FIELD EVIDENCE FOR AN ATTRACTANT PRODUCED BY THE MALE PEPPER WEEVIL (COLEOPTERA: CURCULIONIDAE)

R. J. PATROCK¹, D. J. SCHUSTER² AND E. R. MITCHELL³

¹Department of Zoology

University of Texas

Austin, TX 78712

²Gulf Coast Research & Education Center University of Florida, IFAS 5007 60th Street East Bradenton, FL 34203

³Insect Attractants and Basic Biology Research Laboratory Agricultural Research Service, U.S. Department of Agriculture Gainesville, FL 32604

ABSTRACT

A small-scale field trapping experiment using caged *Anthonomus eugenii* Cano weevil adults in modified PMS Survey boll weevil traps was performed at the Gulf Coast Research and Education Center in Bradenton, Fla. A significantly higher percentage of male-baited traps captured conspecific weevils than did female-baited or unbaited traps. Males and females were captured in all traps in similar proportions over the study. PMS traps alone also were somewhat attractive to the weevils. These field results support earlier laboratory results indicating pepper weevil males produce an aggregation pheromone.

RESUMEN

Un experimento de campo utilizando trampas modificadas de *Anthonomus grandis* ("PMS Survey") fue realizado para capturar adultos de *Anthonomus eugenii* Cano. Las trampas con machos capturaron más adultos de la misma especie que las trampas con hembras o el control. La proporción de machos y hembras capturadas en las trampas fue similar. Estos resultados coinciden con experimentos de laboratorio que indican que *Anthonomus eugenii* produce una feromona de agregación.

Effective control of the pepper weevil, Anthonomus eugenii Cano, is hindered by problems in detecting field populations before economic injury (Genung & Ozaki 1972). Visual sampling currently is used to time pesticide applications on small-scale farms for this pest in Honduras (Andrews et al. 1984). Sticky traps were found to be superior to other sampling methods in Puerto Rico (Segarra-Carmona & Pantoja 1986) but the relationship between population density and trap catch was weak and erratic, particularly at low densities. Cartwright et al. (1990) found that the percentage of bud clusters damaged by adults was correlated with adult density and could be used as an action threshold. Waiting until damage has already occurred before applying an insecticide may allow an infestation to develop protected in buds and small fruit.

Pheromonal trapping is considered to be an effective and economical method of detecting pests, particularly in the early part of the season when pest population levels

are low (Klassen et al. 1982). While it is likely that pheromones play some role in the behavior of most insects, a necessary step in determining the feasibility of using pheromones for pest detection is to demonstrate attractancy, especially in the field (Roelofs 1979). Coudriet & Kishaba (1988) demonstrated in laboratory and field cages that males produced an airborne female attractant.

The objective of this study was to obtain information on pepper weevil attractancy and behavior in the field to support further the existence of a pepper weevil-produced pheromone system. The considerable research on the pheromonal complex and trapping of the related boll weevil (Hardee 1972) was used as a guide for this investigation.

MATERIALS AND METHODS

Handling of Pepper Weevil in the Laboratory

Adult pepper weevils used in the trapping study were collected as pupae from field-collected pepper fruit weekly between March and August 1984. To obtain cohorts of individuals of the same age, the pupae first were segregated according to red or white eye color. Young pupae had white eyes and older pupae had red eyes. These were placed in individual containers for emergence. All containers were used once or washed with 75% ethanol before holding other adults (Bradley et al. 1968). When the adults began walking, they were provided with host flowers and/or fruit every 1-2 days. The adults were held for five days or until they were used as experimental replacements. Initially, the sex of pepper weevil adults was determined using rostrum characteristics and the pygidium. Females typically have a more slender and less punctured beak than males, and the antennae are inserted farther from the mouthparts (Agee 1964). This method was unsuitable because of overlap in these features among some adults. Therefore, only specimens with the above features well-defined were used. Later, the sex of pupae was determined by examining the abdominal sexual characteristics described by Anderson (1968) for the boll weevil. Sex of adults was checked before individuals were used in the trapping study to limit errors in sexual determination.

Trap Design

Modified PMS Survey cotton boll weevil traps (Pest Management Specialists, Inc.) were used. A plastic 100 ml cup housed the bait insects and/or pepper flowers and fruit. Openings were cut (ca. 4 cm²) in the top and on opposite sides of the cup and were covered with organdy cloth for ventilation. The cup was inverted over the inspection dome of the trap. The top one-quarter of the dome was removed and the resulting hole covered with organdy to facilitate diffusion of volatile materials through the trap into the environment. To prevent the escape of captured insects, the original aeration holes in the inspection dome were closed slightly with hot-glue. Four 1-cm diameter holes were drilled in the top of the trap base to allow for movement of pepper weevil from beneath the base (Dickerson et al. 1981).

Field Plots

The trapping study was conducted at the University of Florida, Gulf Coast Research and Education Center (GCREC) in spring and summer of 1984. Peppers (cv. Early Calwonder) were transplanted on February 20 and 28. Four beds were planted on the first date and three beds on the second date in anticipation of late frosts and to extend the fruiting period. The plants were set in single rows (two plants per hole) on raised black plastic mulched beds at ca. 30.5 cm intervals. The beds were ca. 92 m long and

0.8 m wide. Fertilizer was incorporated into the EauGallie fine sand beds. Before the plastic mulch was installed, the beds were fumigated with methyl bromide (67%) and chloropicrin (33%) for disease, weed, and nematode control. Other pesticides were not used on the plants during the study period. Water was provided by seepage furrow irrigation.

