fruit harvest per tree. Fruit were rated for russeting using the following scale modified after Byers et al. (1983): 0 = no russeting or enlarged lenticels; 1 = no russeting but lenticels enlarged, rough and raised to touch; 2 = 1-10% surface russeted; 3 = 11-25% surface russeted; 4 = 26-50% surface russeted; and 5 = >50% surface russeted. Fruit were categorized into U.S. grade classifications (USDA 1972) based on the russet rating as follows: 0-2 = extra fancy/fancy; 3 = U.S. no. 1; and 4-5 = utility. The crop value was calculated for each treatment using an average West Virginia yield of 1236 bu/ha and an average price of $8.00, $5.00 and $4.00/bu for extra fancy/fancy combination, U.S. no. 1 and utility grade, respectively. Data are presented as means with standard error to show variability.

A second experiment was conducted in a 0.37 ha block of 5 yr-old 'Smoothee' (a russet resistant mutation of 'Golden Delicious') on MM 106 rootstock with a tree spacing of 6.1 × 6.1 m. The average tree canopy measured 4.5 m in height and 3.2 m in width. Three treatments, consisting of chlorpyrifos applied alone and in combination with two spray schedules of GA_{4+7} (gibberellic acid), were each applied to 4 single-tree replications in a complete randomized block design. GA_{4+7} has been shown to reduce russeting in 'Golden Delicious' (Wertheim 1982; Steenkamp et al. 1984) and is labeled for this use. It was used in this test to determine its effectiveness in suppressing russeting of fruit treated with chlorpyrifos. Treated trees were separated by two unsprayed rows and at least one unsprayed tree within treatment rows. Chlorpyrifos was used in four applications (PF-3C) in all treatments at the same rate as in the first experiment. In a second treatment, GA_{4+7} (15.3 g [AI]/ha) was applied in four sprays on a 7-day interval as specified in product literature. In the first (PF) and third (1C) application, GA_{4+7} was applied in combination with chlorpyrifos, whereas it was used alone in the second and fourth application. Since the chlorpyrifos applications were scheduled on a 14-day interval, which is standard for insecticides, the 7-day interval for GA_{4+7} necessitated two extra trips through the orchard. As this would present some inconvenience and additional expense for growers, a third treatment was included whereby GA_{4+7} was applied on a 14-day schedule in combination with chlorpyrifos in each of the 4 applications. Azinphosmethyl was applied to all treatments for the second half of the season beginning at fourth cover. Fungicides, as specified in the first experiment, were applied separately to all treatments. Application method and dates were the same as in the first experiment, with the addition of 13 and 28 May for the second and fourth application of GA_{4+7} in the 7-day schedule. All fruit were harvested from each replication on 23 Sep, with treatments evaluated in the same manner as in experiment one. Data were subjected to Duncan's (1951) multiple range test to determine differences between treatment means.

RESULTS AND DISCUSSION

A full season spray program of chlorpyrifos caused more fruit russeting than a comparable azinphosmethyl program, resulting in a 54% reduction of extra fancy/fancy fruit and an estimated decrease in crop value of $967/ha (Table 1). This was accompanied by a three-fold increase of utility grade apples under the chlorpyrifos program. Chlorpyrifos treated fruit had a higher incidence of solid versus net russeting compared to azinphosmethyl. The high susceptibility of 'Starkspur Golden Delicious' to russeting (Cummins 1977) undoubtedly contributed to the poor overall packout in this study.

Substituting azinphosmethyl for chlorpyrifos during the first three applications (PF-2C) reduced russeting and increased crop value to levels similar to the season long program of azinphosmethyl. Of the three applications, the substitution at first cover accounted for most of the reduction in russeting.

During an 8-year study under natural orchard conditions, Creasy (1980) discovered an excellent correlation between russet severity on 'Golden Delicious' and humidity during 16-20 days after full bloom. The first cover application occurred 24 days after full bloom, compared to 9 and 39 days for the petal fall and second cover applications, respectively. It would appear that chlorpyrifos was enhancing the expression of weather-induced russeting, which was reduced by substituting azinphosmethyl during the most susceptible period at first cover. It would also appear that chlorpyrifos may be causing some russeting after second cover, as substitution of azinphosmethyl in the first 3 applications still resulted in a higher percentage of utility grade apples compared to the full season azinphosmethyl program.

Chlorpyrifos is not the only pesticide currently used on apples which may have a detrimental effect on the fruit finish of 'Golden Delicious'. Russeting has also been demonstrated with dodine (Hatch 1975), a fungicide which is used for early season control of apple scab. Creasy and Swartz (1981) found that both oil and diazinon resulted in increased russeting of 'Golden Delicious' following a single application 18 days after full bloom.

Four applications of GA_{4+7}, scheduled 7 days apart, significantly reduced the russeting by chlorpyrifos and increased the packout of extra fancy/fancy fruit by 68% as compared to chlorpyrifos applied alone (Table 2). Although the 14-day schedule of GA_{4+7} also improved fruit finish and packout, it was not significantly better than chlorpyrifos applied alone. The 7-day application schedule of GA_{4+7} increased the crop value of chlorpyrifos treated fruit by $924/ha. Subtracting the cost of the GA_{4+7} and the two extra trips through the orchard necessitated by the 7-day spray interval would result in a benefit of approximately $625/ha.

The decision to use GA_{4+7} will have to be made on an orchard block by block basis with consideration given to the packout percentage needed to justify packing the fruit versus sending the fruit to the processor. For example, an increase in extra fancy/fancy packout from 34 to 58%, as occurred in this study with the use of GA_{4+7}, would not be sufficient to justify packing the fruit if the grower needed 75% packout to break even. In less severely russeted blocks, GA_{4+7} could provide enough benefit to make packing profitable.

Without the aid of a russet suppressant, growers should probably refrain from using chlorpyrifos (1120 g[AI]/ha) at least during the first 25 days, and preferably the first 40 days, after full bloom on 'Golden Delicious' if the crop is to be marketed as fresh fruit. Chlorpyrifos could be used without restriction in orchard blocks of 'Golden Delicious' grown for processing since russeting does not reduce fruit value for this market.

Table 1. Effect of chlorpyrifos 50W application timing on fruit finish and packout of "Starkspur Golden Delicious" in 1987.

Treatment	Time of application	Russet rating*	Distribution of fruit in US grades (%)*			Crop value ($ per hectare*)
			extra fancy/fancy	no. 1	utility	
Azinphosmethyl 50W	PF-8C	2.73 ± 0.13	34.6 ± 7.4	54.4 ± 3.5	11.0 ± 4.2	7327 ± 325
Chlorpyrifos 50W	PF-8C	3.31 ± 0.06	16.0 ± 3.5	50.6 ± 4.7	33.4 ± 3.3	6360 ± 74
Azinphosmethyl 50W	PF	3.15 ± 0.05	15.8 ± 2.2	55.8 ± 2.7	28.4 ± 2.8	6415 ± 103
Chlorpyrifos 50W	1C-8C					
Azinphosmethyl 50W	1C	3.02 ± 0.06	22.4 ± 3.8	54.8 ± 2.6	22.8 ± 1.5	6729 ± 159
Chlorpyrifos 50W	PF, 2C-8C					
Azinphosmethyl 50W	2C	3.10 ± 0.09	17.4 ± 3.4	58.0 ± 7.6	24.6 ± 7.3	6521 ± 108
Chlorpyrifos 50W	PF, 1C, 3C-8C					
Azinphosmethyl 50W	PF-2C	2.91 ± 0.09	25.2 ± 6.2	54.6 ± 5.9	20.2 ± 3.3	6865 ± 246
Chlorpyrifos 50W	3C-8C					

* $\bar{x}$ ± SEM.

Table 2. Effect of GA_{4+7} on fruit finish and packout of "Smoothee" treated with chlorpyrifos 50W in 1987*.

Treatment	Russet rating	Distribution of fruit in US grades(%) extra fancy/fancy	no. 1	utility	Crop value ($ per hectare)
Chlorpyrifos 50W	2.72a	34.3b	59.3a	6.6a	7383b
Chlorpyrifos 50W + GA_{4+7}†	2.42b	57.8a	40.5a	1.8a	8307a
Chlorpyrifos 50W + GA_{4+7}‡	2.53ab	47.6ab	49.0a	3.5a	7908ab

* Means in a given column followed by the same letter are not significantly different by Duncan's (1951) multiple range test, P = 0.05.
† Four applications of GA_{4+7} at 7 day intervals.
‡ Four applications of GA_{4+7} at 14 day intervals.

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OBITUARY

Augustus Burns Weathersby

1913 - 1988

Dr. Augustus Burns Weathersby, Emeritus Professor of Entomology, University of Georgia, died January 26, 1988, of a massive stroke.

A. B., to everyone who knew him, was born May 19, 1913, son of a minister, in Pinola, Mississippi. He received his B.S., M.S. and Ph.D. in entomology from Louisiana State University.

After receiving his master's degree in 1940, he worked for the Louisiana Department of Agriculture as an entomologist. An accomplished story teller, A. B. enjoyed relating such inane trials and tribulations as his attempts to help the city fathers control the town ant. In 1941 he met Olive Hammons, a school teacher who would later become his wife. They were married in April 1945.

With the outbreak of World War II, A. B. went into the U. S. Navy, joining the Epidemiology Unit of the Third Marine Division. His work in mosquito control carried him to Guadalcanal, Bougainville and Guam. From this experience he carried a piece of shrapnel in his finger. Unsure as to whether the shell fragment was of Japanese or American origin, he never applied for the Purple Heart. In 1946 A. B. helped set up the Naval Medical Research Institute in Cairo, Egypt. While in the Middle East, A. B. became an advisor to the Iranian Minister of Health. During an evening flight from Iran, A. B. and Olive were conscripted into the Caterpiller Club, an elite group whose members have abandoned disabled aircraft by parachute, in this case a B - 17. On landing A. B. immediately began searching and calling in the night to find Olive. Arabs gathered in the vicinity of the crash site mistakedly thought he was offering praise to Allah. The respect this misunderstanding garnered helped insure their safe passage through the desert. A. B. returned to Louisiana State University, completed his doctorate in 1954, and continued his military service. From 1947 until his military retirement as a commander in 1962, A. B. held various positions including special lecturer in the U. S. Naval Medical School and several assignments as entomologist - parasitologist.

After retiring from the Navy as a commander in 1962, A. B. joined the Department of Entomology, University of Georgia as a research associate. In 1964 he was named professor, the position he held until retirement in June of 1983. A. B. regularly taught undergraduate and graduate level courses concerned with insects of medical importance. A. B.'s enthusiasm, command of his subject and irrepressible humor made him a favorite of students. He developed an especially innovative and popular course called *Insects and Man*, for the non - science majors. In 1983 he was awarded the Entomological Society of America's Distinguished

Achievement Award in Teaching. A. B. participated in numerous honor and professional societies including: Sigma Xi, Phi Sigma and Gamma Sigma Delta. He was a member of the Entomological Society of America, American Society of Tropical Medicine, American Association for the Advancement of Science, American Mosquito Control Association, Society of Parasitology and the Georgia Entomological Society. He served as president of the Southeastern Society of Parasitiologists, president of the University of Georgia Chapter of Phi Kappa Phi and as a Fellow of the Royal Society of Tropical Medicine and Hygiene.

A. B.'s primary research interests were in the areas of host - parasite relations, insect susceptibility to disease organisms, the behavioral interactions of parasites and vectors, cryobiology and tissue culture.

A. B. was active in community affairs. He served as president of the Clarke Central High School Touchdown Club, president of the National Little League of Athens and was a member of the Dugout Club. The Clarke Central High School baseball field was recently refurbished and named Weathersby Field. A. B. served three terms as chairman of the Board of Deacons of Beech Haven Baptist Church. He also worked with the Chamber of Commerce and the Advisory Committee of the Clarke County Board of Education.

A. B. had many interests and talents. He played a number of instruments in high school and college bands, as well as the piano and violin. Noted for his rich bass voice, he sang in a barber shop quartet and in his church choir. He was an artist. He enjoyed painting, photography, woodworking and lapidary. He was well known for his gardening, having a particular affinity for azaleas.

A. B. is survived by his wife, Olive, and two sons, Richard M. of Athens and Robert B. of Dallas.

Rudolph T. Franklin
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Athens, Georgia

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TRIGONOTYLUS DODDI[1] (DISTANT) AS A PEST OF BERMUDAGRASS: DAMAGE POTENTIAL, POPULATION DYNAMICS, AND MANAGEMENT BY CUTTING

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Georgia Station, Griffin, GA 30223-1797

Abstract: The damage potential, phenology and dispersion of *Trigonotylus doddi* (Distant) were investigated in forage-type bermudagrass, *Cynodon dactylon* (L.) Pers. In a greenhouse study, adults fed on leaves, leaf whorls, and stems resulting in leaf chlorosis and severe stunting of stems. Stem height and plant dry matter production declined substantially as adult density increased. Seasonal trends of nymphs and adults were similar in all fields with population peaks occurring in late May through June and in September through October. Nymphal and adult populations were low in July and August. Both Iwao's regression method and Taylor's power law found that nymphs and adults were aggregated, with nymphs being slightly more aggregated than adults. Cutting of bermudagrass reduced populations of nymphs and adults an average (± SEM) of 82.6 ± 7.9 and 71.6 ± 10.2%, respectively. These results demonstrate the potential of *T. doddi* to damage forage-type bermudagrass with damage being most likely to occur in June and during the fall. Timely cutting of forage may be a useful tool for managing *T. doddi* in bermudagrass.

Key Words: *Trigonotylus doddi*, plant bug, bermudagrass, *Cynodon dactylon*, dispersion.

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Bermudagrass, *Cynodon dactylon* (L.) Pers., is the most widely grown warm-season perennial forage grass in the southeastern United States. The predominant plant bug attacking bermudagrass in Georgia is *Trigonotylus doddi* (Distant) (Byers 1967), which has been misidentified in earlier literature as *T. dohertyi* (Distant). No information is available on the biology and damage potential of *T. doddi* in bermudagrass. A related species, *Trigonotylus coelestialium* (Kirkaldy), has been associated with growth inhibition in small grain crops (Wheeler and Henry 1985), and this species and *Trigonotylus ruficornis* (Geoffroy) are recognized as pests of grass crops in the Palearctic region (Agafonova and Belizin 1964, Korcz 1979, Mikhailova 1981). Injury is caused by stylet penetration of plant tissue producing chlorotic spots on leaves and inhibition of plant growth (Wheeler and Henry 1985).

T. doddi is a tropicopolitan insect (Carvalho and Wagner 1957) and seems to occur wherever bermudagrass is grown. Other host plants of this insect include Japanese brome, *Bromus japonicus* Thunb. ex Murr.; johnsongrass, *Sorghum halpense* (L.); spangletop, *Leptochloa panicoides* (L.); southern crabgrass, *Digitaria ciliaris* (Retz.) goosegrass, *Eleusine indica* (L.); knotroot foxtail, *Setaria geniculata* (Lam.) P. Beav.; little barley, *Hordeum pusillum* Nutt.; and purple nutsedge, *Cyperus rotundus* L. (Snodgrass *et al.* 1984). All species of *Trigonotylus* heretofore studied overwinter in the egg stage and undergo one to several generations per year (Carvalho and Wagner 1957, Wheeler and Henry 1985, Blinn and Yonke 1986).

[1] HETEROPTERA: Miridae. Accepted for publication 23 August 1988.

The first objective of this research was to characterize the injury and assess the damage potential of *T. doddi* to bermudagrass. A second objective was to examine the population phenology and dispersion patterns of *T. doddi* to determine when damage is most likely to occur in the field. Information on the impact of cutting as a management tool also was obtained as part of the second objective.

MATERIAL AND METHODS

Assessment of Damage Potential

The impact of *T. doddi* on bermudagrass growth was examined in the greenhouse. Bermudagrass, cv. 'Coastal', sprigs were established in 15-cm diameter pots containing a 2:1 mixture of potting soil and sand. Pots were watered three times per week, and a complete nutrient solution (200 ppm N:P:K) was supplied weekly. After a six week establishment period, plants were cut to a stubble height of 5 cm. Plants regrew for 10 days before treatments were established in a randomized complete block design with 10 replications. Temperature in the greenhouse during the study ranged from 25 - 33° C with a photoperiod of 14:10 (L:D).

Treatments were a caged and uncaged control and caged plants with 1, 2, 4, and 8 adults per caged pot. Cages consisted of a 30-cm high aluminum screen (18 × 20 mesh) cylinder covered at the top with a clear plastic petri dish. The cages did not significantly ($P > 0.05$) affect any plant growth parameter, and presumably *T. doddi* adults behaved normally on the caged plants. Plants were inspected every 2 - 3 days, and adults were replaced as needed with unsexed, feral adults to maintian the desired treatment density. All adults were obtained from a field of 'Coastal' bermudagrass located near Griffin, GA. Feeding occurred for 14 days after which cages were removed, and foliage was clipped at the soil level. New regrowth was separated from the stubble, and final stem density, mean extended leaf height of stems, and dry weight of regrowth were measured. Plant measurements were analyzed with analyses of variance, and treatment means were separated using Duncan's (1955) multiple range test.

Population Dynamics

Populations of *T. doddi* nymphs and adults were monitored during 1985 and 1986 in 3 bermudagrass hay fields located in Pike Co., GA. Fields were 4 to 10 ha and consisted primarily of 'Coastal' bermudagrass. Other plant species present were common bermudagrass, johnsongrass and crabgrass, *Digitaria* sp. These species also are hosts of *T. doddi* (Snodgrass *et al.* 1984), but if possible they were avoided while sampling.

T. doddi was sampled using a D-Vac vacuum machine (Model 1A, D-Vac Co., Riverside, CA) equipped with a 0.09-m^2 sampling cone. Fifteen subsamples were taken about 1-m apart for each sample, and 8 samples were collected per field. A subsample was collected by placing the sampling cone into the foliage and pressing it to the ground for 15 seconds. Nymphs and adults were separated from the plant debris by using a previously described extraction box (Buntin 1988). A preliminary study found that extraction boxes recovered about 92% (N = 24) of nymphs and 95% of adults. Fields were sampled between 1300 to 1700 hrs every 7 to 10 days from 30 May to 20 October in 1985 and 1 April to 7 November in 1986. Sampling in field 3 in 1986 was discontinued after 14 October when the field was sprayed on 10 October with methomyl at 1.1 kg AI/ha to control the fall armyworm, *Spodoptera frugiperda* (J. E. Smith).

Population Dispersion

Dispersion patterns of nymphs and adults were examined using Iwao's (1968) regression method and Taylor's (1961) power law. These analyses quantify the relationship between sample variance (s^2) and mean density ($\bar{x}$). Iwao's regression method regresses Lloyd's mean crowding index ($\overset{*}{m}$), where $\overset{*}{m} = \bar{x} + [s^2/\bar{x})-1]$ on mean density such that $\overset{*}{m} = \alpha + \beta\bar{x}$. Least squares estimates of α and β were computed (SAS Institute 1985). Taylor's power law relates to s^2 to $\bar{x}$ such that $s^2 = a\bar{x}^b$, where a and b were estimated using a linear regression model. Both dispersion analyses were calculated for nymphs and adults by year and combined for both years. The slope coefficients (b and β) of both methods indicate a uniform, random, and aggregated dispersion pattern when slope < 1, slope $= 1$, and slope > 1, respectively. The intercept term (α) of Iwao's method also can be used as a measure of the basic unit aggregation where $\alpha + 1$ is the number of individuals per aggregate (Iwao and Kuno 1971).

Effect of cutting on T. doddi populations

Cutting and removal of hay had a substantial impact on *T. doddi* populations. The population reductions of nymphs and adults were determined by comparing population densities before and after cutting and calculating the percentage reduction in density. Calculations were made only when population density before cutting exceeded 2 nymphs or adults per sample. Calculated reductions were compared to no reduction using a one-tailed t-test (Steel and Torrie 1960).

RESULTS AND DISCUSSION

Assessment of Damage Potential

Adults fed on leaves, leaf whorls and stems of bermudagrass, which resulted in leaf chlorosis and severe stunting of stems. Feeding at the base of the leaf whorl caused distortion of unfolded leaves within the whorl, which typically killed the apical meristem and leaf whorl. Adult feeding produced large reductions in stem height and plant dry weight (Table 1) with reductions increasing up to 4 adults/cage. Eight adults/cage did not cause additional significant reductions in stem height or plant dry weight. A density of 4 adults/cage reduced stem height by 55.6% and plant dry weight by 56.7%. Stem density tended to delcine as adult density increased, but stem density of no treatment was significantly different ($P > 0.05$) than the stem density of the uninfested, caged plants. *T. doddi,* therefore, reduced plant dry matter production primarily by reducing dry weight per stem.

Table 1. Effect on feeding by *T. doddi* adults on stem density, stem height, and dry weight of bermudagrass.

Adults per pot	Stems per pot	Stem height (cm)	Plant dry weight per pot (mg)
0, uncaged	6.0 ab*	23.4 a	353 a
0, caged	6.1 ab	25.7 a	344 a
1	7.1 a	17.9 b	250 b
2	5.8 ab	18.5 b	235 bc
4	5.2 b	11.4 c	149 cd
8	4.6 b	11.4 c	137 d

* Means followed by the same letter are not significantly different ($P > 0.05$; Duncan's multiple range test).

Population Dynamics

The seasonal trends of nymphs and adults were similar in all fields with two peaks of activity being observed in both years (Fig. 1 & 2). The first peak occurred during late May through June, and the second peak occurred in September and October. The second peak occurred about one month later in 1986 than 1985. Populations of both nymphs and adults in all fields were low during July and August in both years.

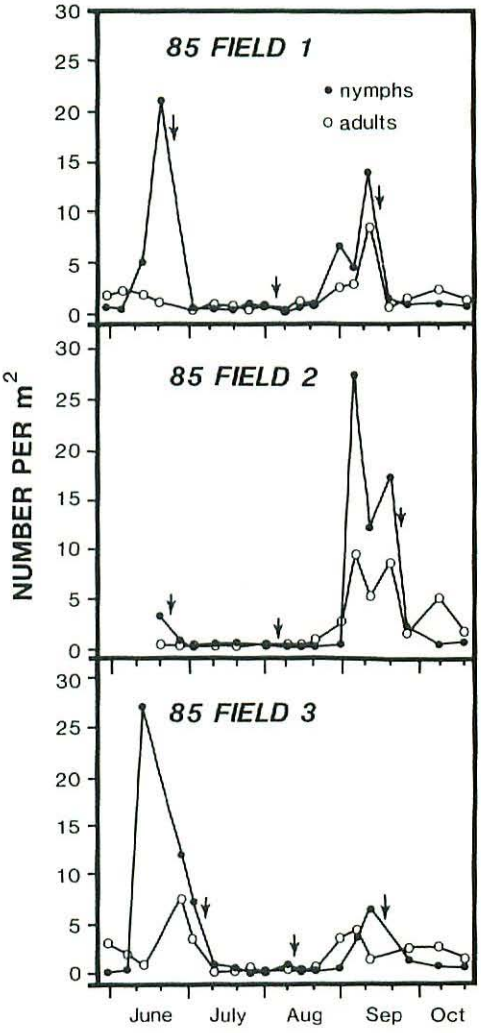

Fig. 1. Phenology of *T. doddi* nymphs and adults in 3 'Coastal' bermudagrass fields in 1985. Arrows indicate cutting.

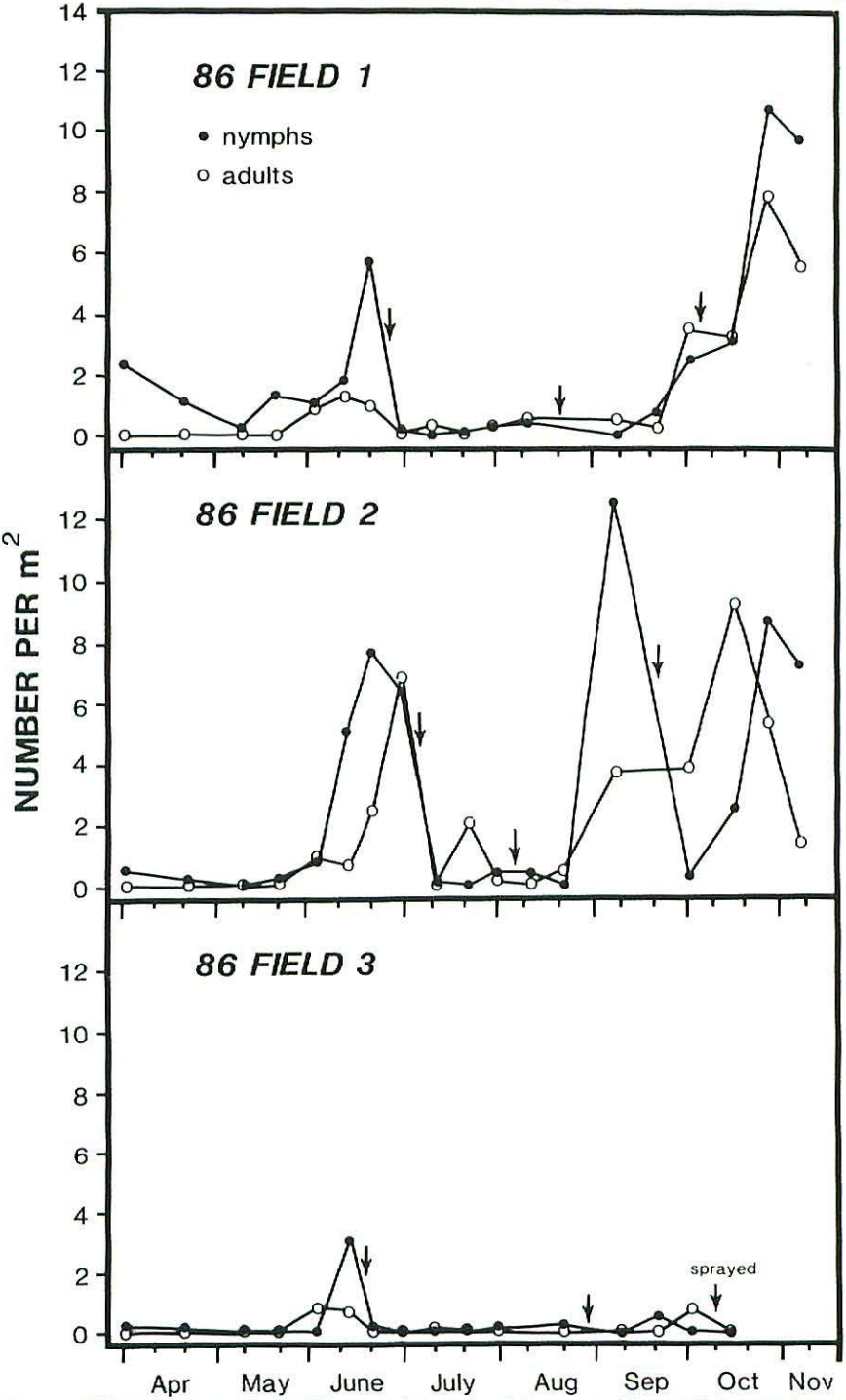

Fig. 2. Phenology of *T. doddi* nymphs and adults in 3 'Coastal' bermudagrass fields in 1986. Arrows indicate cutting.

The number of generations is not clear because cutting interferes with normal phenology, but 3 to 4 generations per year are suggested by the data. The presence of nymphs in April, which were all early-stage nymphs, and the absence of adults until about 1 June in 1986 suggests that the first activity peak arose from overwintering eggs. Although the developmental time of *T. doddi* is not known, a closely related species, *T. coelestialium*, averaged a total of 31.5 days for egg to adult development (26.8 days) and preoviopositional development (4.7 days) at 25° C in the laboratory (Blinn and Yonke 1986). If this value is similar to the minimal generation time to *T. doddi*, then at least one generation should have occurred during July and August. This generation, however, was not distinct in either year, probably because fequent cutting interferred with the life cycle of *T. doddi*. A third and partial fourth generation seemed to occur in the fall with females depositing eggs that overwintered in bermudagrass stubble.

Population Dispersion

Both Iwao's regression method and Taylor's power law indicated that *T. doddi* nymphs and adults were aggregated (Table 2). All of the combined and yearly values of b and β were significantly ($P < 0.01$) greater than 1.0. Both procedures revealed that nymphs were more aggregated than adults. Eggs in the greenhouse experiment were laid in bermudagrass stems in clusters of 2 to 6 which would tend to result in aggregations of nymphs. Increased mobility of adults as compared with nymphs would tend to further reduce aggregation of adults. None of the α terms of Iwao's regression procedure was significantly ($P > 0.05$) greater than zero indicating that the individual is the basic unit of aggregation for both nymphs and adults (Iwao and Kuno 1971).

Table 2. Population dispersion parameters of *T. doddi* nymphs and adults in bermudagrass.

Stage	Year	No. of Observations	Taylor's Power law			Iwao's regression method		
			a	b†	r^2	a‡	β	r^2
Nymphs	1985	44	1.71	1.35**	0.96	0.252	1.29**	0.95
	1986	29	1.24	1.38**	0.88	-0.014	1.35**	0.90
	Combined	70	1.30	1.37**	0.92	-0.011	1.31**	0.93
Adults	1985	42	1.36	1.16**	0.87	0.059	1.25**	0.96
	1986	27	1.14	1.35**	0.93	0.006	1.20**	0.95
	Combined	66	1.07	1.25**	0.86	-0.136	1.23**	0.95

†, *, ** indicate greater t-statistic at $P < 0.05$ and $P < 0.01$, respectively, for test of H_0:b or $\beta = 1$.
‡ No α-term is significantly ($P < 0.05$) different than zero.

Effect of Cutting on T. doddi Populations

Cutting and harvest of bermudagrass substantially reduced populations of both nymphs and adults. The mean ($\pm$ SEM) percentage reduction of nymphs in 1985 ($N = 8$) and 1986 ($N = 8$) was 91.8 $\pm$ 2.5 and 73.5 $\pm$ 15.0%, respectively, with a combined reduction of 82.6 $\pm$ 7.9%. Cutting reduced adult populations in 1985 and 1986 an average of ($\pm$ SEM) of 83.5 $\pm$ 11.4 and 59.0 $\pm$ 16.2%, respectively, with a combined reduction of 71.6 $\pm$ 10.2%. All of these reductions were significant ($P < 0.05$). The generally lower and more variable (i.e., larger SEM) reductions of adults as compared with nymphs probably were caused by the increased mobility of

adults which can escape the direct impact of cutting. Reductions in adult populations probably reflect both direct mortality and dispersal from the field, whereas reductions of nymphal populations are caused by a combination of mortality and progression of nymphs to the adult stage between sampling periods. The typical use of a mower/conditioner, which cuts and crimps the hay in one operation, probably causes substantial direct mortality of both nymphs and adults. Additional causes of mortality, especially for nymphs, are the removal of food and lethal temperature extremes that can occur when the forage is removed.

CONCLUSIONS

T. doddi has the potential to severely reduce forage production of bermudagrass. Although not examined in this study, *T. doddi* injury also could be expected to adversely affect forage quality. The mechanism of injury by *T. doddi* seems similar to that caused by plant bugs in other forage grasses (Arnott and Bergis 1967, Todd and Kamm 1974). The seasonal phenology of *T. doddi* indicates that damage is most likely to occur during June and again in September and October. Shortening day length and cool temperatures limit bermudagrass growth during September and October; consequently, potential yield losses may be greater in June than in the fall. Populations were only moderately aggregated; therefore, damage could be expected to be generally distributed throughout a field rather than to occur in a patchy pattern. Additional research is needed, however, to determine if densities typically observed in the field are sufficiently large to adversely affect bermudagrass forage production. If control is warranted, timely cutting may be a useful method for reducing *T. doddi* populations in bermudagrass.

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INBREEDING DEPRESSION AND GENE FREQUENCY CHANGES FOR AGRONOMIC TRAITS IN CORN SYNTHETIC SELECTED FOR RESISTANCE TO EUROPEAN CORN BORER[1]

J. R. Klenke,[2] W. A. Russell,[3] W. D. Guthrie,[4] and O. S. Smith[5]

Abstract: A corn (*Zea mays* L.) synthetic, BS9, was evaluated to determine the effect that four cycles of S_1 recurrent selection to improve resistance to European corn borer (ECB), *Ostrinia nubilalis* (Hübner), had on several other agronomic traits. The analysis used is a modification of linear regression analysis that is a more powerful technique than a means separation test to measure changes in agronomic traits during recurrent selection for ECB resistance. The model allows partition of the changes in agronomic traits into those caused by changes in allelic frequencies due to selection (implying linkage or pleiotropy), and changes caused by genetic drift, which results because of small population size. Recurrent selection for ECB resistance caused significant decreases in BS9 for grain yield (YLD), 300-kernel weight (KWT), ear diameter (EDI), ear length (ELH), ear height (EHT), and plant height (PHT). Changes in gene frequency ($\Delta p\alpha$) were significant for EDI, EHT, and PHT, and it was suggested that indirect selection for shorter internode length was the primary cause of these changes. Yield decreased from 6.07 Mg ha^{-1} for BS9CO to 4.42 Mg ha^{-1} for BS9(CB)C4, a 27.2% reduction. Change in gene frequency was estimated to cause an 8.4% reduction, and inbreeding depression was estimated to cause an 18.8% reduction, but only the latter was significant. The random fixation with loss of favorable alleles at some loci that were heterozygous in the CO (referred as drift) appears to have lowered the advanced cycle means. Selection to maintain maturity was effective by compensating for the weighted change in gene frequencies at heterozygous loci with an increase in the fixation of the favorable alleles. S_2 recurrent selection, therefore, was recommended to include yield in the selection criteria to reduce the yield loss in advanced cycles from recurrent selection for resistance to the two generations of ECB.

Key Words: Maize, *Zea mays*, *Ostrinia nubilalis*.

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The primary objective of a recurrent selection program is to increase the frequency of the favorable alleles, while maintaining the genetic variability, for a quantitatively inherited trait such as yield or European corn borer (ECB), *Ostrinia nubilalis* (Hübner), resistance (Hallauer and Miranda 1981). Usually, it is preferred that other agronomic traits do not change in their phenotypic characteristics. Assessment of the progress from recurrent selection, therefore, involves evaluation of traits under selection and also indirect effects of selection on other traits.

Studies on the progress of many recurrent selection programs have been summarized by Hallauer and Miranda (1981). Usually, the analysis used to evaluate progress from selection regresses changes in means of populations on cycles of selection by using least-squares procedures (Eberhart 1964).

Realized gains from recurrent selection programs are often less than predicted gains because of inbreeding depression caused by small population size (Smith 1984).

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Realized response in the change of the mean of the population is a function of the change in gene frequency and inbreeding depression (Smith 1979a). Smith (1979a,b, 1983) proposed a modification of the Hammond and Gardner model to separate changes in the mean caused by increase in frequencies of favorable alleles from changes in the mean due to inbreeding depression.

The Smith model has been applied to results of several recurrent selection programs to demonstrate the effects of drift on grain yield. Eberhart et al. (1973) reported that the means of two populations in a reciprocal recurrent selection (RRS) program for grain yield had not changed significantly. Smith (1979a), however, was able to detect from this study that RRS was effective in increasing the frequencies of favorable alleles. The estimate of drift effects accounted for the lack of significance in the change of the means of the populations. Similar results were obtained by Smith (1979b) in two populations in a recurrent selection program with an inbred tester reported by Russell et al. (1973). Effects of inbreeding depression due to small population size were evident in the means of the populations per se in three other studies by Smith (1979b, 1983, 1984).

BS9(CB)C4 was released to the seed industry in 1982 after four cycles of S_1 recurrent selection for ECB resistance because of its potential value in breeding programs for resistance to the two generations of ECB normally found in Iowa (Russell and Guthrie 1982). The objectives of this study were to estimate gene frequency changes and inbreeding depression on agronomic traits of BS9 by using the model proposed by Smith.

MATERIALS AND METHODS

The original population, BS9CO, was developed by mating 10 corn inbred lines that were selected for their combining abilities and for their resistance to both ECB generations (Pesho et al. 1965). A more detailed description of the lines and development of BS9 is given by Russell and Guthrie (1982).

Experimental plots for this study were planted in eight environments, four Iowa locations in 1983 and 1984, and the experiment consisted of five entries: BS9CO, BS9(CB)C4, BS9C × BS9(CB)C4, BS9CO selfed, and BS9(CB)C4 selfed. The experimental design was a split-plot, with random-mated and selfed populations being nested in separate, bordered subplots within replications.

Agronomic practices, including planting date, fertilization, and weed control, were followed in all environments to promote high productivity. The Ames and Ankeny locations were hand-planted in single-row plots, 0.76 m × 4.32 m. Seeds were planted two per hill in 17 hills spaced 25.4 cm, and plants were later thinned to one per hill for a final density of 51,666 plants per hectare. The Kanawha and Martinsburg locations were machine-planted in single-row plots, 0.76 m × 5.00 m. Thirty-four seeds were planted, and plants were later thinned to a uniform stand of 19, for a final density of 50,000 plants per hectare.

Anthesis (POL) and silking (SIL) dates were recorded at the Ames location when 50% of the plants in a row were shedding pollen and silking, respectively. After pollination had been completed, plant and ear heights were taken on five competitive plants in each plot at all locations. Plant height (PHT) was measured to the flag leaf at the base of the tassel, and ear (EHT) was measured to the top ear node.

Ten competitive plants per plot were hand-harvested and the ears were dried to a uniform moisture. Data were taken on the diameter of the primary ears (EDI)

and length of all ears (ELH) and recorded on a per-plant basis. The ears were then shelled, and the grain was weighed and recorded on a per-plot basis (YLD). A sample of the grain was saved of each plot to permit a determination of 300-kernel weights (KWT).

The statistical model used to analyze changes in the population means for agronomic traits is based on the model developed by Smith (1979). In this analysis, the population mean for the original cycle of selection (CO) is written as a function of the homozygous (A_0) and heterozygous (D_0) effects in the CO. The population means and crosses after n cycles of selection are expressed as changes in allelic frequencies association with selection ($A_l + D_l$), and changes in allelic frequencies caused by drift (D_q). The A_0, D_0, A_l, D_l, and D_q terms can be related to additive and dominance effects at a single locus and the allelic frequencies at those loci as follows (Smith 1979b) and Table 1:

A_0 — the weighted sum (where the weights are the allelic frequencies) of the contribution of homozygous effects (a) to the mean of the CO:
D_0 — the weighted sum of the contribution of heterozygous effects (d) to the mean of the CO;
A_l — the linear function of the changes in allelic frequencies caused by selection ($\Delta\rho$), weighted by additive effects;
D_l — the linear function of $\Delta\rho$ due to selection, dominance effects; and
D_q — the quadratic function of $\Delta\rho$ caused by selection drift, weighted by dominance effects.

The mathematical expectations of the parameters are presented in Table 1. The assumptions are that the base population (CO) is diploid and that there is no epistasis. Estimates of the parameters were derived in this study from the following equations (Smith 1979b);

$$\begin{aligned}
\text{CO} &= A_0 + 2D_0 \\
\text{C4} &= A_0 + 2D_0 + 2A_l(N) + 2D_l(N) + 2D_q(N^2) \\
\text{CO selfed} &= A_0 + D_0 \\
\text{C4 selfed} &= A_0 + D_0 + 2A_l(N) + D_l(N) + D_q(N^2) \\
\text{CO} \times \text{C4} &= A_0 + 2D_0 + A_l(N) + D_l(N)
\end{aligned}$$

where N is the number of cycles of selection. A more thorough description of the parameters is given below in the order that they were estimated.

D_0 can be estimated from the difference of the CO and CO selfed. One generation of selfing reduces the number of heterozygous loci by half. D_0, therefore, is an estimate of the amount of inbreeding depression observed after one generation of selfing. Doubling D_0 ($2D_0$) estimates the total contributions of the heterozygous loci to the CO mean.

A_0 is the remainder of the contribution to the CO mean, which is an estimate of the homozygous loci effects. A_0 also is the estimate of the mean of a random sample of inbred lines (homozygous) developed from the base popualtion.

$A_l(N) + D_l(N)$ is the difference between the CO and the population cross (CO $\times$ Cn), which is $(A_l + D_l)(N)$. This is an estimate of the weighted change in allelic frequencies, independent of the effect of drift, because $A_l + D_l = \Delta\rho a + \Delta\rho(q\text{-}p)d = \Delta\rho\alpha$, which is half the change in the mean of a random-mating population from

selection (Kempthorne 1973). The difference between CO and CO × Cn, therefore, is the linear function of the change in allelic frequencies due to selection, weighted by additive and dominance effects.

Table 1. Two-allele and multiple-allele notations for expectations of parameters used in the model to estimate change in gene frequency in BS9.

Parameter	One-locus two-alleles (Smith 1979)*	More than one locus, multiple-alleles (Smith 1979)
A_O	$\mu + (p-q)a$	$\sum_{k=1}^{l} (\mu_k + 2 \sum_{i=1}^{n} \rho_{ik} a_{ik})$
D_O	$2pqd$	$\sum_{i \neq i'}^{n} \sum_{k=1}^{l} \rho_{ik} \rho_{i'k} d_{ii'k}$
A_l	$\Delta\rho a$	$\sum_{i=1}^{n} \sum_{k=1}^{l} \Delta\rho_{ik} a_{ik}$
D_l	$\Delta\rho(q-p)d$	$\sum_{i \neq i'}^{n} \sum_{k=1}^{l} \Delta\rho_{ik} \rho_{i'k} d_{ii'k}$
D_q	$\Delta\rho^2 d$	$\sum_{i \neq i'}^{n} \sum_{k=1}^{l} \Delta\rho_{ik} \Delta\rho_{i'k} d_{ii'k}$

* ρ = frequency of favorable allele, $q = 1-\rho$, a = homozygous effect of favorable allele, d = heterozygous effect, and $\Delta\rho$ = change in allele frequency after selection.

The differences between CO × Cn and Cn is $(A_l + D_l)(N) + 2D_q(N^2)$. The Cn contains twice the effect of the weighted change in gene frequency due to selection $(A_l + D_l)$ compared with CO × Cn, and removing this effect provides an estimate of D_q. Quadratic changes in allelic frequencies from a small number of cycles of selection are usually small relative to the effects of drift (Smith 1979b). The D_q term, therefore, is primarily the effect of drift or loss of heterozygotes in the Cn population causing inbreeding depression after the effects of selection have been removed. D_q also is a measure of the loss in amount of predicted gain due to finite population size.

For a quantitative trait controlled by many loci, the expected change in allelic frequency due to drift (sampling) is zero (i.e., $\Sigma\Delta\rho = 0$). Drift, therefore, should have little effect on the estimates of A_l and D_l. The sum of the change in allelic frequencies squared, however, is always positive and can be large relative to $\Delta\rho$ due to selection, depending on the number of lines recombined. From the equation $D_q = \Delta\rho^2 d$, drift is the square of the change in allelic frequencies, weighted by the dominance effects. This, by definition, is the estimate of mid-parent heterosis (Falconer 1981). The D_q term, therefore, can also be considered an estimate of heterosis in the CO × Cn cross (Smith 1983).

A_O and D_O in $\Delta p\alpha$ can be separated because of the loss of half the heterozygous loci in the selfed population affects D_l but not A_l. The D_l term can be estimated from a linear function of the population per se, per se selfed, and the cross of the CO to the Cn. The A_l term is finally estimated from the remainder.

The standard errors for the parameter estimates were calculated as described by Draper and Smith (1966).

RESULTS AND DISCUSSION

Least-square estimates of genetic parameters and means for various agronomic traits are presented in Table 2. Means decreased significantly in the C4 population relative to CO for YLD, KWT, EDI, ELH, EHT, and PHT and increased significantly for SIL. The means for CO × C4 were significantly lower than the CO population per se for EHT, PHT, and EDI, which is also indicated by the significant $\Delta p\alpha$.

The A_l and D_l terms were not significant for EDI, but the sum of the two ($\Delta p\alpha$) was significant, indicating that a change in allelic frequencies caused by selection, weighted by additive and dominance effects, significantly decreased EDI. For EHT and PHT, the D_l term was significant, indicating that selection caused a change in gene frequency detected by a decrease in the contribution of the heterozygous loci to the mean. A A_l term, however, was not significant for EHT and PHT, indicating no detectable change in the contribution of the homozygous loci.

The cause for the indirect selection on EDI, EHT, and PHT was not obvious. Russell et al. (1979) reported a change in gene frequency for these traits in three synthetics after three cycles of recurrent selection for first-generation resistance. They stated that a change in gene frequency may have been caused by pleiotropism, linkage, and independent selection. In the present study, no selection was imposed on EHT or PHT because no measurements were taken. Additionally, first-generation ECB damage ratings tend to favor more vigorous plants because they can "grow-out" of the larval feeding sites more quickly and thus reduce the size of the lesions. Chromosome linkage and pleiotropism are unlikely causes because taller and later lines tend to be the most resistant.

Selection for second-generation ECB resistance (SGR) in BS9 was primarily based on resistance to stalk tunneling by the fifth-instar larvae (borers), which was determined by cavity counts (CVC) (one cavity ca. 2.5 cm), with secondary emphasis on resistance to sheath-collar feeding in the last two cycles of selection (W. A. Russell, W. D. Guthrie, and P. R. White, unpublished, Maize Breeding Research Project Annual Reports, 1972, 1975, 1978, 1980, Dep. of Agronomy, Iowa State Univ., Ames). Selection for a low number of cavities based on total plant counts may have been a possible cause for reduction of EHT and PHT, but the amount of tunneling in the S_1 lines of BS9 populations was too low for plant size to be a contributing factor. Although a high correlation exists between SGR and CVC (Guthrie et al. 1978), progress in SGR was slower than CVC in BS9. This may suggest that resistance to sheath-collar feeding was caused by some kind of antibiosis, but that resistance to tunneling was caused by hardness of the stalk (W. A. Russell, W. D. Guthrie, and P. R. White, unpublished, Maize Breeding Research Project Annual Report, 1980, Dep. of Agronomy, Iowa State Univ., Ames). Martin and Russell (1984) reported that internode length was significantly correlated with hardness of stalk, measured as rind strength, and ear diameter ($r = -0.62^{**}$ and $r = 0.74^{**}$, respectively). Selection for reduced CVC, therefore, could indirectly influence internode length, which caused the decrease in EDI, EHT, and PHT.

Table 2. Least-square estimates of genetic parameters and means for yield, yield components, and other agronomic traits of BS9 populations evaluated in six environments.

Parameter‡	Traits†							
	POL	SIL	YLD	KWT	EDI	ELH	EHT	PHT
			(Mg ha^{-1})	(g)	cm			
A_0	29.3 ± 0.6	34.2 ± 1.3	1.49 ± 0.41	60.1 ± 3.4	3.58 ± 0.09	10.6 ± 1.0	77.1 ± 3.5	155.7 ± 5.4
D_0	-3.5 ± 0.4	-4.5 ± 0.8	2.29 ± 0.26	5.3 ± 2.2	0.32 ± 0.06	3.2 ± 0.6	12.0 ± 2.2	27.5 ± 3.3
A_1	-0.4 ± 0.1	-0.3 ± 0.2	0.02 ± 0.07	0.5 ± 0.6	-0.01 ± 0.02	0.1 ± 0.2	-0.5 ± 0.6	0.4 ± 1.0
D_1	0.3 ± 0.1	0.2 ± 0.3	-0.09 ± 0.11	-0.1 ± 0.9	-0.02 ± 0.02	-0.1 ± 0.3	-2.2 ± 0.8	-3.6 ± 1.2
$\Delta\rho\alpha$§	-0.1 ± 0.1	-0.0 ± 0.2	-0.06 ± 0.06	-0.5 ± 0.5	-0.03 ± 0.01	0.1 ± 0.2	-2.7 ± 0.3	-3.2 ± 0.4
D_q	0.0 ± 0.1	0.1 ± 0.1	-0.04 ± 0.01	-0.1 ± 0.1	0.00 ± 0.01	-0.1 ± 0.1	0.1 ± 0.1	0.1 ± 0.2
Means								
CO	22.3	25.2	6.07	70.7	4.21	17.0	102.0	211.1
C4	22.3	26.5**	4.42**	61.4**	3.84**	15.7*	80.6**	185.2**
CO selfed	25.8**	29.7**	3.78**	65.4*	3.89**	13.8**	89.3**	183.2**
C4 selfed	24.2**	29.3**	3.04**	62.6**	3.66**	13.6**	76.7**	171.7**
CO × C4	22.0	25.0	5.81	68.5	4.08*	17.2	89.8**	196.9**
Mean	23.3	27.1	4.62	65.7	3.94	15.5	87.7	189.6

*, ** Significantly different from CO at the 0.05 and 0.01 probability levels, respectively.

† Abbreviations for traits: POL = date of anthesis, SIL = date of silking, YLD = grain yield, KWT = 300-kernel weight, EDI = ear diameter, ELH = ear length, EHT = ear height, and PHT = plant height.

‡ Parameters are defined in Smith (1983).

§ α = average effect of an allelic substitution, $\alpha = a + (q - p)d$.

The D_q term was significant only for YLD. Substitution of the estimates for the parameters in the equation for the C4 population, $\Delta p\alpha[2A_l(N) + 2D_l(N)]$ and $D_q[2D_q(N^2)]$, caused reductions of 0.51 and 1.14 Mg ha^{-1}, respectively, from the CO mean for YLD. Changes in gene frequency ($\Delta p\alpha$) and inbreeding depression (D_q), thus, were estimated to account for 8.4 and 18.8%, respectively, of the reduction in yield in BS9(CB)C4. The D_q term, however, was the only estimate that was significant. If the Δp^2 term in D_q was caused primarily by drift (Smith 1979b), then the greatest decrease in the C4 yield was caused by inbreeding depression due to a small population size.

The inbreeding coefficient of the C4, where $N_e \simeq N$ (Robertson 1961), was calculated to be 6.9% (Klenke 1985), but the D_q term was 18.8%. The difference between these two estimates may be accounted for by using the product for the effects of drift on the components of yield. Although none was significant, the reductions due to D_q for KWT, EDI, and ELH were 6.9, 2.8, and 10.3% of the BS9CO mean, respectively. Subtraction of these three terms from unity gives the values of C4 for the three traits, expressed as a percentage of the CO caused by drift, of 93.1, 97.2, and 89.7%, respectively. The product of the components is 81.2%, which is an 18.8% reduction and equal to the 18.8% reduction in yield. Similarly, the product of the three components based on $\Delta F = 6.9\%$ is $(.931)^3 = 80.7\%$, which is a 19.3% reduction. In this study, the reduction in yield due to drift, therefore, seems to be related to the inbreeding depression of the three components of yield.

Another explanation, indicated by the Smith model, for the decrease in yield of the C4 could be a large amount of random fixation at heterozygous loci (drift). Estimates for the contributions to yield of the BS9CO from the homozygous loci (A_0) and heterzygous loci ($2D_0$) were 1.49 and 4.58 Mg ha^{-1}, or 24.5 and 75.5%, respectively.

The A_l term is not significant but positive, indicating that there was no loss in the frequency of the favorable alleles; thus, repulsion-phase linkage was not a contributing factor. Linkage to alleles for other traits under direct and indirect selection (FGR, SGR, CVC, and POL, and EDI, EHT, and PHT, respectively) could contribute to inbreeding depression because populations in linkage equilibrium would fix alleles not influenced by selection at random. The greatest decrease in subsequent cycles of BS9, therefore, seemed to be the result of random fixation of alleles contributing to yield, perhaps increased by linkages to other traits affected by selection, rather than by a change in gene frequencies due to selection.

The A_l term was significant for only POL, which was the only trait influenced by direct selection in BS9. Selection was imposed on POL in BS9 so that the relative maturity of the selected S_1 lines was no later than the mean of the S_1 population. The means of the CO, C4, and CO × C4 were not significantly different; thus D_q and $\Delta p\alpha$ were not significant. The A_l and D_l terms, however, were significant but of opposite sign. Because dominance of POL was for earliness and earliness is the most desirable direction of selection, the A_l and D_l terms are more easily understood if they are expressed as days before a certain date. To keep the mean of the CO and C4 populations approximately the same, the POL data were converted to the number of days before August 14, which will make dominance and gain, in the desired direction, positive. The only parameter in the Smith model that is changed by this transformation is A_0, which will not affect the interpretations.

The estimates of the parameters become:

A_0	15.8 ± 0.6
D_0	3.5 ± 0.4
A_l	0.4 ± 0.1
D_l	-0.3 ± 0.1
$\Delta\rho\alpha$	-0.1 ± 0.1
D_q	0.0 ± 0.1

The A_l and D_l terms are of approximately equal magnitude, which sum to produce a $\Delta\rho\alpha$ term that is not significant. The weighted change in gene frequencies at heterozygous loci (D_l), or heterozygous effects, therefore, was compensated by the fixing of the favorable alleles or homozygous effects. Fixation of the favorable allele rather than random fixation was caused by selection.

The effects of selection on POL may also perform similarly with other traits such as yield. Selection may not reduce drift or may increase D_q if $\Delta\rho_{ik}$ and $\Delta\rho_{i'k}$ (Table 1) are increased. The effect of selection to reduce unfavorable changes in the mean would be to ensure that fixation is primarily of the favorable allele. Because of limited resources for replication yield trials and the importance of population size to reduce drift, a S_2 recurrent selection program as described by Hallauer and Miranda (1981) would be the most desirable method to implement. Although this would require an extra year in temperate zones, selection can be conducted in two seasons. The S_1 lines could be evaluated for FGR, POL, PHT, and SGR in one or two replications to eliminate the most undesirable lines. These traits could then be evaluated again in a small population of S_2 lines, in addition to evaluations for yield in replicated trials. The main emphasis of selection should be on SGR and YLD, with lesser emphasis on FGR, while attempting to maintain the means for POL and PHT.

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DISRUPTION OF OLFACTORY COMMUNICATION IN ORIENTAL FRUIT MOTH AND LESSER APPLEWORM IN A VIRGINIA PEACH ORCHARD[1]

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Abstract: Dispensers (20-cm polyethylene capillary tubing; "ropes") containing Oriental fruit moth pheromone were placed in a 4-ha block of peaches shortly before first male flight. This placement of ropes disrupted attraction to conventional pheromone traps for the duration of the season. Fruit damage was not significantly different from that in a control block (conventional pesticide application); twig damage was significantly lower relative to the control. Implications for pest management in mature and nonbearing blocks are discussed.

Key Words: Oriental fruit moth, *Grapholita molesta*, lesser appleworm, *G. prunivora*, pheromone, disruption, peach.

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Oriental fruit moth, *Grapholita molesta* (Busck), is an introduced pest of peach and apple. It has three to four generations in the northern part of its range (four in Virginia) and six to seven in the south (Bobb 1973; Brunner and Howitt 1981). Larvae of early generations tunnel in ends of twigs, while those of later generations bore into fruit flesh.

Lesser appleworm, *G. prunivora* (Walsh), is a native species that causes damage similar to that of *G. molesta* (Brunner and Howitt 1981). It appears to have only one to two generations throughout its range (Quaintance 1908; Brunner and Howitt 1981), and is attracted to pheromone traps used for *G. molesta* (Gentry et al. 1974, 1975; Cardé et al. 1977).

Several studies (reviewed by Gentry et al. 1982) have employed the sex pheromone of *G. molesta* in a variety of dispensers to attempt disruption of olfactory communication in that species. Most studies examined disruption of attraction to conventional traps but did not examine damage. Such disruption of male capture using varying pheromone-release technology has been reported for periods ranging from two to five weeks (Gentry et al. 1974, 1975) to 121 days (Cardé et al. 1977). Other studies have used small experimental orchard units which may have allowed immigration of gravid females from outside the treated area (Rothschild 1982).

The present study, in which a new type of dispenser was used in a block large enough to minimize the effect of immigrating gravid females, was undertaken to determine not only the disruption of attraction of *G. molesta* to conventional traps, but also the feasibility of this technique for preventing damage by this species in the Appalachian fruit-growing region.

MATERIALS AND METHODS

The treated area was a 4-ha 'Monroe' peach block at Batesville, Virginia. Dispensers were made of 20-cm polyethylene capillary tubing (hereafter called

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"ropes"; Shin-Etsu Chemical Co., Ltd., Tokyo), each loaded with 75 mg of pheromone [93% (Z)-8-dodecenyl acetate: 6% (E)-8-dodecenyl acetate: 1% (Z)-8-dodecenol]. On 4 April 1986 (about petal fall stage), two ropes were placed in each tree at ca. 2-m height to achieve a rate of 1000/ha. In 1987, ropes were placed on 14-15 April (full bloom). Release rate from ropes is 5-12 mg/ha/hour (P. Kirsch pers. comm.). The block was 26 rows wide at the widest point but narrowed to a bottleneck of 10 rows at one edge where it adjoined an additional peach orchard. Except for an apple orchard across a road from the treated area, the block was surrounded mainly by mixed deciduous woodland. The treated block had no insecticide treatment for the entire season, except for one parathion spray applied mistakenly.

The control area for both years was a 4-ha block of 'Rio-Oso-Gem' peaches separated from the treated block by 0.2 km of apple orchard and woodland. The control treatment consisted of a program of conventional insecticide applications, parathion 0.85 kg ai/ha, every two weeks from petal-fall until two weeks before harvest. The control treatment was defined as such (rather than a totally untreated control) because of the need to compare disruption technology with conventional insecticide practices. The control cultivar is expected to be equivalent to the treatment cultivar in susceptibility to injury by *G. molesta* because both have late-maturing fruit, an important factor in varietal susceptibility to this species.

Because of the large size of plots deemed necessary, the cost of replication would have been very high. Therefore each plot was treated as a nonreplicated experimental unit and inferential statistics were not applied (Hurlbert 1984). Descriptive statistics were employed (Altieri and Schmidt 1985).

As a means of determining disruption, seven commercially available *G. molesta* pheromone traps (Trece Inc., P. O. Box 5267, Salinas, CA 93915) were placed in each block on 11 April 1986. In 1987, four traps were placed in each block on 13 April. Each trap was separated by at least 60 m. Numbers of captured *G. molesta* and *G. prunivora* were recorded weekly and the moths were removed from the traps. Identifications were periodically confirmed by dissecting genitalia (Heinrich 1926). Lures were replaced after six to seven weeks. Trap bottoms were replaced when they became excessively contaminated with insect parts, dust, etc.

In 1986, damage was evaluated on 3 July and 21 August (harvest). On 3 July, five trees per block were randomly selected for sampling; on 21 August, ten trees per block were sampled. In 1987, damage was evaluated on 21 July on five trees per block. Twig damage was recorded as the number of affected twigs seen during a two-minute examination of each sampled tree. Fruit damage was recorded as the number of fruit damaged in a sample of 20 fruit per tree. Damage was assumed to be caused by *G. molesta*.

RESULTS AND DISCUSSION

Olfactory communication in both *G. molesta* and *G. prunivora* was totally disrupted for the entire growing season as determined by pheromone trap captures in both 1986 and 1987 (Table 1). Relatively low population densities of *Grapholita* spp. were present in both orchard blocks; population suppression by olfactory disruption is more effective at low population densities because males experience increased difficulty in finding the lower number of females (Kydoneius and Beroza 1982). Population densities of *Grapholita* spp. and twig damage may have been

reduced by early hardening off of terminals induced by drought (Schoene et al. 1937). The years of the study were drought years [1986: 5.9, 7.2, 6.6, 6.5, and 11.2 cm rainfall in April through August, respectively; 1987: 23.2, 6.3, 3.0, 10.9, and 3.0 cm in the same months (unpublished data)].

Table 1. Seasonal trap capture data for *Grapholita molesta* and *G. prunivora* in peach orchards at Batesville, VA.*

	Mean *G. molesta* per trap		Mean *G. prunivora* per trap	
	1986	1987	1986	1987
Pheromone-treated	0.0(0.0)	0.0(0.0)	0.0(0.0)	0.0(0.0)
Insecticide-treated	40.7(7.4)	64.5(9.8)	46.6(10.5)	43.2(3.1)

* Numbers in parentheses represent standard deviations.

No fruit or twig damage was observed in the pheromone-treated block in either year (Table 2). Fruit damage was also very low in the parathion-treated control block in both years. More affected twigs were seen in the control block than in the pheromone treatment in 1986. Fewer damaged twigs were seen on 21 August than on 3 July. This is probably due to the majority of this damage being caused by larvae of earlier generations, and after weathering and plant development some affected twigs were not clearly recognizable as such. Random variation may also present a factor in the discrepency between the two dates since different trees were selected for each sampling date. It was concluded that disruption of olfactory communication was at least as effective as a conventional insecticide program.

Table 2. Damage data for *Grapholita molesta* in a peach orchard at Batesville, VA.*

1986				1987	
Infested twigs per tree		Damaged fruit per 20 fruit		Infested twigs	Damaged fruit per 20
3 Jul	21 Aug	3 Jul	21 Aug	per tree	fruit
Pheromone-treated					
0.0(0.0)	0.0(0.0)	0.0(0.0)	0.0(0.0)	0.0(0.0)	0.0(0.0)
Insecticide-treated					
6.0(2.3)	2.0(0.6)	0.0(0.0)	0.1(0.1)	2.0(1.1)	0.0(0.0)

* Numbers in parentheses represent standard deviations

The source of the dispensers (Pacific Biocontrol, 1121 L. Street, Sacramento, CA 95833) directs that ropes be applied prior to first moth emergence and replaced 90 days later. Cost of ropes is estimated to be $225-240/ha for two applications. This cost would be reduced to $112-120/ha by using a single application, thus enhancing economic feasibility. The present study used a single placement to effect season-long disruption. Insecticide costs for seasonal control are $116, 52, or 90/ha for azinphosmethyl 50W, parathion 25W, and carbaryl 50W, respectively [based on seven insecticide applications recommended against *G. molesta* (Virginia 1987)]. Current assessments of economic feasibility of the mating disruption technique vary (Rothschild 1975; Vickers et al. 1985). The use of mating disruption for control of such low population densities of *G. molesta* can be justified since disruption is more effective against low population densities and will more effectively maintain this pest at low levels.

It is important to note that the above costs for pheromone and insecticide do not include labor or other related costs (fuel, machinery, etc.). In this study, the application of ropes was timed to precede flight of *G. molesta* and occurred in the bloom to petal-fall period, a time when laborers are not normally in the orchard. Labor for such an application would cost $12-15/ha (at a $4-5/hour base). However, first-generation *G. molesta* larvae damage only the shoot tips and this damage is not considered important except in trees three years old or younger (Bobb 1973). Therefore it may be possible to wait until slightly later in the season, that is until fruit-thinning operations when workers give individual attention to each tree. This would greatly reduce the labor cost.

Insecticide application still retains the benefit of controlling other pests, such as catfacing insects and plum curculio in the early part of the season and Japanese beetle later in the season. However, this pheromone technology may lend flexibility to orchard pest management programs by eliminating the need to monitor and control a direct pest, thereby allowing orchardists to concentrate on other insect pests. An alternative emphasis of a pheromone suppression program centers on young orchards. An early placement of ropes could significantly reduce twig damage relative to insecticide applications. Twig damage is more important in young orchards than in bearing orchards because this is when initial tree training occurs. Actively-growing twigs are present on such young trees for a greater proportion of the growing season than on mature trees (Schoene et al. 1937). Lack of fruit in young orchards eliminates the need for insecticides directed against fruit pests, so an early placement of pheromone dispensers could greatly reduce total insect control costs in addition to elimination of twig damage.

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CORRELATION OF CROP AGE WITH POPULATIONS OF SOIL INSECT PESTS IN FLORIDA SUGARCANE[1]

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Abstract: Correlation between crop age and populations of soil insect pests was measured in 18 commercial sugarcane fields in Florida. *Melanotus communis* (Gyllenhal) was the most abundant and largest wireworm species found in these fields. Wireworm populations were not significantly correlated (r = -0.18) with crop age (years). *Ligyrus subtropicus* (Blatchley) was the most abundant and largest grub species found in these fields. In contrast to wireworms, grub populations were significantly correlated (r = 0.74) with crop age (years). Data presented in this study indicate the importance of old sugarcane fields in harboring grub populations and these data also suggest reduced ratooning of Florida sugarcane as a possible means of grub control.

Key Words: Sugarcane, Elateridae, Scarabaeidae

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Sugarcane is Florida's most valuable field crop and is primarily grown in the Everglades area of southern Florida. Currently this crop is attacked by two major groups of soil insect pests, wireworms (Family Elateridae), which cause damage to newly planted sugarcane (Hall 1985) and several species of grubs (Family Scarabaeidae). Of these grubs, *Ligyrus subtropicus* (Blatchley), the primary species of economic importance (Gordon and Anderson 1981), in areas of high infestation has been shown to reduce harvest of sugarcane by 39% (Sosa 1984).

After being planted, Florida sugarcane is harvested at ca. yearly intervals from October to April. Florida sugarcane is allowed to ratoon after each harvest and fields remain in production for several years before being replanted. Each ratoon crop is in general less productive than the previous crop and replanting occurs when sugarcane yield drops to a level unacceptable to the producer (Alvarez et al. 1979). The objective of this study was to determine the correlation between crop age and populations of grubs and wireworms in Florida sugarcane since these date are not currently available for these important pests.

MATERIALS AND METHODS

Eighteen commercial sugarcane fields were sampled on farms in southern Florida. The 18 fields were selected from different areas and different sugarcane growers in order to obtain a representative sample of the grub and wireworm populations in Florida sugarcane fields. Fields were also selected to represent a range of crop ages so that some fields were 1 year old, 2 years old, etc. up to 5

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years old. All sugarcane plants in each field were the same age since each field was planted in its entirety at the same time. Mature sugarcane is a very difficult crop in which to sample insects (Southwood 1969) and Florida sugarcane may be 3 to 4 m high before harvest. Therefore, all fields were sampled after harvest for easy access. Nine fields were sampled from October, 1985 to May, 1986 and nine fields were sampled from October, 1986 to May, 1987. Since digging and sorting grubs and wireworms is very labor intensive, the 14 month period was required to obtain a sufficient number of fields sampled for statistical analysis. Each field was sampled once and all sampling for each field was completed in 1 to 2 weeks. Each field was 5.3 to 16.2 ha and fields were sampled one at a time. Since the objective of this study was to correlate crop age (years) with grub and wireworm populations, fields were sequentially sampled so that field age (years) was randomized within the 14 month period to reduce bias due to monthly changes in population abundance. Sugarcane plants were used for sample units since most soil-dwelling pests of sugarcane become aggregated around sugarcane plants (Southwood 1969) as also occurs with Florida sugarcane grubs (Cherry 1984). One hundred sugarcane plants were randomly sampled for soil insects from each field. Sugarcane plants may merge together, especially in older fields, making separation of a plant difficult. Thus, each plant was defined by a 50 × 50 cm area and the soil was dug by hand to a depth of 20 cm. The sugarcane plant and the soil were examined for grubs and wireworms in the field. After collection, insects were taken back to a laboratory, frozen, and later identified.

Skips (denuded areas) in sugarcane fields may appear due to removal of sugarcane plants. Factors responsible for skips are field traffic, harvesting equipment, cold damage, and other factors (Eiland and Lyrene 1977). Since soil insects aggregate under sugarcane plants (Southwood 1969), skips could bias sampling since soil insects would aggregate under remaining sugarcane plants. Therefore, sugarcane plants in 100 m of plant row in the center of each field were also counted to determine if increasing crop age reduced the number of sugarcane plants left in the fields. Sugarcane plants were counted by walking along the row and counting plants, each plant was defined as a 50 × 50 cm area in the row.

Data were analyzed by regression analysis using a general linear models (GLM) procedure (SAS Institute 1985). Soil insect data were analyzed by using the total number of wireworms or grubs collected per field as the dependent variables and crop age in years as the independent variable. Crop age in years refers to the time interval from when the sugarcane was planted until the time the sugarcane field was sampled for soil insects. Fewer old fields (i.e. four and five year old) were sampled since these old fields are normally disced and replanted shortly after harvest. Data on skips were analyzed by using sugarcane plants/100 m as the dependent variable and crop age in years as the independent variable. Analysis of variance showed quadratic models did not add significantly ($P > 0.05$) to the predictability of linear models. I used the simpler linear models in all data analysis.

RESULTS AND DISCUSSION

The number of sugarcane plants/100 m versus crop age (years) were described by the equation, $y = 119.5 - 4.1x$ where y equals sugarcane plants/100 m and x equals crop age in years. The low correlation coefficient of -0.23 for this equation

was not significant ($P > 0.05$). These data indicate that use of sugarcane plants as sample units was not biased by field age since the number of sugarcane plants did not significantly decrease in older fields in this study.

A total of 3,854 wireworms was collected in the 18 fields. Of these wireworms, 63.5% were *Melanotus communis* (Gyllenhal), 28.6% were *Glyphonyx bimarginatus* Schaeffer, 6.9% were *Conoderus* spp., and 1.1% were unknown. Ingram et al. (1938) described *M. communis* as the most abundant wireworm injuring sugarcane in the Everglades, but provided no data for this statement. Hall (1985) has reported on damage by *M. communis* to sugarcane during germination and early growth. In this study, *M. communis* was the most abundant and largest commonly found wireworm in Florida sugarcane fields. Wireworm abundance was quite variable between fields ranging from a high of 722 wireworms found in a field with 2 year old sugarcane to a low of 30 wireworms also found in a field with 2 year old sugarcane (Fig. 1). Wireworm populations were not significantly correlated ($P > 0.05$) with crop age as indicated by the low correlation coefficient of -0.18. The presence of moderate levels of wireworm populations in 1 year old sugarcane shows that residual wireworm populations are still present in newly planted fields and/or fields are rapidly reinfested by the pests.

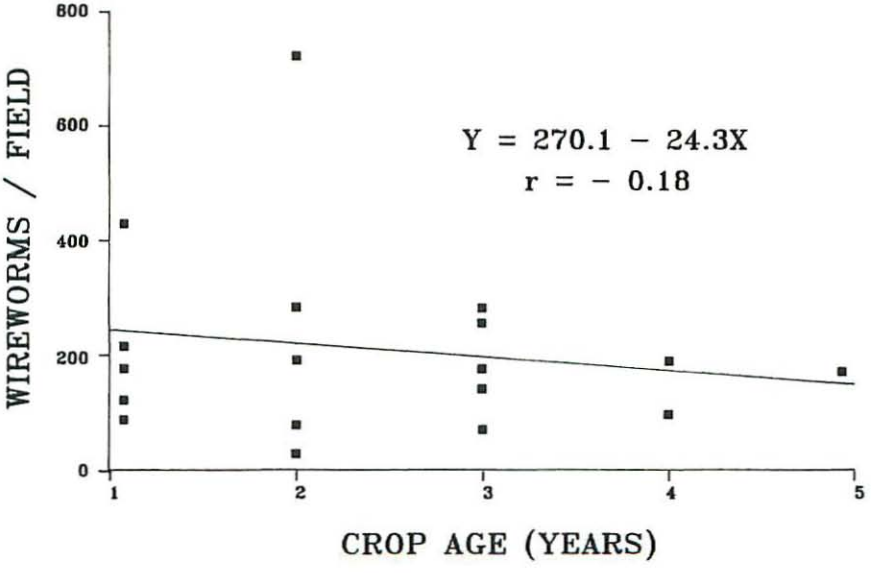

Fig. 1. Correlation of total number of wireworms collected per field with crop age (years) in 18 sugarcane fields in Florida. One hundred sugarcane plants were sampled in each field.

A total of 1,576 grubs was collected in the 18 fields. Of these grubs, 70.6% were *L. subtropicus*, 24.4% were *Cyclocephala parallela* Casey, 2.8% were *Anomala marginata* (F.), and 2.2% were *Phyllophaga latifrons* (LeConte). As noted earlier, Gordon and Anderson (1981) reported that *L. subtropicus* was the primary grub

species of economic importance in Florida sugarcane, but provided no data to support this statement. Sosa (1984) showed that this grub could reduce sugarcane yield by 39% in Florida. Grub abundance was quite variable between fields ranging from a high of 352 grubs found in a field with 3 year old sugarcane to a low of no grubs found in a field with 1 year old sugarcane (Fig. 2). In contrast to wireworms, grub populations were significantly correlated ($P < 0.05$) with crop age ($r = 0.74$). Also in contrast to wireworms, grub populations were very low in all 5 one year old sugarcane fields. These low grub populations show that few grubs are usually present in newly planted fields and fields are only slowly reinfested by the pests. In Florida sugarcane, each ratoon crop is generally less productive than the previous crop (Alvarez et al. 1979). Data in Fig. 2 suggest that increasing grub populations in older sugarcane fields is a factor in the reduced yield of older sugarcane fields. These data also suggest that grub damage can be reduced by reducing the number of years that a crop is left in a ratoon program. Reduction of ratoon crops for grub control in sugarcane has been suggested by Van Dine (1913) in Puerto Rico. Pierce (1930) and Saplala (1959) in the Philippines condemned leaving old grub infested sugarcane plants undisturbed. Mungomery (1930) considered that long ratooning fostered grub infestations in Australian sugarcane. In Burma, Ghosh (1937) found fallowing for one year and taking only one ratoon crop the only practical control against grubs in sugarcane. More recently, Veerish (1984) reported that ratoon crops must be avoided as part of the integrated management of white grubs in sugarcane fields in India. Data in this study show the importance of old sugarcane fields in harboring grub populations and suggest reduced ratooning of Florida sugarcane as a means of grub control.

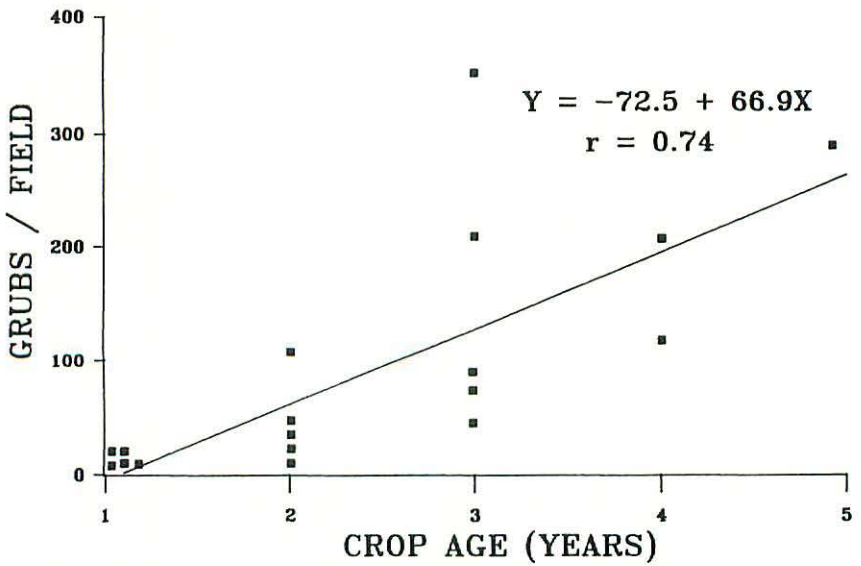

Fig. 2. Correlation of total number of grubs collected per field with crop age (years) in 18 sugarcane fields in Florida. One hundred sugarcane plants were sampled in each field.

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THE EFFECTS OF PREBIOASSAY TREATMENT OF RESISTANT AND SUSCEPTIBLE CORN SILKS ON THE DEVELOPMENT OF THE CORN EARWORM[1] AND FALL ARMYWORM[1,2]

B. R. Wiseman[3] and D. J. Isenhour[4]

Abstract: The effects of handling resistant or susceptible corn silks during the drying process were evaluated for their effects on the biology of the corn earworm, *Heliothis zea* (Boddie), and the fall armyworm, *Spodoptera frugiperda* (J. E. Smith). Subtle differences in weights of larvae, developmental time, and weights of pupae among the drying processes for each silk type were measured. However, in each case and for each insect species, large differences were detected between the measurement parameters for resistant and susceptible corn silks, regardless of handling and drying process treatment. Silks that were directly oven- or freeze-dried showed the best consistency for biological activity. For ease of handling and processing and its biological activity on the insects, the direct oven-drying process will be adopted.

Key Words: Bioassay, *Heliothis zea*, *Spodoptera frugiperda*, Antibiosis, *Zea mays*.

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Silks of some of the corn (*Zea mays* L.), particularly those of 'Zapalote Chico', have been found to be resistant to both fall armyworm (FAW), *Spodoptera frugiperda* (J. E. Smith), and corn earworm (CEW), *Heliothis zea* (Boddie) (Wiseman and Widstrom 1986; Wiseman and Wilson 1987). Although maysin, a flavone glycoside, has been identified as an antibiotic factor in the silks of 'Zapalote Chico', the levels in other corn lines, as determined by spectrophotometeric analysis of freeze-dried silks, have not correlated with CEW biological activity (Wiseman et al. 1985). To date, laboratory bioassays have been required to identify silks of corn lines that possess antibiosis. Thus, because of the volume of materials that must be assayed, techniques had to be developed to preserve the silks for assays at a later date while still maintaining biological activity.

The laboratory bioassay (Wilson et al. 1984; Wiseman and Wilson 1987), which utilizes the pinto bean diet (Burton 1967) and fresh or dry silks, pollinated or nonpollinated, has been an efficient and effective method of assaying for resistance to these insect species. This bioassay is extremely effective for determining if antibiosis is one of its resistance mechanisms. In the past we have experienced some slight difficulties in obtaining uniform weights of larvae from test to test. Silks that were handled differently during the drying process have been implicated.

Wilson et al. (1984) showed that the use of 80 g of fresh ZC (Zapalote Chico) silks/diet was required to equal the effects of feeding fresh ZC silks directly to CEW. In addition, they reported the greatest differences in 8-d weights of larvae on SEG (Stowell's Evergreen) vs. ZC diets occurred when the silks were processed immediately and added to the diets. However, to evaluate cultivars of different maturities the silks must be frozen or dried for periods of time.

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In this study, we varied the handling of the silks of 'Zapalote Chico' (resistant) and 'Stowell's Evergreen' (susceptible) in either the freeze or oven-drying process before they were fed to FAW or CEW neonates to detect if the silk-handling processes grossly affected insect growth and development.

MATERIALS AND METHODS

The CEW and FAW used in this study were obtained from colonies maintained at the Insect Biology and Population Management Research Laboratory, Tifton, GA (Perkins 1979; Perkins et al. 1973).

Two corn genotypes, a susceptible, 'Stowell's Evergreen' (SEG) and a resistant, 'Zapalote Chico 2451# (PC3)' (ZC), were selected for evaluation. The corns were grown in bulk plantings at Tifton, Georgia using agronomic practices common to the area.

Open-pollinated silks of each cultivar were harvested when the silks had emerged for 2 d. Silks were excised to the ear tip, removed from the husk channel, and bulked. The bulked silks for each frozen treatment were placed in plastic bags and placed in a freezer. Processing treatments for the silks were as follows:

1. Silks fresh from the field, frozen at -10°C in a freezer for 2 wk, thawed at room temperature for 2 to 3 hr, and freeze-dried (See Table 1, FTFD).
2. Silks fresh from the field, frozen at -10°C in the freezer for 2 wk, thawed, refrozen at -10°C in the freezer for 2 wk, and freeze-dried (FTFFD).
3. Silks fresh from the field, frozen at -10°C in the freezer for 2 wk and freeze-dried (FFD).
4. Silks fresh from the field and freeze-dried (FD).
5. Silks fresh from the field and oven-dried at 41°C for ca. 2 wk (OD).
6. Silks fresh from the field, frozen at -10°C in the freezer for 2 wk, oven-dried at 41°C for 2 wk (FOD).

Each silk type was bulked by cultivar and treatment, then ground to a fine powder (1.0 mm screen) using a Cyclotec® (Fisher Scientific) TC 1093 sample mill. The powdered silks were processed into a diluted pinto bean diet of 250 ml bean diet:100 ml distilled water at a concentration of 45 mg silks per 1 ml of dilute diet. Diets were dispensed into 36 one-oz. plastic diet cups at ca. 8 ml/cup for each insect species and diet treatment. The treatment diets were allowed to solidify for ca. 2 h, then one neonate FAW or CEW was placed in each cup which was capped with a paper lid. The experiments were held under a constant temperature of 25 ± 2°C and an RH of 80 ± 5% with a photoperiod of 14:10 LD.

Each experiment (CEW or FAW) was arranged as a randomized complete block design with 18 replications with 2 cups/replicate. Data recorded were weights of larvae (FAW 10 d, CEW 9 d), developmental time to pupation, and weights of pupae. Replicate means were used for the analysis. Data were subjected to an analysis of variance (SAS Institute 1985) and means were separated by Waller-Duncan k-ratio t test; k ratio = 100; $P < 0.05$ (Waller and Duncan 1969).

RESULTS AND DISCUSSION

Corn earworm larvae fed all the ZC diet mixtures weighed consistently and significantly less than those fed the SEG diet mixtures (Table 1). The CEW larvae

fed ZC freeze-dried and ZC frozen freeze-dried silks weighed significantly less than all other larvae fed ZC silk treatments except ZC oven dried. In addition, larval developmental time revealed clear differences between larvae fed SEG and ZC. The larval developmental time for those fed on each of the ZC silk treatments were significantly different from each other. The differences in weights of pupae for larvae fed the various SEG or ZC silk diet treatments were not as distinct. This may be due in part to the extended time of development that permits compensation for previous weight differences in larvae. However, the pupae from larvae fed the freeze-dried material in ZC weighed significantly less than the remaining pupae from the ZC silk treatments. It is apparent that freezing and thawing before or during the drying process alters the biological effect on growth and development of CEW larvae that were fed either the resistant or susceptible silks. The magnitude of differences due to silk alterations are greatest for the ZC treatments, probably because smaller numerical values occur on ZC treatments than for those from the SEG silk treatments.

Table 1. Mean weights of larvae, developmental time, and weights of pupae for corn earworm fed various processes of resistant (ZC) and susceptible (SEG) silks in meridic diets.*

Treatment	Wt (mg) of larvae 9 d	Developmental time (d)	Weights of pupae (mg)
Bean	583a	15a	517b
1. SEG-FTFD	562a	15a	537ab
2. SEG-FTFFD	544a	15a	565a
5. SEG-OD	424b	15a	530ab
3. SEG-FFD	354c	15a	503bc
4. SEG-FD	308cd	16a	537ab
6. SEG-FOD	280d	16a	531ab
6. ZC-FOD	178e	18b	472cd
2. ZC-FTFFD	148e	20c	452de
1. ZC-FTFD	91f	22d	418ef
5. ZC-OD	50fg	25e	387f
4. ZC-FD	19g	32g	295g
3. ZC-FFD	17g	28f	406f

* Means within a column not followed by the same letter are significantly different. (MSD = 48-weights of larvae; 1.4-Development time; 40-weight of pupae). $P < 0.05$ (Waller and Duncan 1969). Treatments were as follows: 1. Silks fresh from the field, frozen at -10°C for 2 wk, thawed, and freeze-dried (FTFD); 2. Silks fresh from the field, frozen at -10°C for 2 wk, thawed, refrozen at -10°C for 2 wk, and freeze-dried (FTFFD); 3. Silks fresh from the field, frozen at -10°C for 2 wk and freeze-dried (FFD); 4. Silks fresh from the field, freeze-dried (FD); 5. Silks fresh from the field, oven-dried at 41°C for ca. 2 wk (OD); 6. Silks fresh from the field, frozen at -10°C for 2 wk, oven-dried at 41°C for 2 wk (FOD).

The differences in weights and larval developmental time of FAW fed the various silk diet treatments were significantly different for larvae fed SEG compared with those fed ZC (Table 2). However, very little difference existed within each silk type for weights of larvae. Also, no differences were found for larval developmental time for those fed SEG silk diets. But larvae fed ZCOD silks required longer to pupate than those fed ZCFTFD, ZCFTFFD, and ZCFOD. The larvae fed ZCFTFD, ZCOD, ZCFD and ZCFFD formed pupae that weighed significantly less than those from any other silk diet treatments. As with the CEW,

it is apparent that different processes or handling of silks before and during the drying process affects the biological responses measured for the FAW. The differences seemed to be greater for ZC than SEG. But regardless of the treatments, if compared individually, the biological responses for larvae fed SEG vs. ZC were of a large enough magnitude to significantly measure the differences consistently between the resistant and susceptible. However, these results may not be applicable if silks with lesser amounts of antibiosis resistance are used.

Table 2. Mean weights of larvae, developmental time, and weights of pupae for fall armyworm fed various processes of resistant (ZC) and susceptible (SEG) silks in meridic diets.*

Treatment	Wt (mg) of larvae 10 d	Developmental time (d)	Weights of pupae (mg)
Bean	347b	15a	252c
1. SEG-FTFD	413a	14a	271bc
2. SEG-FTFFD	382ab	15a	306ab
5. SEG-OD	246c	15a	277bc
4. SEG-FD	232c	15a	263bc
6. SEG-FOD	231c	16a	330a
3. SEG-FFD	210c	15a	287bc
6. ZC-FOD	55d	20b	258c
2. ZC-FTFFD	52de	21b	257c
1. ZC-FTFD	28de	24c	195d
5. ZC-OD	24de	27d	172d
4. ZC-FD	19de	26cd	186d
3. ZC-FFD	11e	25cd	184d

* Means within a column not followed by the same letter are significantly different. (MSD = 42-for weights of larvae; 2-for development time; and 44-for weight of pupae). $P < 0.05$ (Waller and Duncan 1969). Treatments were as follows: 1. Silks fresh from the field, frozen at -10°C for 2 wk, thawed, and freeze-dried (FTFD); 2. Silks fresh from the field, frozen at -10°C for 2 wk, thawed, refrozen at -10°C for 2 wk, and freeze-dried (FTFFD); 3. Silks fresh from the field, frozen at -10°C for 2 wk and freeze-dried (FFD); 4. Silks fresh from the field, freeze-dried (FD); 5. Silks fresh from the field, oven-dried at 41°C for ca. 2 wk (OD); 6. Silks fresh from the field, frozen at -10°C for 2 wk, oven-dried at 41°C for 2 wk (FOD).

In mixing the diets, the silks of the oven-dried materials are easier to handle, especially for the ZC silks. Freeze-dried silks usually required more water in mixing to obtain the same diet consistency. Since the oven-dried silks did not deviate drastically from the freeze-dried silks in their biological activity, we prefer to use the oven-dried silks in both of our insect bioassays. The process of oven drying permits small or large amounts of silks to be dried easily and conveniently at one time. But very few freeze driers have the capacity to handle the quantity of material that we have to process. In addition, the storage of oven-dried silks in a freezer is easier because they are not as bulky. Since the biological impact on these two insects is not altered by the oven-dried silks, there should be little if any biochemical differences between oven or freeze-dried silks. Thus, for our purposes we will continue to bioassay oven-dried silks to determine relative levels of antibiosis factors.

ACKNOWLEDGMENTS

We thank J. L. Skinner and Charles Mullis for their technical assistance in this study.

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ERRATUM

In the article "Distribution and Monitoring Lesser Mealworms, Hide Beetles, and Other Fauna in High-Rise Caged-Layer Poultry Houses" by Stafford, et al. which appeared in the Volume 5, Number 2, April 1988 issue of *Journal of Agricultural Entomology* on pages 80-101:

Line 4 on page 89 should read "high-rise, caged-layer poultry houses. Both species normally live in the"

Line 12 on page 90 should read "corrugated cardboard inserted into sections of polyvinyl chloride (PVC) pipe were "

Line 11 on page 93 should read "sample size formulae may be given in terms of their parameters (Karandinos 1976)."

Line 1 on page 94 should read "(Table 1). There was a significant difference in the distribution of lesser mealworm."

Line 1 of footnote to Table 1 on page 94 should read "* Means along the manure profile for all species except *L. campestris* were significantly different by"

Line 2 of footnote to Table 2 on page 96 should be "protected LSD (α = 0.05). Steel and Torrie (1980)."

Line 5 from bottom of page 98 should be "(1984) found only ca. 10 traps were required at at high beetle densities ($\geq$ 350 beetles) for a"

Last line on page 98 should be "rise houses than the turkey and broiler houses. In contrast, beetle"

NOTE: These are not author's errors. The author had marked these changes in the final proof, and the correction was made on the final galley; however, when the final typeset copy was produced, the old disk was inadvertently utilized. The editor regrets any inconvenience caused by these errors.

PREDICTING *HELIOTHIS* SPP. OVIPOSITION PEAKS USING TEXCIM[1]

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Abstract: The Texas Cotton-Insect Model (TEXCIM) was used to predict the timing of oviposition peaks of *Heliothis* spp. in South Carolina. TEXCIM's predictions were compared with scouting report data collected over multiple locations and years. The model's predictions were within 1 day of actual peak oviposition reported by field scouting.

Key Words: Model, oviposition, cotton, bollworm.

J. Agric. Entomol. 5(4): 253-256 (October 1988)

The Texas Cotton-Insect Model (TEXCIM) (Hartstack and Sterling 1988) contains a fleahopper model (Hartstack and Sterling 1986) and the *Heliothis* spp. model, MOTHZV (Hartstack and Witz 1982). TEXCIM was designed to help farmers make management decisions concerning the need for insect control and to estimate the severity of future pest problems. To estimate future pest problems, information from scouting reports are entered into the model and short-range predictions are made. However, these predictions must compare favorably with historical data, collected within a region, before any confidence can be placed in the model's predictions. In this study we compared TEXCIM's predicted time of peak oviposition by *Heliothis* spp. with actual ovipositional peaks in South Carolina cotton fields.

MATERIALS AND METHODS

Data used to validate TEXCIM's ability to predict the days of peak oviposition were from South Carolina cotton scouting reports from 1979-1987 in Lee County, and from 1986 in Dillon and Sumter Counties. Data from Lee County were collected by scouts hired by the Wateree Association and trained by Clemson University Extension Service. In Sumter and Dillon Counties, data were collected by scouts hired through the Clemson University Extension Cotton Pest Management project. A random scouting technique was used at all locations. Scouts examined 100 plants once each week for *Heliothis* spp. eggs in each 8 ha of cotton. Multiple fields were examined each year, and data collected in fields which had at least two oviposition peaks were used in this study.

[1] In cooperation with the South Carolina Agricultural Experiment Station. This paper reports the results of research only; mention of a commercial or proprietary product does not constitute an endorsement for its use by the USDA. Accepted for publication 19 September 1988.

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Weather data needed to run TEXCIM are daily rainfall, and minimum and maximum temperatures. These data were obtained from the weather station at the USDA-ARS, Coastal Plains Soil and Water Conservation Research Center at Florence, S.C. The scouted cotton fields were within 30 miles of the weather station.

To test TEXCIM's ability to predict the time of the next oviposition peak, we entered egg counts up to the first oviposition peak. The model was run for the full season and the date of the next oviposition peak was recorded. A straight line was fitted to the predicted data versus observed data using PROC REG (Statistical Analysis Systems 1985).

RESULTS AND DISCUSSION

Observed and predicted oviposition peaks are shown in Table 1. Six of the 10 predictions were within 1 d of the observed oviposition peak, with the largest difference being 5 d. The model's prediction differed from observed data an average of 1.9 d.

Table 1. Comparison of the predicted and the observed day of peak oviposition by *Heliothis* moths.

			Second Peak	
Location	Year	1st Peak	Observed	Predicted
Lee Co.	1979	6/19	7/24	7/28
Lee Co.	1980	6/17	7/22	7/22
Lee Co.	1982	6/23	7/27	7/27
Lee Co.	1983	6/22	7/27	7/28
Lee Co.	1984	6/20	8/01	7/27
Lee Co.	1984	6/14	7/18	7/19
Lee Co.	1986	6/18	7/23	7/19
Dillon Co.	1986	6/13	7/8	7/12
Sumter Co.	1986	6/19	7/17	7/17
Lee Co.	1987	6/24	7/29	7/29

To test the validity of the model's prediction, we graphed the predicted versus observed values and fitted a straight line to these data (Fig. 1). If the model's predictions were perfect, regression analysis would result in a line with a slope of 1 and an intercept of 0. The slope of this regression line was not significantly different ($P > 0.05$) from 1 ($b_1 = 0.762 \pm 0.118$), and the intercept ($b_0 = 48.58 \pm 24.09$) ($R^2 = 0.84$) was not significantly different ($P > 0.05$) from 0. Regression analysis showed that the model's predictions were 0.23 d earlier than observed.

Predicting the future occurrence of an oviposition peak would be useful in scheduling irrigations and fertilizer applications. Slosser (1980) found that irrigating cotton during peak oviposition resulted in increased oviposition and larval survival. He recommended not irrigating cotton for 7-10 days before and 3-4 days after peak oviposition to decrease the potential for damaging bollworm infestation. Fletcher (1941) found a high correlation between percent moisture in the plant terminals and the number of eggs and larvae. Fye and Surber (1971) reported that increased humidity could result in higher egg hatch during periods of high temperatures. High nitrogen fertilization can also result in increased bollworm numbers and damage (Adkisson 1958).

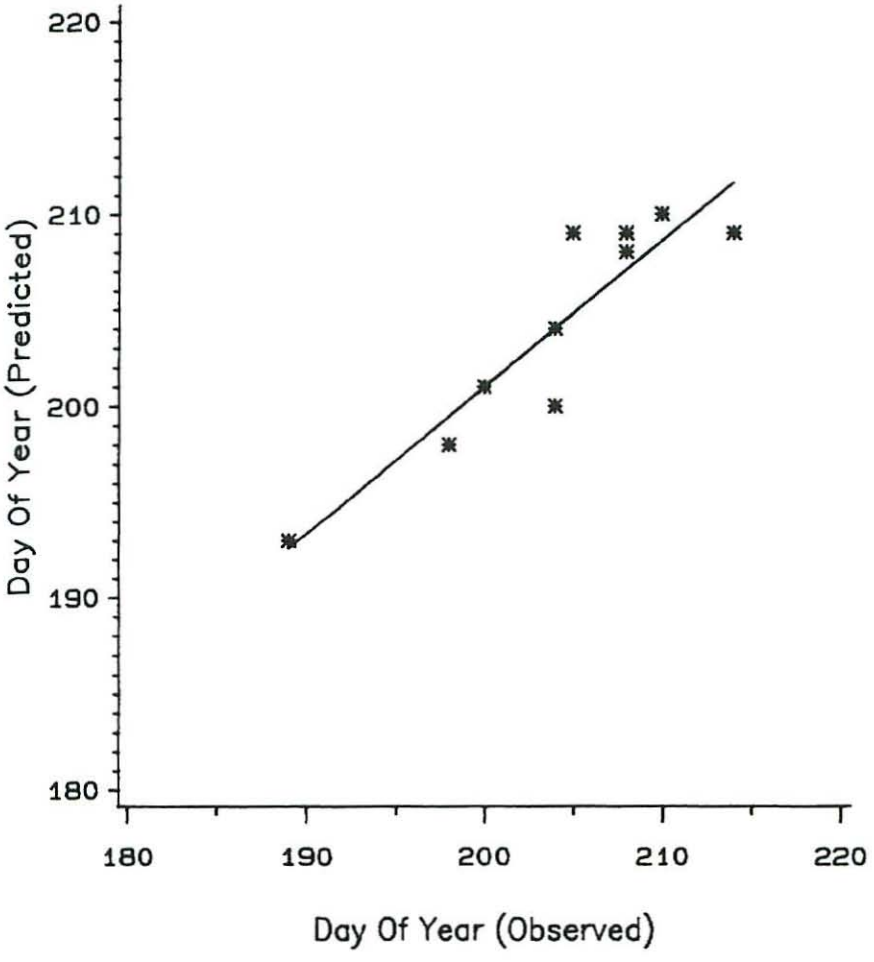

Fig. 1. Comparison of TEXCIM's predicted time of peak oviposition with observed data (*). Solid line represents regression line (Y = 48.58 + (0.762*X)) (R^2 = 0.84).

The TEXCIM model was able to predict within 1 day the occurrence of *Heliothis* oviposition peaks in South Carolina cotton fields; however, TEXCIM was not able to predict the number of eggs. To predict the number of eggs laid, TEXCIM needs information concerning the number of immigrating moths, and the number of different insect predators present in the cotton field. Since we did not have this information, we could not adequately test TEXCIM's ability to predict the number of eggs laid. However, predicting the population trends would still be useful in making management decisions.

The cotton crop model GOSSYM (Baker et al. 1983) with the expert system COMAX (Lemmon 1986) simulates the effect on crop yield of various irrigation and fertilizer application schedules; however, it does not consider the effect of these practices on insect development. Linking a *Heliothis* model, such as TEXCIM,

to GOSSYM would provide farmers with additional information needed to decide if the benefits of irrigation and fertilization outweigh the possible risk of increased *Heliothis* infestation.

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HOP LEAF PHENOLICS AND RESISTANCE TO THE TWOSPOTTED SPIDER MITE[1]

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Abstract: Leaves from ten cultivars of hop, *Humulus lupulus* L., were analyzed for five classes of phenolic compounds as well as for 12 phenolic aglycones. Twospotted spider mites, *Tetranychus urticae* Koch, on variety 'BOR-704' had significantly lower intrinsic rates of increase, r_m's than on 'Eroica', 'Nugget', and 'Cascade' in both trials of the experiment, indicating a difference in resistance among cultivars. Amounts of leaf phenols varied significantly among cultivars. However, the correlations of *T. urticae* r_m's with phenolic concentrations were not consistently significant between the two trials of the experiment. Our study indicates that phenols are not major factors in the resistance of hops to *T. urticae*.

Key Words: Twospotted spider mite, *Tetranychus urticae*, hop, *Humulus lupulus*, phenols, host plant resistance, life tables, intrinsic rate of increase.

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The twospotted spider mite, *Tetranychus urticae* Koch, is a serious pest of hop, *Humulus lupulus* (L.). It feeds on the leaves and cones (female flowers), reducing quality and yield. Several studies have shown differences in resistance to *T. urticae* among hop varieties (Mayberry 1968; Regev and Cone 1975; Peters and Berry 1980a, b), indicating that it may be possible to develop resistant varieties.

Phenolic compounds are allelochemicals that have been connected with resistance to *T. urticae*. Dabrowski and Rodriguez (1972) found that strawberry leaf phenols added to a synthetic diet reduced ingestion of food, increased repellency of the diet, and increased mortality of *T. urticae*. Larson and Berry (1984) found that as the amount of combined total phenols in peppermint leaves increased, the number of eggs laid by *T. urticae* decreased, dispersal of immatures increased, and development time of immatures lengthened. Hop cultivars that contained higher levels of phenolic compounds were found to be less susceptible to aphids and fungal diseases (Lyashenko et al. 1982; Piotrowski and Milczak 1982). Hop stems, leaves, flowers, and cones have been shown to contain many phenolic compounds (Lebreton 1958; Lyashenko 1977; Verzele 1979; McMurrough 1981; McMurrough et al. 1982). The purpose of this study was to determine if quality and quantity of phenolic compounds in hop leaves are involved in resistance to *T. urticae*. Most previous studies on the role of phenols in plant resistance have measured total phenols. We have taken a more detailed approach by analyzing hop leaves for the amounts of phenols in five classes of phenols and for individual phenolic aglycones.

MATERIALS AND METHODS

Preparation of Hop Leaves for Phenolic Analyses

The cultivars chosen were representative commercial varieties: 'BOR-704' (a selection from Hallertauer), 'Cascade', 'Chinook', 'Eroica', 'Galena', 'L1', 'L8',

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'Nugget', 'Olympic', and 'Willamette'. Twelve leaves were collected from field-grown plants for each of the four replications of each variety on 26 June 1986 for trial 1 and on 24 July 1986 for trial 2. To standardize leaf age as much as possible, we collected the third and fourth leaves proximal to the growing tips of the lateral branches (sidearms). Leaves for phenolic analysis were immediately placed on dry ice and later freeze-dried. Leaf disks, as used in the mite bioassay, were not analyzed for phenols because more leaf material was needed for analysis. The samples were ground in a Wiley mill to pass through a No. 40 screen, placed in sealed glass bottles, and kept in a desiccator at 3°C until analysis.

Extraction of Phenolic Compounds

The freeze-dried material (0.5g per sample) was defatted in a Soxhlet apparatus with chloroform, air dried, and extracted with 70% ethanol for 6 h. The remaining residue was hydrolyzed with 2N HCl at 80°C for 20 h. The alcohol extract and hydrolyzed portion were filtered through Whatman No. 1 filter paper. The analyses were of two basic types: classes of phenolic compounds (total phenols soluble in ethanol, total phenols insoluble in ethanol, ortho-dihydroxy phenols, and flavonols) analyzed by spectrophotometric techniques, and individual phenolic aglycones analyzed by High-Performance Liquid Chromatography (HPLC).

Estimation of Total Phenols

The ethanol extract was analyzed by the method of Singh et al. (1978) to determine the total phenols soluble in ethanol. The method was also used to analyze the hydrolyzed fraction for total phenols insoluble in ethanol. Water was added to 0.5 ml of the extract to make 15 ml of solution, to which 5 ml ferric chloride-potassium ferricyanide reagent (0.3% $FeCl_3 \cdot 6H_2O$ in 0.4N HCl and 0.3% $K_3[Fe(CN)_6]$; 1:1 v/v) was added. After 60 min at room temperature, the readings were taken at 700 nm. Pyrocatechol was used for the standard curve. Combined total phenols were determined by adding the amounts of total soluble phenols to the amounts of total insoluble phenols.

Estimation of Ortho-dihydroxy Phenols

Arnow's (1937) procedure was used to determine the amount of ortho-dihydroxy phenols. The following reagents were added to 5 ml of the ethanol extract in the order given: 1 ml of 0.5 N HCl, 1 ml of Arnow's reagent (10 g $NaNO_2$ and 10 g Na_2Mo_4 in 100 ml H_20), 2 ml of 1 N NaOH and 1 ml of H_2O. The absorbance was read at 520 nm. Pyrocatechol was used for the standard curve.

Estimation of Flavonols

The concentration of flavonols was estimated according to the Christ-Müller method (Strzelecka et al. 1978). One half ml of 0.5% hexamethylenetetramine and 2 ml of $AlCl_3$ solution (2 g $AlCl_3 \cdot 6H_2O$ in 100 ml of 5:95 v/v acetic acid-methanol) were added to 10 ml of the ethanol extract. Acetic acid-methanol (5:95 v/v) was added to make 25 ml. After 45 min, absorbance was measured at 425 nm. Quercetin was used as the standard.

Analysis of Phenolic Aglycones by HPLC

One hundred ml of hop ethanol extract was hydrolyzed with 30 ml of 2N HCl at 80°C for 1 h. The hydrolyzed material was condensed under vacuum at 40°C to

an acidified water phase, filled to 50 ml with H_2O, and extracted three times with 50 ml of ethyl acetate. The combined ethyl acetate phases were dried with 20 g anhydrous sodium sulfate and evaporated to dryness under vacuum at 40°C. The dry residue was dissolved in 5 ml ethanol containing 0.5 mg of flavone (retention time = 14.49 min) as an internal standard. The mixture was shaken and filtered through a 0.45 µm pore size filter. Sample size used in the HPLC was 20 µl. The phenolic aglycones were identified by comparison of retention times of unknowns with those of standards and by addition of standards to the hop extracts. Replicate samples from a variety were pooled for HPLC analysis, resulting in one analysis per variety for each trial.

HPLC analysis was performed using a Versapack C_{18} (10 u particles) 25 cm × 4.0 mm I. D. column (Alltech Assoc., Deerfield, Illinois) at room temperature. Separation was accomplished by gradient elution with solvent A (acetic acid - H_2O, 1:99 v/v) and solvent B (acetonitrile) according to the method of Tamma et al. (1985). The gradient was linear from 20 to 90% of solvent B in 20 min. Column pressure was 8.27 MPa (1,200 psi) and solvent flow rate was 1.5 ml per min. Aglycones were detected at 254 nm.

Mite Life Tables

Leaves were collected from the same growing positions and same plants as the leaves for the phenolic analyses. The two trials were started in late June and late July to correspond to the two collections for phenolic analyses. Ten leaf disks (17 mm diameter) were cut from the leaves of each variety with a cork borer and placed ventral side up on moist cotton in square (9 × 9 cm) plastic culture dishes. The culture dishes with the mites were placed 40 cm below four 40 w, 1.2 m long Phillips F40D daylight fluorescent lamps. Temperature in trial 1 was 24.8 ± 0.48°C during the day and 21.7 ± 0.39°C at night. Photophase was 16 h. In trial 2, daylength was 24 h with an average temperature of 25.4 ± 0.44°C. Daylength in trial 2 was changed without our knowledge by another worker using the growth chamber. The error was discovered about one week after the start of trial 2, so we decided to leave the daylength at 24 h. Leaf disks were changed at about one week intervals, which was before leaves began to turn yellow and before the appearance of mold. Changing leaf disks involved the chance of disturbing or injuring the mites. We tried to minimize handling of the mites while maintaining leaf disk quality. All leaf disks of a variety were placed in one dish. This was done so that a mite that traveled from her disk to another could be put back on her disk without the observer confusing varieties. Mites did not leave their disks very often, but it happened occasionally. Also, recording and mite transferring errors should be reduced by putting one variety in one disk. Conditions were kept as uniform as possible by placing all plates together under the same bank of lights, by watering all dishes with deionized water at the same time and to about the same level, and by changing leaf disks of all varieties on the same day. Disks came from different leaves. Hop plants within a variety are genetically identical since plants are propagated vegetatively.

One adult *T. urticae* female (F1 generation) was placed on each leaf disk for 24 h and removed. All offspring (F2 generation) from the F1 females were left on the leaf disks until sex of the mites could be determined. Then one female and at least one male (so females would be mated) were transferred to a new leaf disk. Eggs (F3 generation) from the F2 females were counted daily. The survival of the F2

females was also recorded. The experiment ran for 20 days starting at the time the F2 eggs were laid. The data were used to construct life tables and calculate the intrinsic rate of increase, r_m (Birch 1948) for individual mites (n = 10 r_m's per variety). The mean r_m of mites on a variety was used as a measure of hop resistance to *T. urticae*. A single sex ratio was calculated for each trial from all F2 mites on all varieties combined.

Statistical Analysis

A test of equality of variances was done to determine if r_m data from the two trials could be pooled. Differences among varieties in r_m's and in amounts of the classes of phenolics were analyzed by completely random design ANOVA and Duncan's Multiple Range Test. Since only one HPLC analysis of phenolic aglycones was made per variety, no ANOVA was done on these data. Relationships between mean amounts of phenols (classes of phenols and phenolic aglycones) and r_m means for a variety were analyzed by linear correlation (Steel and Torrie 1960).

RESULTS AND DISCUSSION

Mites r_m's were higher ($P < 0.01$) on 'Eroica', 'Nugget', and 'Cascade' than on 'Bor-704' in both trials (Table 1), indicating that 'BOR-704' was the most resistant of the above cultivars. The order of the r_m's of mites on the different varieties was about the same between the two trials except that 'L1' and 'L8' were substantially lower in the order in trial 2 and 'Cascade' was much higher than in trial 1. This relative consistency indicates real differences in resistance among varieties. Mite r_m's were higher on all varieties in trial 2 except on 'L1' and 'L8'. Apparently, the resistance of 'L1' and 'L8', which are closely related, increased as the season progressed. The higher r_m's in trial 2 were probably due to the increased temperature in that trial. Variances of r_m's between the two trials were not equal (F = 3.12, 99,99 df, $P < 0.01$). Therefore, data from the two trials could not be pooled. The proportion of females was 0.680 (n = 1,128) in trial 1 and 0.722 (n = 583) in trial 2.

Table 1. Intrinsic rate of increase, r_m*, of *T. urticae* on different hop varieties.

	Trial 1		Trial 2	
Variety	$\bar{x}$	SEM	$\bar{x}$	SEM
Eroica	0.2830a†	0.0065	0.2988ab	0.0059
L1	0.2827a	0.0084	0.2650bcd	0.0177
Nugget	0.2822a	0.0127	0.3150a	0.0042
Olympic	0.2811a	0.0026	0.2972abc	0.0044
L8	0.2692a	0.0084	0.2584d	0.0263
Cascade	0.2535a	0.0090	0.3120a	0.0027
Galena	0.2488a	0.0121	0.2901abcd	0.0063
Willamette	0.2356a	0.0206	0.2767bcd	0.0047
Chinook	0.2346a	0.0198	0.2763bcd	0.0047
BOR-704	0.1474b	0.0434	0.2627cd	0.0036

* $\log_e$ of number of female offspring per female per day.

† Means in a column followed by the same letter are not statistically different ($P > 0.05$; 90 df for each trial; Duncan's multiple range test).

The concentrations of the five classes of phenols varied significantly among cultivars (Table 2). 'Bor-704' was significantly lower in combined total phenols than the other varieties in both trials (Table 2). However, the r_m's of mites on 'BOR-704' were among the lowest in both trials (Table 1). The order of the varieties in the amounts of phenolics in their leaves was about the same within a date among the five classes of phenolics (Table 2). The amounts of phenols were generally higher in late June than in late July. The cultivars with the highest amounts in late June were generally among the highest in late July, and the lowest in late June were usually among the lowest in late July. This indicates that there were consistent differences among varieties.

We found no statistically significant correlations between leaf total phenol concentrations (including soluble and insoluble total phenols) and mite r_m's (Table 3). Although we could not show that the hop leaf total phenols affected the mites, total phenols have been shown to have a negative effect on *T. urticae* on peppermint (Larson and Berry 1984).

Ortho-dihydroxy phenols were positively correlated with r_m in trial 1 (Table 3). This indicates that high levels of ortho-dihydroxy phenols are beneficial components in the diet of *T. urticae*. Since we know of no data on ortho-dihydroxy phenols to support this idea, and because the correlation in trial 2 was not significant, we suspect that the positive correlation is an artifact. At low protein concentrations, the ortho-dihydroxyphenyl groups of polyphenols are the main sites of hydrogen bonding with proteins (McManus et al. 1983). Therefore, the ortho-dihydroxy phenols are usually expected to be important in plant resistance. However, Jordens-Rottger (1979) showed that broad bean leaf surface extracts containing a phenolic compound were attractants for apterous *Aphis fabae* (Scopoli). Moreover, Bernays and Woodhead (1982) showed that *Anacridium melanorhoda* (Walker) survived better and grew faster when certain phenols were added to the food plant.

The concentration of flavonols was not significantly correlated with mite r_m (Table 3). Apparently, the flavonols are not very important in resistance of hops to *T. urticae*.

We found no consistent, significant correlations between phenolic aglycones and r_m (Table 3). However, in the first trial r_m was negatively correlated with p-coumaric acid and was positively correlated with the concentration of quercetin (Table 3). Concentrations of p-coumaric acid as low as 10^{-5} and 10^{-6} M reduced the feeding rate of *T. urticae* on strawberries and higher concentrations prevented feeding for 12 to 24 h (Dabrowski and Rodriquez 1972). Unfortunately, information on the affect of quercetin on *T. urticae* is lacking. Levin (1971) states that the "active" phenolics occur mostly as aglycones but most of the phenolic aglycones in plants are glycosylated and must be hydrolyzed or oxidized for maximum effectiveness. The relationships between r_m and the concentrations of p-coumaric acid and quercetin were not significant in trial 2.

We found no clear qualitative differences in the composition of phenolic aglycones among varieties. HPLC separation of the hydrolyzed hop leaf alcohol extract produced peaks (retention times in min) corresponding to gallic (1.78), chlorogenic (2.63), protocatechuic (2.91), caffeic (4.13), p-coumaric (5.88), and trans-cinnamic (9.25) acids as well as leucocyanidin (1.41), (+)-catechin (3.34), cyanidin (5.37), myricetin (7.13), quercetin (8.84), and kaempferol (10.56) (Table 4). The phenolic aglycones found in the highest amounts were cyanidin, kaempferol, and p-coumaric acid (Table 4). Similar results were obtained by McMurrough (1981) and McMurrough et al. (1982) who identified high amounts of kaempferol, quercetin, cyanidin, (+)-catechin, and (-)-epicatechin, which were released from the flavonol glycosides and flavonol oligomers after acid hydrolysis of the cone extract. Except

Table 2. Mean (± SD) amount (mg per g freeze-dried weight) of classes of phenolic compounds in leaves of different hop varieties.

Variety	Combined total phenols (soluble + insoluble in ethanol)	Soluble in ethanol			Insoluble in ethanol
		Total	Ortho-dihydroxy phenols	Flavonols	Total
Trial 1 - 26 June 1986					
Willamette	33.27 ± 0.48a*	12.46 ± 0.78a	3.30 ± 0.11a	3.92 ± 0.24c	20.81 ± 0.60a
L8	25.71 ± 0.58b	10.37 ± 0.24bc	2.74 ± 0.11bcd	3.30 ± .08de	15.34 ± 0.35b
Olympic	22.52 ± 0.63c	10.72 ± 0.37b	2.78 ± 0.31bc	4.52 ± 0.20b	11.80 ± 0.38c
L1	21.57 ± 0.19cd	9.62 ± 0.40c	2.41 ± 0.04d	2.94 ± 0.06e	11.95 ± 0.58c
Eroica	20.94 ± 0.42d	10.16 ± 0.18bc	2.62 ± 0.22bcd	6.28 ± 0.15a	10.78 ± 0.39cd
Chinook	17.84 ± 0.73e	8.42 ± 0.43d	1.85 ± 0.04e	1.66 ± 0.08f	9.42 ± 0.86ef
Cascade	17.51 ± 0.61e	8.54 ± 0.24d	1.57 ± 0.05ef	3.52 ± 0.27cd	8.99 ± 0.57ef
Nugget	16.89 ± 0.47e	7.15 ± 0.69e	2.83 ± 0.21b	3.25 ± 0.23de	9.86 ± 0.30de
Galena	15.85 ± 0.39f	7.27 ± 1.75e	2.45 ± 0.22cd	2.89 ± 0.43e	8.58 ± 0.90f
BOR-704	13.75 ± 0.47g	5.18 ± 0.13f	1.38 ± 0.05f	3.03 ± 0.08e	8.56 ± 0.58f
Trial 2 - 24 July 1986					
Willamette	19.52 ± 0.28a	8.65 ± 0.48a	2.78 ± 0.13a	4.01 ± 0.13b	10.88 ± 0.42a
Eroica	15.78 ± 0.20b	6.62 ± 0.48b	1.54 ± 0.04de	4.34 ± 0.15a	9.16 ± 0.54b
L8	14.29 ± 0.52c	4.60 ± 0.08d	1.89 ± 0.06b	1.61 ± 0.04f	9.71 ± 0.59b
Olympic	13.72 ± 0.24c	6.70 ± 0.16b	1.88 ± 0.06bc	3.26 ± 0.11c	7.05 ± 0.25c
Nugget	11.27 ± 0.20d	5.53 ± 0.18c	1.95 ± 0.05b	2.19 ± 0.22e	5.74 ± 0.23de
Chinook	11.05 ± 0.60d	5.25 ± 0.28c	2.04 ± 0.09b	2.79 ± 0.23d	5.68 ± 0.55de
L1	10.96 ± 0.25d	4.22 ± 0.45d	1.46 ± 0.04e	2.15 ± 0.07e	6.74 ± 0.22c
Galena	10.61 ± 0.19d	4.07 ± 0.22d	2.05 ± 0.14b	1.24 ± 0.15g	6.55 ± 0.38cd
BOR-704	9.74 ± 0.32e	4.30 ± 0.48d	1.70 ± 0.16cd	2.74 ± 0.23d	5.45 ± 0.34e
Cascade	7.08 ± 0.47f	3.07 ± 0.19e	0.62 ± 0.06f	1.40 ± 0.03fg	4.01 ± 0.57f

* Means in columns followed by the same letter are not significantly different ($P > 0.01$; Duncan's multiple range test). Each trial analyzed separately.

Table 3. Correlation coefficients showing the relationships between hop leaf phenolic and mite intrinsic rate of increase.

Class of Phenolic Compound	Trial 1	Trial 2
Total phenols	0.301	-0.207
Total soluble phenols	0.546	0.036
Ortho-dihydroxy phenols	0.603*	-0.321
Flavonols	0.361	-0.003
Total insoluble phenols	0.149	-0.363
Phenolic aglycones		
Leucocyanidin	-0.304	—
Gallic acid	0.328	0.119
Chlorogenic acid	0.275	—
Protocatechuic acid	-0.065	0.314
(+)-Catechin	0.400	—
Caffeic acid	0.202	-0.434
Cyanidin	0.004	-0.285
p-Coumaric acid	-0.847**	-0.061
Myricetin	0.047	0.171
t-Cinnamic acid	0.593	-0.527
Quercetin	0.786**	0.010
Kaempferol	-0.063	-0.344

*, ** Significant at the $P < 0.05$ and 0.01 levels, respectively, (df = 8).

for 'BOR-704' and 'Olympic', the concentration of p-coumaric acid was higher in trial 2 than in trial 1 (Table 4). 'L1' and 'L8' had the largest relative increases (about 3-fold) in p-coumaric acid in trial 2 while 'Olympic' had the largest decrease (about 9-fold). Quercetin concentrations declined in 7 varieties and increased in 3. These changes in the concentrations of phenolic aglycones through time may partially account for between-trial differences in the correlations with r_m. Changing the relative amounts of phenolics or other allelochemicals over time may help prevent phytophagous animals from developing resistance to these compounds.

We were unable to show a clear relationship between hop leaf phenols and resistance to *T. urticae*. However, we studied only the third and fourth leaves back from the growing tips of lateral branches. Phenols may be important in leaves in other positions or other maturities or in other organs. All of the varieties we studied may have been too low in phenols or the range of concentrations may not have been great enough to show an effect on the mites. Moreover, varietal differences in nutritional suitability for the mites or morphology, such as density of leaf glands or hairs (Peters and Berry 1980b), may have masked the effects of phenols. Leaf disks may not be representative of intact hop leaves, although Dabrowski and Bielak (1978) found that fecundity and survival of *T. urticae* reared on strawberry leaf disks were not significantly different from those of mites on leaves growing on plants. Also, the chemical analysis of phenols may give misleading results for two reasons: 1. The strength of the reactions of the colorimetric reagents with phenols may vary from one phenol to another. 2. Phenolic compounds in plants are extremely complex and varied. If specific phenolic compounds are involved in resistance, as Zucker (1983) believes, these individual compounds would be missed by chemical analyses that almost necessarily involve the destruction of the complex phenolic compounds present in the plant into more simple compounds.

Table 4. The concentrations (mg/g freeze-dried weight) of phenolic aglycones determined by HPLC after acidic hydrolysis of ethanol soluble fraction of hop leaves.

Phenolic compound	Variety									
	BOR-704	Cascade	Chinook	Eroica	Galena	L1	L8	Nugget	Olympic	Willamette
Trial 1, 26 June 1986										
Leucocyanidin	0.942	+*	1.449	+	+	1.811	0.362	+	—	1.201
Gallic acid	0.101	0.171	0.179	0.138	0.165	0.174	0.559	0.151	0.300	0.168
Chlorogenic acid	0.331	0.403	0.423	0.305	0.340	0.559	0.334	0.257	0.658	0.237
Protocatechuic acid	0.241	0.372	0.905	0.290	0.286	0.210	0.270	0.206	0.372	0.106
(+)-Catechin	+	1.127	+	0.414	1.196	1.434	+	—	1.564	+
Caffeic acid	0.555	0.953	2.671	0.523	1.585	1.974	1.837	0.893	1.136	0.407
Cyanidin	3.412	4.301	8.275	4.301	3.909	2.960	3.388	2.717	6.359	2.927
p-Coumaric acid	4.413	1.324	1.446	1.253	1.274	0.135	0.316	0.805	2.200	2.211
Myricetin	0.080	0.071	0.378	0.210	0.045	0.037	0.103	0.126	0.147	0.058
t-Cinnamic acid	0.238	0.508	0.189	0.705	0.501	0.273	0.453	0.732	0.553	0.384
Quercetin	0.081	1.458	0.415	2.325	1.200	1.159	1.922	1.843	1.316	0.444
Kaempferol	3.023	2.805	1.052	5.590	1.492	0.592	1.277	1.097	3.096	2.470
Trial 2, 24 July 1986										
Leucocyanidin	+	+	+	+	+	+	+	+	+	+
Gallic acid	0.110	0.135	0.104	0.196	0.207	0.140	0.271	0.242	0.209	0.254
Chlorogenic acid	+	+	+	+	0.431	+	0.019	+	+	0.031
Protocatechuic acid	0.090	0.242	0.126	0.178	+	0.291	+	0.109	0.369	+
(+)-Catechin	—	—	—	—	—	—	—	+	—	
Caffeic acid	0.485	0.070	0.581	0.660	0.674	0.235	1.981	0.581	0.230	0.316
Cyanidin	3.492	1.309	3.355	3.750	1.643	1.387	1.845	1.780	1.185	4.028
p-Coumaric acid	3.157	2.370	1.613	1.661	1.798	0.425	0.922	1.409	0.240	3.985
Myricetin	0.114	0.098	0.034	0.139	0.241	0.021	0.138	0.180	+	0.201
Quercetin	0.233	—	0.661	0.786	0.819	0.849	0.728	0.925	0.855	0.475
t-Cinamic acid	0.385	0.272	0.396	0.454	0.381	0.451	0.541	0.384	0.453	0.375
Kaempferol	2.994	0.754	2.237	2.363	1.151	2.013	0.680	0.510	2.888	2.453

* + = trace amount, — = none detected.

Plant resistance to herbivores is a complex system of chemical and physical factors (Feeny 1970; Reese 1983). The role of phenols in this system is not well understood and is currently a controversial issue (Reese 1983; Zucker 1983; Martin and Martin 1984). These allelochemicals may be an important part of the defensive system. However, in our study, we were unable to show that phenols were a major factor in the resistance of hops to *T. urticae*.

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INSECTS FROM NATIVE AND CULTIVATED SUNFLOWERS (*HELIANTHUS*) IN SOUTHERN LATITUDES OF THE UNITED STATES[1]

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Abstract: Non-pest species of insects found on both cultivated sunflower and native species of *Helianthus*, as well as pests alternatively found on native sunflowers in southern latitudes of the United States, are categorized by order, family, genus, and species and by host species and location. Brief comments about the more prominent species of insects associated with cultivated sunflower are included in the text. Also included is an addendum to an earlier paper on natural enemies of insect pests of sunflower.

Key Words: *Helianthus* species, Coleoptera, Lepidoptera, Diptera, Natural Enemies, Insecta.

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Sunflower, *Helianthus annus* (L.), as a native cultivated crop of North America, is sometimes inundated by indigenous insect faunas. Naturalists of the early 1900's reported on insects visiting the common annual sunflower, *H. annuus* (Cockerell 1915; Robertson 1922; and Walker 1936). Several papers also have described insects associated with cultivated sunflower in localized areas (Genung and Green 1979; Lynch and Garner 1980; Phillips et al. 1973; Walker 1936), bees visiting sunflower (Hurd et al. 1980; Parker 1981; Robertson 1922), the incidence of insect species on native *H. annuus* (Hilgendorf and Goeden 1981), and management of insect pests on cultivated sunflower (McBride et al. 1981; Schulz 1978). Bibliographies of insect pests of sunflower and their natural enemies also are available (Rajamohan 1976; Rogers 1979a, 1980). Charlet et al. (1987) recently reported on insects associated with cultivated sunflower in the northern Plains. However, there is a paucity of literature for insect faunas of native sunflower (*Helianthus*) species. This paper reports the occurrence of innocuous insects on cultivated and native species of sunflower, as well as records of pest species on native sunflower hosts in mostly southern latitudes of the United States.

MATERIALS AND METHODS

Miscellaneous and innocuous species of insects were collected from cultivated sunflower during field plot work from native species of sunflower during excursions to collect *Helianthus* germplasm (Rogers et al. 1982). Standard entomological collecting techniques were used to document host, habitat, etc., of collected specimens. Representative specimens of initial collections of unidentified species were sent to taxa specialists (see acknowledgments) for species determination. Voucher specimens of selected species were deposited in the USDA Systematic Entomology Research Laboratory at the time of determination. Reference specimens of each species were subsequently deposited in the Department of Entomology, Texas A&M University, for proper cataloging and preservation.

RESULTS AND DISCUSSION

Native sunflowers are recognized by ecologists as key niches for the sustenance of beneficial insects (Rogers 1985a) and as alternate hosts for several pests of

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cultivated sunflower (Cockerell 1915; Rogers 1988). The extent to which native species of sunflowers function in the natural history of native insect fauna is not recognized by most entomologists. The species reported herein exclude major economic pests except to report their occurrence on non-cultivated sunflowers, and certainly are not exhaustive of the insect fauna encountered on *Helianthus*. However, this listing may serve as a reference from which more detailed studies may be undertaken. Species are listed below by order and family to facilitate discussion.

Coleoptera

The Coleoptera are well represented among the insect fauna of sunflowers (Table 1). Cerambycids commonly found on sunflowers throughout southern latitudes, e.g., *Ataxia hubbardi* Fisher, *Dectes texanus* LeConte and *Mecas* spp., may become serious pests of cultivated sunflower (Rogers 1977a). Although adult cerambycids are common on native species of sunflowers, visual damage that may be caused by larval burrowing is rare.

Sunflowers harbor several species of Chrysomelidae, among which the sunflower beetle, *Zygogramma exclamationis* (Fabricius) is a serious pest in the Great Plains (Rogers 1977b; Westdal 1975). Pollen of sunflower appears to be a major attractant for the chrysomelids, for these species most often are found on the inflorescence. The flea beetle, *Systena blanda* Melsheimer, often becomes extremely abundant and skeletonizes the bracts of the capitulum in late summer in the southern Plains.

The family Curculionidae is of particular importance to the sunflower industry of the United States. Species of *Apion*, *Cylindrocopturus*, *Haplorhynchites*, and *Smicronyx* often cause economic losses in cultivated sunflower, both as direct pests and as mechanical vectors of plant pathogens (Charlet 1983; Gaudet and Schulz 1981; Oseto and Braness 1979; Rogers et al. 1983; Yang et al. 1983). *Haplorhynchites aeneus* (Boheman) and other species of this genus also frequent several species of *Helianthus* (Table 1) (Hamilton 1974). The cocklebur weevil, *Rhodobaenus tredecimpunctatus* (Illiger), is a common pest of cultivated sunflower (Vaurie 1981) and occurs on several species of *Helianthus*. Although not collected by me, species of *Lixus* have been collected from *H. annuus*, *Helianthus grosseserratus* Martens, *Helianthus rigidus* (Cass.) Desf. and *Helianthus tuberosus* L. (Webster 1889; Williams 1942.)

Colorful meloids are common pollen and nectar-feeders on sunflowers. *Gnathium* sp. and *Nemognatha lurida lurida* (LeConte) are among the most common insect inhabitants of cultivated sunflower in the southern Plains. The adults of these species have siphoning mouth parts and feed on floral nectar. Masses of several hundred orange eggs of *N. lurida* commonly occur on the back of sunflower capitula. When larvae hatch, they crawl to the inflorescence and attach themselves to visiting bees. At the nest or hives of the bees, the larvae become parasitic on bee eggs and food stores in brood cells (White 1983). The impact of *N. lurida* on bee broods is unknown. Mordellids (tumbling flower beetles) also are common on the inflorescence of cultivated sunflower in the southern Plains. Larvae of *Mordellistena* sp. are frequently found burrowing in the stalk of sunflower.

Scarabs sometimes becomes pests of cultivated sunflower and often are found on native *H. annuus* in the southern Plains (Rogers 1974; Rogers and Morrison 1978). Curiously, the adult rather than the grub is the stage that attacks sunflowers, and they may be found feeding on the roots, stems, florets, and immature seeds. *Cotinis nitida* (L.) often congregates on diseased tissue of sunflowers and feeds on fermenting plant exudates. Adults of *Cyclocephala dimidiata* Burmeister, *Euphoria inda* (L.), and *Euphoria kermi* Haldeman sometimes feed on immature achenes in large enough populations to be of concern to farmers.

Table 1. Coleoptera collected from cultivated and wild sunflower (*Helianthus*) species in southern latitudes of the United States (1974-1983).

Insect species	Host species	Collection site and year
COLEOPTERA		
Anthicidae		
Notoxus constrictus Casey	cultivar: Capitulum*	Vernon, TX 1976
Bruchidae		
Acanthoscelides prosopoides (Schaeffer)	cultivar: Capitulum	Chillicothe, TX 1976
Buprestidae		
Acmaeodera scalaris Mann	*praecox* Engleman: Capitula	Encinal, TX 1976
Acaemodera, mixta LeConte	cultivar: Captiulum	Vernon, TX 1975
Cantheridae		
Chauliognathus scutellaris (LeConte)	cultivar: Capitulum & Leaves	Texas (General) 1975-1982 & Capulin, NM
Carabidae		
Microlestes sp.	cultivar: Seedling	Friona, TX 1979
Cerambycidae		
Hippopsis lemniscata (Fabricius)	cultivar: Stalk	Vernon, TX 1975
" "	*atrorubens* L.: Stalk	McGee, MS 1977
Strangalia sexnotata (Haldeman)	*resinosus* Small: Capitula	McGee, MS 1977
Tragidion coquus (L.)	cultivar: Stalk	Bushland, TX 1977
Typocarnus sinuata (Newman)	cultivar: Stalk	Vernon, TX 1977
" "	*resinosus* Small: Stalk	McGee, MS 1977
" "	*heterophyllus* Nuttall: Stalk	Hattiesburg, MS 1977
Chrysomelidae		
Diabrotica tricinta (Say)	cultivar: Capitulum	Vernon, TX 1974
Diabrotica undecimpunctata howardii Baker	cultivar: Capitulum	Vernon, TX 1974
Diachus auratus (Fabricius)	*annuus*: Leaves	Uvalde, TX 1975
Disonycha sp.	cultivar: Leaves	Uvalde, TX 1975

Table 1. continued.

Insect species	Host species	Collection site and year
Labidomera clivicallis Kirby	cultivar: Leaves	Uvalde, TX 1975
Omophoita octomaculata Crotch	cultivar: Leaves	Uvalde, Weslaco, TX 1975
Ophraella notulata (Fabricius)	cultivar: Leaves	Uvalde, Corpus Christi, TX 1975
Sinea didema (Fabricius)	cultivar: Leaves	Bushland, TX 1979
	anomolus Blake: Leaves	Sand Mountain, UT 1978
Systema blanda Melsheimer	cultivar: Bracts	Texas (General) 1975-1983
Trirhabda nr. *bacharidis* (Weber)	*pumilus* Nuttall: Capitula	Manitoba Springs, CO 1977
Zygogramma conjuncta pallida Beland	cultivar: Leaves	Bushland, TX 1977
Zygogramma exclamationis (Fabricius)	cultivae & *annuus*: Leaves & Capitula	Texas (General) 1976-1983
Cleridae		
Enoclerus zonatus (Klug)	cultivar: Leaves	Bushland, TX 1977
Coccinellidae		
Neomysia pullata (Say)	cultivar: Leaves	Bushland, TX 1976
Curculionidae		
Anthonomus heterothecae Pierce	cultivar: Leaves	Bushland, TX 1976
Apion occidentale Fall	*annuus*: Stalk	
" "	*argophyllus* Torrey & Gray: Stalk	Texas (General) 1975-1983
Apion sp.	cultivar: Stalk	Uvalde, TX 1976†
Baris strenua LeConte	*paradoxus* Heiser: Leaves	Ft. Stockton, TX 1977
Baris sp.	cultivar: Stems	Bushland, TX 1978
Centrinaspis picumnus (Herbst)	*carnosus* Small: Capitulum	San Mateo, FL 1977
Chalcodermus aeneus Boheman	*annuus* (L.) Stems	McLoud, OK 1976
Compsus auricephalus (Say)	cultivar: Capitulum	Uvalde & Vernon, TX 1975
Contrahelus leucophaeatus (Fabricius)	cultivar: Capitulum	Jones Co., TX 1975
Curculio sp.	cultivar: Capitulum	Vernon, TX 1974
Curculio victorienses (Chittenden)	cultivar: Capitulum	Jones Co., TX 1975
Cylindrocopturus adspersus (LeConte)	cultivar & *annuus*: Stalk & Roots	Texas (General), 1976-1983

Haplorhynchites aeneus (Boheman)	*annuus*: Capitula	Texas (General), 1976-1983
" "	*argophyllus*: Capitula	South Texas 1978
" "	*nuttallii* Torrey & Gray: Capitula	Trinidad, CO 1977
Haplorhynchites pseudomexicanus Hamilton	*rigidis* (Cass.) Desf.: Capitula	Lorenzo, NM 1977
" "	*lacineatus* Gray: Capitula	Lorenzo, NM 1977
" "	*niveus* (Benth.): Capitula	Sanderson, TX 1976
Haplorhynchites quadripennis (Fall)	cultivar: Leaves	Vernon, TX 1974
Hyperodes dorsalis (Dietz)	cultivar: Leaves	Bushland, TX 1976
Hyperodes sp.	cultivar: Leaves	Bushland, TX 1976
Microlarinus lareynii Jac duVal	cultivar: Capitulum	Vernon & Bushland, TX 1975, 1976
Ophryastes tuberosus LeConte	cultivar: Capitulum	Chillicothe, TX 1975
Pantomorus cervinus (Boheman)	*tuberosus* L. Leaves	Bamberg, SC 1977
Pantomorus elegans (Horn)	cultivar: Capitulum	Vernon, TX 1974
Rhodobaenus tredecimpunctata (Illiger)	*annuus*: Stalk	TX & OK (General), 1974-1983
" "	*grosserratus* Martens: Stalk	TX & OK (General), 1974-1983
Rhodobaenus tredecimpunctata	*maximiliani* Schrader: Stalk	" "
Rhynchites sp.	cultivar: Capitulum	Vernon, TX 1974
Smicronyx fulvus LeConte	cultivar: Capitulum	Texas (General) 1974-1983
" "	*annuus*: Capitula	Sand Mountain, UT 1977
" "	*anomalus* Blake: Capitula	Sand Mountain, UT 1977
Smicronyx rectirostris Blatchley	*angustifolius* L.: Capitula	Alvin, TX 1976
Smicronyx scapalis (LeConte)	cultivar: Capitulum	Vernon, TX 1976
Smicronyx sordidus LeConte	cultivar: Capitulum	Texas (General) 1974-1983
Tanymecus confertus Gyllenhal	*annuus*: Capitula & Stems	Vernon, TX 1974
" "	cultivar: Capitulum	Vernon, TX 1976
Trichobaris texana LeConte	cultivar: Bud	Bushland, TX 1978
Melanopleurus belfragei (Stral)	cultivar: Leaves	Vernon, TX 1974
Elateridae		
Conoderus sp.	cultivar: Stalk	Bushland, TX 1982
Languriidae		
Languria sp.	cultivar: Stems & Capitula	Uvalde, TX 1975
Lathridiidae		
Melanophthalmus sp.	*annuus*: Leaves	Trivoli, TX 1976

Table 1. continued

Insect species	Host species	Collection site and year
Meloidae		
Gnathium sp.	cultivar: Capitulum	Texas (General) 1975-1983
"	*annuus*: Capitula	
"	*anomalus*: Capitula	Sand Mountain, UT 1977
"	*praecox* ssp. *hirtus* Heiser: Capitula	Crystal City, TX 1976
Nemognatha lurida lurida (LeConte)	cultivar: Capitulum	TX & KS (General) 1977-1982
Zonitis bilineatus (Say)	cultivar: Capitulum	Bushland, TX 1976
Zonitis dunniana Casey	*neglectus* Heiser: Capitula	Monohans, TX 1976
Zonitis punctipennis (LeConte)	*niveus* (Benth.) Brandegee: Capitula	Sanderson, TX 1976
Mordellidae		
Mordellistena sp.	*paradoxus* Heiser: Stalk	Ft. Stockton, TX 1976
"	cultivar: Stalk	Texas (General) 1976-1980
Nitidulidae		
Carpophilus mutitabus Erichson	cultivar: Capitulum	Weslaco, TX 1975
Phalacridae		
Phalacrus sp.	*annuus*: Leaves	Tivoli, TX 1976
Scarabaeidae		
Anomala foraminosa Bates	*annuus*: Capitula	Chillicothe, TX 1975
Cotinis nitida (L.)	cultivar: Stalk & Leaves	Texas (General) 1976-1983
Cyclocephala dimidiata Burmeister	cultivar: Seedlings	Brownsfield, TX 1977
Euphoria inda (L.)	*annuus*: Stems	Brownsfield, TX 1977
Euphoria kermi Haldeman	*annuus*: Capitula	Chillicothe, TX 1975
Phyllophaga lanciolata Say	*annuus*: Capitula & Stems	Chillicothe, TX 1975
Phyllophaga sp.	*annuus*: Capitula & Stems	Chillicothe, TX 1975
Tenebrionidae		
Blapstinus pratensis LeConte	cultivar: Seedlings	Friona, TX 1979
Bothrotes plumbeus plumbeus (LeConte)	cultivar: Leaves	Vernon, TX 1974
" " "	*maximiliani*: Capitula	Big Springs, TX 1974
Eleodes opaca Say	cultivar: Seedlings	Friona, TX 1979

* Cultivar refers to cultivated *H. annuus*.

† Also collected from a cultivar in Glyndon, MN 1976.

Tenebrionids are uncommon on sunflower. However, sunflower seedlings emerging from fallow wheat stubble have been destroyed in the High Plains of Texas by adults of *Blapstinus pratensis* LeConte and *Eleodes opaca* Say.

Diptera

Larvae of several species of Diptera use sunflowers as hosts (Table 2). *Melanagromyza* (Agromyzidae) spp. have larvae that commonly burrow in the capitula, stems (and stalk), and leaves of sunflowers. Larvae of *Hylemya* (Anthomyiidae), *Euxesta* (Otitidae), *Seioptera* (Ptitidae), and *Resseliella* (Cecidomyiidae) are stem borers in sunflowers. The Cecidomyiidae associated with sunflowers have been reported (Rogers et al. 1979). Larvae of the sunflower midge, *Contarinia schulzi* Gagné, and the sunflower seed midge, *Neolasioptera helianthi* (Felt), have become economically damaging to cultivated sunflower seed production (Schulz 1973; Kreitner and Rogers 1981). Adults of *Physiophora demandata* (Fabricius) frequently congregate near the terminals of sunflowers and feed on extrafloral nectar.

Tephritids often may be the most noticeable insects on sunflowers, with the wing-waving adults engaged in mating and territorial duels. The larva of these species is the stage that inflicts damage to sunflower plants. Larvae of *Neotephritis finalis* (Loew) develop in immature achenes (Foote 1960). Both the capitula and the stalk of sunflowers are attacked by larvae of *Paracantha cultaris* Coquillett (Cavender and Goeden 1984). *Trupanea* spp. larvae burrow in the capitula of both cultivated and native sunflowers (Cavender and Goeden 1982). Larvae of the sunflower maggot, *Strauzia longipennis* (Wiedemann), tunneling into the stalk of cultivated sunflower, sometimes cause economic losses in the northern Plains (Westdal and Barrett 1962). *Gymnocarena diffusa* Snow larvae are common in the capitula of cultivated sunflower, *Helianthus annuus* and *H. maximiliani* Schrader (Kamali and Schulz 1974).

Hemiptera

Hemipterans are common but not abundant on sunflowers in the United States (Table 3). *Nysius* spp. (Lygaeidae) sometimes colonize the capitulum of cultivated sunflower in the southern Plains where pigweeds (*Amaranthus* spp.) infest fields. *Chlamydatus associatus* (Uhler) adults often feed on the leaves of sunflower in the southern Plains.

Homoptera

Aphids (Aphididae) colonize both cultivated sunflower and native species of *Hellianthus* (Rogers et al. 1978) (Table 3). Leafhoppers (Cicadellidae) of *Aceratagallia* and *Empoasca* spp. attack the leaves of sunflowers, often resulting in chlorosis of the plants. A treehopper, *Vanduzea laeta* (Goding), is a common insect on cultivated sunflower as well as on several species of *Helianthus* in southern latitudes. This treehopper colonizes stems of plant terminals and leaf petioles, and usually is tended by ants. *Phenacoccus solani* Ferris (Pseudococcidae) is a common pest of sunflower in the greenhouse, and *Phenacoccus solenopsis* Tinsley occurs on *Helianthus paradoxus* Heiser, where colonies develop on the rootcrown and hypocotyl of the plant at the soil surface.

Lepidoptera

Species of Lepidoptera rival Coleoptera in dominating insect faunas of sunflowers (Table 4). Several lepidopterous species become serious economic threats to

Table 2. Diptera collected from cultivated and wild sunflower (*Helianthus*) species in southern latitudes of the United States (1974-1983).

Insect species	Host species	Collection site and year
Agromyzidae		
Calycomyza sp.	cultivar: Leaves*	Bushland, TX 1976
Melanagromyza minimoides Spencer†	cultivar: Stalk	Vernon, TX 1974
Melanagromyza viridis (Frost)	*arizonensis* R. Jackson: Capitula	Snow Flake, AZ 1977
" "	*deserticola* Heiser: Capitula	Virgin, UT 1977
Anthomyiidae		
Calythea monticola (Bigot)	cultivar: Stem	Bushland, TX 1978
Adia platura (Meigen)	cultivar: Leaves	Chillicothe, TX 1976
Adia cinerella (Fallen)	cultivar: Capitulum	Vernon, TX 1974
Bibionidae		
Plecia neartica Hardy	*annuus*: Leaves	Moscow, TX 1976
Bombyliidae		
Phthiria sp.	cultivar: Capitulum	Bushland, TX 1976
Cecidomyiidae		
Contarinia schulzi Gagné	*annuus*: Capitula	Knox County, TX 1972
Neolasioptera helianthi (Felt)	cultivar & *Helianthus* spp.: Capitula	General So. U.S., 1974-1983
Resseliella sp.	*nuttalii* Torrey & Gray: Stalk	Bushland, TX 1983
Chironomidae		
Procladius sp.	cultivar: Bud	Bushland, TX 1978
Chloropidae		
Elachiptera costata (Loew)	cultivar: Stalk	Bushland, TX 1981
Fiebrigella n. sp.	cultivar: Leaves	Chillicothe, TX 1975
Thaumatomyia glabra (Meigen)	cultivar: Leaves	Bushland, TX 1976
Calliphoridae		
Cochliomyia macellaris (Fabricius)	cultivar: Capitulum	Vernon, TX 1974
Phaenicia sericata (Meigen)	cultivar: Leaves	Bushland, TX 1974
" " "	cultivar: Leaves	Vernon, TX 1975
Muscidae		
Musca domestsica L.	cultivar: Leaves	Bushland, TX 1971
Orthellia caesarion (Meigen)	cultivar: Capitulum	Vernon, TX 1974
Spilogona sp.	cultivar: Capitulum	Vernon, TX 1974
Otitidae		
Euxesta nitidiventris Loew	cultivar: Leaves	Bushland, TX 1976-1978
Euxesta pechumani Curran	cultivar: Stalk	Bushland, TX 1983

Euxesta sp.	cultivar: Stalk	Bushland, TX 1981
Physiphora demandata (Fabricius)	cultivar: Stalk	Bushland, TX 1978
Tetanops leuridipennis Loew	cultivar: Leaves	Uvalde, TX 1975
Genus unknown	cultivar: Stalk	Bushland, TX 1981
Ptitidae		
Seioptera vibrans (L.)	cultivar: Stalk	Bushland, TX 1981
Sarcophagidae		
Blaesoxipha kellyi (Aldrich)	cultivar: Leaves	Texas (General) 1975-1983
Blaesoxipha rudis (Aldrich)	cultivar: Leaves	Texas (General) 1975-1983
Sciaridae		
Bradysia impatiens (Johannsen)	cultivar: Soil	Bushland, TX 1976-1983
Tachinidae	cultivar: Leaves	Chillicothe, TX 1976
Clausicella opaca (Coquillett)	cultivar: Leaves	Chillicothe, TX 1976
Nemorilla pysta (Walker)	cultivar: Leaves	Vernon, TX 1974
Hyalomya sp.	cultivar: Leaves	Vernon, TX 1974
Euphoracera sp.		
Syrphidae	cultivar: Capitulum	Vernon, TX 1977
Eristalis tenax (L.)	cultivar: Capitulum	Vernon, TX 1974
Eupeodes volucris Osten Sacken	cultivar: Capitulum	Vernon, TX 1974
Metasyrphus americanus (Wiedemann)	cultivar: Capitulum	Vernon, TX 1974
Toxomerus marginatus (Say)		
Tephritidae	cultivar: Stalk	Bushland, TX 1983
Gymnocarena diffusa Snow	*annuus*: Capitula	Variadero, NM 1976
Euarestoides acutangulus (Thomson)	*petiolaris*: Capitula	Variadero, NM 1976
" "	cultivar: Capitulum & Stalk	Texas (General) 1975-1983
Paracantha cultaris Coquillett	*annuus*: Capitula	Variadero, NM 1977
" "	*debilis* spp. *cucumerifolius* Torrey & Gray: Capitula	East Texas 1977
" "		
Neotephritis finalis (Loew)	cultivar: Capitulum	Texas (General) 1974-1983
Orellia palposay (Loew)	cultivar: Capitulum	Vernon, TX 1976
Strauzia longipennis (Weidmann)	cultivar & *annuus*: Stalk	Bushland, TX 1978
Trupanea nigricans (Coquillett)	*annuus*: Capitula	Variadero, NC 1976
" "	*ciliaris* DC: Capitula	Roswell, NM 1976
" "	*neglectus*: Capitula	Monohans, TX 1976
" "	*petiolaris*: Capitula	Variadero, NM 1976

* Cultivar refers to cultivated *H. annuus*

† *Melanagromyza* sp. also collected from a cultivar at Glyndon, MN , 1975; *M. splendida* Frick was collected from a cultivar in Santiago, Chile, 1978.

Table 3. Heteroptera collected from cultivated and wild sunflower (*Helianthus*) species in southern latitudes of the United States (1974-1983).

Insect species	Host species	Collection site and year
HEMIPTERA		
Coreidae		
Archimerus alternatus (Say)	cultivar: Leaves & Stalk*	Bushland, TX 1976
Largidae		
Largus sp.	cultivar: Leaves	Chillicothe, TX 1975
Lygaeidae		
Nysius californicus Stal	*petiolaris*: Buds	Chillicothe, TX 1976
Nysius raphanus Howard	cultivar: Capitulum	Sunray, TX 1977
Xyonysius californicus (Stal)	cultivar: Leaves	Vernon, TX 1975
Miridae		
Chalmydatus associatus (Uhler)	cultivar: Leaves	Munday, Bushland, TX 1975
Lopidea sp.	cultivar: Leaves	Uvalde, 1975, Chillicothe 1980
Rhopalidae		
Harmostes reflexulus (Say)	cultivar: Leaves	Chillicothe, TX 1976
HOMOPTERA		
Cercopidae		
Clastoptera xanthocephala Germar	cultivar: Stems	Corpus Christi, TX 1975
Philaenus spumarius (L.)	*maximiliani*: Leaves	Paris, TX 1976
Cicadellidae		
Aceratogallia uhleri (Van Duzee)	cultivar: Leaves	Bushland, Uvalde, TX 1975
Aceratogallia sp.	cultivar: Leaves	Vernon, Uvalde, TX 1974-1975
Empoasca abrupta DeLong	cultivar: Leaves	Bushland & Chillicothe, TX 1975-1978
Empoasca erigeron DeLong	cultivar: Leaves	Uvalde, TX, Davis, CA 1975
Empoasca sp.	cultivar: Leaves	Vernon, TX 1974
Homalodisca coagulata (Say)	*argophyllus*: Leaves & Stalk	Vernon, TX 1974

Homalodisca coagulata (Say)	*argophyllus*: Leaves & Stalk	Falfurias, TX 1976
Xeropholea viridis (Fabricius)	cultivar: Leaves	Vernon, TX 1974
Oncometopia nigricans (Walker)	cultivar: Leaves	Vernon, TX 1974
Scaphytopius sp.	cultivar: Leaves	Uvalde, TX 1975
Cixiidae		
Oliarus aridus Ball	cultivar: Leaves	Vernon, TX 1984
Delphacidae		
Stobaera tricarinata (Say)	cultivar: Leaves	Uvalde, TX 1975
Membracidae		
Publilia modesta Uhler	*annuus*: Stems	Chillicothe, TX & Variadero, NM 1976
Vanduzea laeta (Goding)	cultivar: Stems	Texas (General) 1974-1983
" "	*annuus*, *maximiliani*,	Texas (General) 1974-1983
" "	*petiolaris*: Stems	
Vanduzea triguttata (Burmeister)	cultivar: Stems	Vernon, TX 1974
Pseudococcidae		
Phenacoccus solenopsis Tinsley	*paradoxus*: Stalk	Ft. Stockton, TX 1976
Phenacoccus solani Ferris	cultivar: Stalk	Bushland, TX 1980-1983
Psyllidae		
Heteropsylla texana Crawford	cultivar: Leaves	Lorenzo, TX 1976

* Cultivar refers to cultivated *H. annuus*.

Table 4. Lepidoptera collected from cultivated and wild sunflower (*Helianthus*) species in southern latitudes of the United States (1974-1983).

Insect species	Host species	Collection site and year
Arctiidae		
Estigmene acrea (Drury)	cultivar: Leaves*	Bushland, Vernon, TX 1974-1983
Cochylidae		
Cochylis hospes Walsingham	*floridanus* Gray ex Chapman: Capitula	Tallahassee, FL 1977
" "	*petiolaris*: Capitula	Channing, TX 1976
" "	*praecox*: Stalk	Eagle Lake, TX 1976
Cochylis sp.	*argophyllus*: Capitula	Ingleside, TX 1976
" "	*petiolaris*: Capitula	Channing, FL 1976
Gelechiidae		
Isophrictis similiella Chambers	*annuus*: Stalk & Capitula	Bushland, TX 1975-1983
" "	*argophyllus*: Capitula	Rufegio, TX 1976
" "	*hirsutus* Rafinesque: Capitula	Bushland, TX 1982
" "	*neglectus*: Capitula	Monohans, TX 1976
" "	*longifolius* Pursh: Capitula	Guest, AL 1977
" "	*petiolaris*: Capitula	Channing, TX 1976
" "	*rigidus*: Capitula	Manitor Springs, CO 1977
Isophrictis sp.	cultivar: Stalk	Uvalde, TX 1975
Geometridae		
Synchlora sp.	*occidentalis* Riddell: Capitula	Kemah, TX 1976
Eupithecia miserulata (Grote)	cultivar: Capitulum	Vernon, TX 1974
Gracillariidae		
Cremastobombycia ignata (Fabricius & Brants)	*annuus*: Leaves	McLoud, OK 1977
Hesperiidae		
Hylephila phyleus (Drury)	cultivar: Capitulum	Vernon, TX 1974
Lerodea sufala (Edwards)	cultivar: Capitulum	Vernon, TX 1974

Lyonetiidae		
Bucculatrix magnelta Chambers	*annuus*: Stems	McLoud, OK 1976
Bucculatrix simulans Braum	*annuus*: Stems	Buckeye, AZ 1977
Bucculatrix sp.	*angustifolius*: Stems	Muenster, TX 1976
Noctuidae		
Amphipyinae (unknown genus)	*maximiliani*: Capitula	Abilene, Austin, TX 1976, 1981
"	*eggertii* Small: Capitula	Rocky Dale, IN 1977
"	*tuberosus*: Capitula	Stewart, OK 1977
Heliothis zea (Boddie)	cultivar: Capitulum	Texas (General) 1974-1983
Heliothis virescens (Fabricius)	cultivar: Capitulum	Texas (General) 1974-1983
Pseudaletia unipuncta (Haworth)	cultivar: Capitulum	Vernon, TX 1974
Pseudanthoecia tumida (Grote)	*annuus*: Leaves	Capulin, NM 1977
Pseudoplusia includens (Walker)	cultivar: Capitulum	Vernon, TX 1974
Spodoptera exigua (Hübner)	cultivar: Leaves	Vernon, TX 1974
Spodoptera frugiperda (J. E. Smith)	cultivar: Capitulum	Vernon, TX 1974
Stibadium spumosum Grote	*annuus*: Capitula	Bushland, TX & Jal, NM 1978
Tarachida tortricina (Zeller)	cultivar: Capitulum	Vernon, TX 1974
Trichoplusia ni (Hübner)	cultivar: Capitulum	Vernon, TX 1974
Nymphalidae		
Chlosyne lacini Geyer	cultivar: Leaves	Texas (General) 1976-1983
" "	*annuus*: Leaves	" " "
" "	*argophyllus*: Leaves	" " "
" "	*neglectus*: Leaves	" " "
" "	*niveus*: Leaves	" " "
Vanessa cardui (L.)	cultivar: Leaves	Texas (Generral) 1974-1983
" "	*maximiliani*: Capitula	Sherman, TX 1977
Tortricidae		
Suleima helianthana Riley	*annuus*: Capitula	Manitoba Springs, CO 1977
" "	*anomalus*: Capitula	Sand Mountain, UT 1977

Table 4. continued

Insect species	Host species	Collection site and year
Suleima helianthana Riley	*argophyllus*: Capitula	Rufegio, TX 1976
" "	*maximiliani*: Capitula	Muenster, TX 1977
" "	*niveus*: Capitula	Sanderson, TX 1974
" "	*occidentalis* ssp. *plantagineous* (T&G) Heiser	Ringgold, TX 1975
" "	*paradoxus*: Stems	Ft. Stockton, TX 1977
" "	*petiolaris* ssp. *petiolaris*: Capitula	Texas (General) 1975, 1982
Suleima helianthana	*praecox* ssp. *hirtus*: Capitula	Crystal City, TX 1977
" "	*rigidus*: Capitula	Adin, CA 1977
Pyralidae		
Achyra ramtalis (Guenee)	cultivar: Capitulum & Leaves	Bushland, TX 1977
Cadra cautella (Walker)	cultivar: Stored seed	Chillicothe & Bushland, TX 1974-1983
Hahncappsia coloradensis (Grote & Robinson)	*annuus*: Leaves	Bushland, TX 1980
Hahncappsia huachualis (Capps)	cultivar: Leaves	Bushland, TX 1978-1983
Hahncappsia rantalis Guenee	cultivar: Capitulum	Bushland, TX 1977
Hahncappsia sp.	cultivar: Stalk	Uvalde, TX 1975
Nomophila nearctica Munroe	cultivar: Capitulum	Vernon, TX 1974
Tortricidae		
Platynota sp.	*carnosus*: Capitulum	San Mateo, FL 1977

* Cultivar refers to cultivated *H. annuus*.

cultivated sunflower. These species have received adequate coverage in the economic entomology literature; hence, they will be deleted from this paper. Salt marsh caterpillar, *Estigmene acrea* (Drury), larvae frequently defoliate cultivated sunflower and *H. annuus* in the southern Plains. *Cochylis hospes* Walsingham (Cochylidae), the banded sunflower moth, infests the inflorescence of several *Helianthus* species in the southern Plains (Table 1), and it sometimes becomes an economic threat to seed production in cultivated sunflower in the northern Plains (Charlet and Busacca 1986). Likewise, larvae of the gelechiid *Isophrictis similiella* Chambers infest several *Helianthus* species on the High Plains of Texas and New Mexico and sometimes cause severe tunneling in the stalk of cultivated sunflower (Underhill et al. 1987). Larvae of *Bucculatrix magnella* Chambers and *B. simulans* Braum (Lyonetiidae) develop in stem galls of *H. annuus*.

Many species of Noctuidae that are agricultural pests may be collected from sunflowers (Table 4). For the most part, adult noctuids are collected while resting on the plant or while feeding on floral nectar. Occasionally, however, larvae of *Heliothis zea* (Boddie) (corn earworm) attack developing achenes, and larvae of *Spodoptera exigua* (Hübner) (beet armyworm) sometimes defoliate cultivated sunflower (Lynch and Garner 1980). Larvae of an unknown noctuid genus were collected from *Helianthus eggerti* Small, *H. maximiliani*, and *H. tuberosus* L. *Stibadium spumosum* Grote is one of a few noctuids that may be restricted to sunflower. Adults of *S. spumosum* oviposit on the back of the capitula of *H. annuus*. Small larvae burrow through the capitula and develop in the inflorescence while consuming the achenes (Rogers et al. 1986)

Two nymphalids are frequent defoliators of sunflowers. Gregarious larvae of *Chlosyne lacinia* (Geyer) feed on several species of *Helianthus*, as well as on cultivated sunflower (Table 1, Neck 1973). Larvae of *Vanessa cardui* (L.) are solitary feeders and are found on sunflowers and other composites from Canada into Mexico (Williams 1970). This migratory species may appear without warning and completely defoliate fields of cultivated sunflower. The sunflower bud moth, *Suleima helianthana* (Riley) (Tortricidae), is common in cultivated sunflower as well as on several species of *Helianthus* (Rogers 1979b). Larvae of pyralids are common on *Helianthus* spp. in southern latitudes. *Hahncappsia coloradensis* (Grote and Robinson) and *H. huachualis* (Capps) larvae construct leaf-rolls from which they skeletonize one leaf at a time. Leaf-roller larvae are heavily parasitized on *H. annuus* in Texas (Table 2, Rogers 1980). *Eucosma womonana* Kearfott (Tortricidae) larvae may cause extensive damage to the roots of cultivated sunflower and *H. tuberosus* (Rogers 1985b).

Miscellaneous Orders

Native bees (Hymenoptera) of several families are almost always encountered on the inflorescence of sunflowers (Table 5). The interaction of these insects with sunflowers has been adequately discussed by Hurd et al. (1980) and Parker (1981). Parasitic Hymenoptera encountered on sunflowers were presented by Rogers (1980) and are updated later in this paper.

A tree cricket, *Oecanthus argentinus* Saussure (Orthoptera), frequently is seen feeding on pollen of sunflower in northern Texas. *Melanoplus differentialis* (Thomas) and other species of grasshoppers (Acrididae) consume *H. annuus*, *H. petiolaris*, and other rangeland species of *Helianthus* in the Great Plains (Lewis 1979).

Ectopsocopsis crytomeriae (Enderlein) (Psocoptera) are found in decaying capitula of cultivated sunflower that are infected by the *Rhizopus* head rot pathogen.

Thrips (Thysanoptera) are common inhabitants of the sunflower inflorescence, where the life history of hundreds of specimens sometimes occur (Table 5). Presumably, most of the thrips in the inflorescence are feeding on pollen. The bionomics of thrips on sunflower should be investigated.

Natural Enemies

The natural enemies of pests of cultivated sunflower were cataloged by Rogers (1980). The listing in Table 6 is an update of the previous work. *Perilloides bioculatus* (Fabricius) is an effective predator of the sunflower beetle, *Z. exclamationis*, in the southern Plains. Both larvae and adults of this pentatomid prey on larvae and adults of *Z. exclamationis*. Two color variations of *P. bioculatus* in sunflower are black stripes on a bright red background, and brown stripes on a cream-colored background. The latter color variation closely resembles the color of adults *Z. exclamationis*. *Perilloides bioculatus* has been considered an effective predator of the Colorado potato beetle, *Leptinotarsa decemlineata* (Say) (Knight 1923).

The leaf-rollers, *Hahncappsia coloradensis* and *H. huachualis*, are heavily parasitized by species from several families of Hymenoptera on native *H. annuus* in the southern Plains. Because these Lepidoptera rarely occur on cultivated *H. annuus*, parasites must be effectively controlling leaf-roller populations on their native hosts. An investigation of the bionomics of parasites of *H. coloradensis* and *H. huachualis* might be of interest to sunflower pest managers.

Neuropteran species are poorly represented among the insect faunas of sunflowers. *Chrysopa carnea* Stephens (Chrysopidae) larvae and adults are often encountered in the inflorescence, or on petioles, feeding on floral and extrafloral nectar (Table 6).

CONCLUSIONS

From the diversity of taxa reported herein, it is apparent that the *Helianthus* species have rich insect faunas in their native habitats, and that some of the insect species found on native sunflowers have adapted to colonization of *H. annuus* in cultivated conditions. Considering that *Helianthus* species are native to North America (Heiser et al. 1969), that cultivated sunflower has become an important oilseed crop in several areas of the United States, and that native sunflowers are important genetic resources for improving cultivated *H. annuus* (Thompson et al. 1981), it is important that sunflower entomologists become familiar with these faunas in their natural habitats as a basis on which to implement ecologically and economically sound pest management strategies for cultivated sunflower.

Table 5. Hymenoptera, Orthoptera, and Thysanoptera collected from cultivated and wild sunflower (*Helianthus*) species in southern latitudes of the United States (1974-1983).

Insect species	Host species	Collection site and year
HYMENOPTERA		
Halictidae		
Agapostemon texanus Cresson	cultivar: Capitulum*	Vernon, TX 1974
Agapostemon trespennis (Cresson)	cultivar: Capitulum	Vernon, TX 1974
Megachilidae		
Megachile sp. nr. *texana* Cresson	cultivar: Capitulum	Vernon, TX 1974
Perilampidae		
Perilampus similis Crawford	cultivar: Capitulum	Vernon, TX 1974
ORTHOPTERA		
Gryllidase		
Oecanthus argentinus Saussure	cultivar: Capitulum	Vernon, TX 1974
PSOCOPTERA		
Ectopsocidae		
Ectopsocopsis cryptomeriae (Enderlein)	cultivar: Capitulum	Weslaco, TX 1975
Lachesillidae		
Lachesilla nubilalis (Aaron)	*microcephalus* Torrey & Gray: Capitula	Asheboro, NC 1977
THYSANOPTERA		
Aeolothripidae		
Aerlothrips duvali Moulton	cultivar: Bud	Chillicothe, TX 1976
Thripidae		
Frankliniella occidentalis (Pergande)	cultivar: Bud	Chillicothe, TX 1976
Microcephalothrips abdominalis Crawford	*annuus*: Capitula	Weslaco, TX 1975
Orellia palposa (Loew)	cultivar: Bud	Chillicothe, TX 1976
Thrips tabaci Lindeman	cultivar: Bud	Chillicothe, TX 1976

* Cultivar refers to cultivated *H. annuus*.

Table 6. Natural enemies of insects found on sunflowers in southern latitudes of the United Stated (1974-1983).

Natural enemy	Host species	Collection site and year
HEMIPTERA		
Pentatomidae		
Perilloides bioculatus (Fabricius)	*Zygogramma exclamationis* (Fabricius)	Bushland, TX 1976-1983
Reduviidae		
Sinea didemo (Fabricius)	*Hippodamia convergens* Guerin-Meneville	Bushland, TX 1977
" "	Inflorescence of *H. anomalus*	Sand Mountain, UT 1977
HYMENOPTERA		
Braconidae		
Apanteles sp.	*Chlosyne lacinia* (Geyer)	McLoud, OK 1979
Chelonus sp.	*Homoeosoma eletellum* (Hulst)	Bushland, TX 1983
Cremnops virginiensis (Morrison)	*Hahncappsia coloradensis* (Grote & Robinson)	Bushland, TX 1983
" "	*Hahncappsia huachualis* (Capps)	Bushland, TX 1983
Schizoprymus sp.	*Mordella* sp.	Bushland, TX 1973
Chalcididae		
Brachymeria ovata (Say)	*Hahncappsia coloradensis*	Bushland, TX 1983
"	*Hahncappsia huachualis*	Bushland, TX 1983
Eucoilidae		
Hexacola n. sp.	*Seioptera vibrans* (L.)	Bushland, TX 1982
Eupelmidae		
Eupelmus sp.	*Bucculatrix magnelta* Chambers	McLoud, OK 1976
Eurytomidae		
Eurytoma sp.	*Bucculatrix magnelta*	McLoud, OK 1976
Ichneumonidae		
Eipshosoma pyralidis Ashmead	*Hahncappsia coloradensis*	Bushland, TX 1983
" "	*Hahncappsia huachualis*	Bushland, TX 1983
Mesochorus discitergus (Say)	*Chlosyne lacinia*	McLoud, OK 1976
Sinophorus n. sp.	*Stibadium spumosum* Grote	Bushland, TX 1983
Pteromalidae		
Heteroschema sp.	*Seioptera vibrans*	Bushland, TX 1982
NEUROPTERA		
Chrysopidae		
Chrysopa carnea Stephens	Inflorescence of cultivars	Vernon, TX 1974
" "	Petioles of *H. annuus*	Bushland, TX 1982

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ERRATUM

In the Journal of Agricultural Entomology, Volume 5, Number 3, July 1988, there are two errors in Table 1 on page 157 under "June 11 -- ECB stage†" the correct entry for Aug. 7 should have been "L_5P" and the correct entry for Aug. 14 should have been "P,L_1." The following is the corrected table in its entirety:

Table 1. Weekly ECB feeding damage in early-(May), mid-(June), and late-(July) season plantings of hybrid sweet corn at Hamden, Connecticut 1986.

	May 12			June 11			July 10		
Sample date	Corn plant stage*	ECB stage†	Mean ± SE % plants damaged‡	Corn plant stage*	ECB stage†	Mean ± SE % plants damaged‡	Corn plant stage*	ECB stage†	Mean ± SE % plants damaged‡
June 26	W	L_1	21.0 ± 2.9ab						
July 3	W	L_2	26.0 ± 4.0a						
July 11	T	L_2,L_3	36.0 ± 3.6d	W	L_1,L_2	17.0 ± 4.9a			
July 17	T	L_3	28.0 ± 4.3a	W	L_2,L_3	8.7 ± 1.6b			
July 23	S	L_3,L_4	12.4 ± 3.4c	W	L_3,L_4	4.3 ± 2.4c			
July 30	S	L_4,L_5	10.0 ± 2.3c	W	L_4,L_5	6.3 ± 1.2bc	W	L_1	10.0 ± 2.0a
Aug. 7	S	L_5P	14.0 ± 1.0bc	T	L_5,P	4.0 ± 0.9c	W	L_1	16.0 ± 4.3b
Aug. 14				S	P,L_1	8.7 ± 1.6b	W	L_2	17.0 ± 2.7bc
Aug. 21				S	L_2	40.0 ± 5.4d	W	L_2	22.3 ± 4.8cd
Aug. 29				S	L_2	52.0 ± 2.0e	W	L_2,L_3	19.7 ± 3.2bc
Sept. 5							W	L_2,L_3	27.3 ± 4.3de
Sept. 10							T	L_2,L_3	28.0 ± 4.9de
Sept. 17							S	L_2-L_4	33.6 ± 8.2e
Sept. 26							S	L_3,L_4	51.0 ± 5.7f
Oct. 3							S	L_3,L_4	52.0 ± 4.6f

* W = whorl, T = tasseling, S = silking.

† L_1 - L_5 = larval instars, P = pupae.

‡ Means within columns which are followed by the same letter are not significantly different ($P > 0.05$) as determined by chi-square contigency table analysis.

ERRATUM

In the Journal of Agricultural Entomology, Volume 5, Number 3, July 1988, the data in Table 2 on page 158 were inadvertently reversed in the two columns under "Means ± SE % damaged†"; under "Ears" in the column "Mean ± SE no. ECB/50*," the first entry should be "1.1 ± 1.6a." The following is the corrected table in its entirety:

Table 2. ECB densities and feeding damage at harvest in early-(May), mid-(June), and late-(July) season plantings of hybrid sweet corn at Hamden, Connecticut 1986.

		Mean ± SE no. ECB/50*		Mean ± SE % damaged†		
Planting date	Harvest date	Stalks	Ears	Stalks	Ears	Mean ± SE % damaged plants with live ECB†
May 12	August 12	6.7 ± 2.3a	1.1 ± 1.6a	32.3 ± 5.3a	3.6 ± 1.0a	34.2 ± 6.6a
June 11	September 3	63.8 ± 6.5b	19.0 ± 1.6b	73.0 ± 4.8b	37.0 ± 3.3b	92.9 ± 1.1b
July 10	October 9	43.8 ± 7.8b	9.0 ± 2.0c	59.7 ± 2.1c	19.0 ± 2.1c	82.1 ± 7.4b

* Means within columns followed by the same letter are not significantly different ($P > 0.05$) as determined by Duncan's (1955) multiple range test.
† Percentages within columns followed by the same letter are not significantly different ($P > 0.05$) as determined by chi-square contigency table analysis.

NOTE: These are not author's errors. The author had marked these changes in the final proof, and the correction was made on the final galley; however, when the final typeset copy of the sideways-tablular material was produced, the old disk was inadvertently utilized. The editor regrets any inconvenience caused by these errors.

ERRATUM

In the Journal of Agricultural Entomology, Volume 5, Number 3, July 1988, the data in Table 2 on page 158 were inadvertently reversed in the two columns under "Means ± SE % damaged†"; under "Ears" in the column "Mean ± SE no. ECB/50*," the first entry should be "1.1 ± 1.6a." The following is the corrected table in its entirety:

Table 2. ECB densities and feeding damage at harvest in early-(May), mid-(June), and late-(July) season plantings of hybrid sweet corn at Hamden, Connecticut 1986.

		Mean ± SE no. ECB/50*		Mean ± SE % damaged†		
Planting date	Harvest date	Stalks	Ears	Stalks	Ears	Mean ± SE % damaged plants with live ECB†
May 12	August 12	6.7 ± 2.3a	1.1 ± 1.6a	32.3 ± 5.3a	3.6 ± 1.0a	34.2 ± 6.6a
June 11	September 3	63.8 ± 6.5b	19.0 ± 1.6b	73.0 ± 4.8b	37.0 ± 3.3b	92.9 ± 1.1b
July 10	October 9	43.8 ± 7.8b	9.0 ± 2.0c	59.7 ± 2.1c	19.0 ± 2.1c	82.1 ± 7.4b

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ERRATUM

The following reference was inadvertently omitted from the index on the front cover of the Journal of Agricultural Entomology, Volume 5, Number 3, July 1988:

COLEMAN, RUSSELL E, and REID R. GERHARDT — Frequency of regurgitation by laboratory-reared face flies fed trypticase-soy broth 185

The editor regrets any inconvenience caused by this omission.

COCHRAN SCHOLARSHIP PATRONS[1]

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[1] The J. H. Cochran Memorial Scholarship is awarded annually by the South Carolina Entomological Society to a deserving graduate student. Donors who contribute $1,000 or more to the Cochran Scholarship Fund are recognized by the Society as Cochran Scholarship Patrons. Such donations continually support graduate work in entomology since only the annual interest is used to support the award.

INFORMATION FOR AUTHORS: VOUCHER SPECIMENS

Voucher specimens are preserved organisms (or parts thereof) that serve as a future reference for a name used in a scientific publication. Voucher specimens ensure the credibility and endurance of research results because they document the identity of studied organisms. Therefore, although not required for publication in the Journal of Agricultural Entomology, the preservation of voucher specimens is recognized by the Editorial Board as one of the most important responsibilities when publishing research. Accordingly, the Board has adopted the following policy statement:

> 'Authors are encouraged to designate, properly prepare, label and deposit high quality voucher specimens and cultures documenting their research in an established permanent collection and to cite the repository in publication.'

Recommended Procedure:

(1) The author should contact the curator of the voucher repository prior to deposition to assure that the collection will accept voucher material and to learn specific procedures required by that repository. (Note that in accepting voucher specimens, the repository assumes responsibility for protecting them from casual use or exchange and for encouraging their review by future taxonomic specialists. For performing this service, most repositories will appreciate the author's assistance in obtaining material and financial support in order to help maintain the scientist's reseach-vouchering specimens.)

(2) The number of voucher specimens designated should be sufficient, in the author's opinion, to represent the range of variation normally expected in the studied organism, but the number should not be so great that limited space and technical assistance in the repository are strained. In insects, for example, five to ten specimens each of males, females, pupae, larvae, and eggs (when available) of each species usually is adequate. In cytological or electrophoretic studies, photographs or permanent preparations should also be designated as vouchers.

(3) Specimens should be mounted and preserved using the standard, accepted techniques and *each* voucher specimen or specimen vial should have the following information provided at the time of deposition: (a) full, standard collection data (such as date, locality, and ecological habitat or host from which the specimen was collected, collector's name, and whether or not the specimen is from a laboratory culture), (b) the identification, identifier, and date of identification, and (c) a label (often green) designating the specimen as 'VOUCHER' (often including an access number and/or the name of the senior author and an abbreviated publication reference).

(4) Authors should include in the text of their JAE article, usually in the 'Materials and Methods' section, a statement naming the voucher repository, the number of voucher specimens deposited, any access numbers, and any noteworthy particulars about their preservation or labeling.