Field Setup

Trapping was initiated on March 21 when the plants began to fruit. A completely randomized design with five replications was used from March 21 to May 23. On May 24, the arrangement of the traps in the field was changed to a randomized complete block design and the number of replicates reduced to three. The traps were set at ca. 9 m intervals. The height of the traps was adjusted with the growth of the pepper plants so that the bottom of the yellow base was approximately even with the top of the plants. Three trap treatments were used: 1) pepper weevil male with food (pepper flowers and fruit); 2) pepper weevil female and food; and 3) pepper flowers and fruit only. To determine whether pepper weevils were attracted to the trap alone (i.e., when it did not contain an adult or food), an empty trap was placed in each block May 31 to June 20.

Five-day-old adults were placed in the trap housings in the field on Wednesday of each week. The traps were examined at least four times each week, and the flowers and fruit were changed at each inspection. If a caged adult had died, the trap was changed and the dead insect was replaced with a similarly aged adult. After May 2, 70% ethanol was used to clean hands following the handling of pepper weevil and traps to reduce the possibility of contamination. The traps were changed at the end of each week and washed in 70% ethanol before being used again in the field. After May 17. the traps were changed and washed after each inspection to reduce the possibility that field insects were responding to insects previously captured (Van Cleave & Harp 1971). After each inspection, the traps were rotated within the field (before May 23) or in the block (after May 23) to reduce positional bias. Captured insects were counted and transported to the laboratory for sex determination. After May 23, traps in blocks with bait insect mortality or loss between inspection dates were not counted. Rostrum and pygidium characteristics were used in sex evaluation if genitalia were not exposed. A one-way analysis of variance applied to ranks was used to compare the percentage of traps capturing adults, using SPSS (SPSS Inc.). This test, which is equivalent to the Kruskel-Wallis non-parametric test (Ray 1982), was used due to the change in experimental design and the heterogeneous population variances of the season-long data. The numbers of adults captured after May 23 were analyzed by ANOVA (Ray 1982) combining captures for each trapping interval and using trapping intervals as blocks.

Visual Sampling of Adults

Visual sampling of adult pepper weevil on pepper terminal leaf clusters was conducted weekly from March 1 to June 20 (Andrews et al. 1986). The purpose of this sampling was to compare adult density to counts of trapped pepper weevil adults.

The plants were inspected between 0800 and 1000 hr. Two procedures were used during the study. Between March 1 and May 18, one set of 40 terminals was sampled around each of the fifteen traps (i.e., 20 terminals from 20 plants were sampled on the two rows adjacent to each trap). The sampling program was modified to a systematic sampling after May 22 because of the change in the trapping experimental design. First, 40 terminals were sampled on pepper plants on beds next to a randomly assigned aisle. Twenty terminals per 20 plants on each of the beds were inspected. This procedure then was continued in successive aisles until 15 samples had been taken.

RESULTS AND DISCUSSION

A significantly higher percentage of male-baited traps captured pepper weevil adults than did female-baited or control traps between March 21 and June 20 (Table 1, P =0.0189). There was no significant difference between the percentages of female-baited and control traps that captured adults. Similar results were obtained considering the numbers of adults captured during the last four trapping intervals (Table 1). Males and females were attracted to the male-baited trap in approximately equal numbers (1.9 \pm 3.4(SD) and 2.5 ± 3.8 per trap per week, respectively). Therefore, our data suggest that the male-produced attractant may have a dual sex attractant and aggregation function. This is contrary to Coudriet & Kishaba (1988) who found mostly females responding in laboratory and field cage studies. These authors used callow adults in all of their studies and didn't use plant material in combination with the adults in the traps in their field cage studies. Therefore, there may have been a response of adults to the pepper flowers and fruit included in the traps in our study. However, a total of 15 adults in 8 traps (7 males and 8 females) was captured in empty traps from May 31 to June 20. This number is similar to the number of adults captured in control traps (18) and female-baited traps (13) during the same period. These results indicate that pepper flowers and fruits were not attractive but that the boll weevil trap may be inherently attractive to the pepper weevil, possibly because of the yellow color. Yellow and white sticky traps were shown to be more attractive to the pepper weevil in the field than six other colors in Puerto Rico (Segarra-Carmona & Pantoja 1988). Other Anthonomines (e.g., Anthonomus captivus Clark) also are known to be attracted to some aspects of boll weevil traps (Clark 1988).

The highest number of trapped adults was recorded in May and June (Table 2). This coincided with a decline in the number of adults observed on terminal leaves (Fig. 1) as well as with a decline in the numbers of flowers and fruit on the plants. The relationship between the percentage of traps capturing adults and counts of adults by visual sampling is linear and significant (Fig. 2). The increased numbers of pepper weevil adults captured in late May and in June might be attributed to increased movement of adults in search of feeding and ovipositional sites, or the change in the trap arrangement.

The low number of pepper weevils captured in the traps precluded detailed observations of adult movement towards or on the trap. Nevertheless, some observations were made in the field and in small cage tests in the laboratory. Adults were observed flying and landing on the yellow base of the trap on several occasions. These adults walked on the outside of the base. One adult walked into the funnel but after a few minutes

TABLE 1. CAPTURES OF PEPPER WEEVIL ADULTS (ANTHONOMUS EUGENII CANO) WITH MODIFIED PMS BOLL WEEVIL TRAPS WHEN THE TRAPS WERE BAITED WITH ONE MALE OR FEMALE PEPPER WEEVIL ADULT PLUS PEPPER FLOWERS AND FRUIT OR A CONTROL TRAP BAITED WITH PEPPER FLOWERS AND FRUIT ONLY.

Treatment	Percentage of traps capturing A. eugenii ^a	Avg. no. per trapping interval ^b	
Male	26.8 a ^c		
Female	15.8 b	4.8 b	
Control (food only)	17.3 b	$6.5\mathrm{b}$	

^aPercentages ranked for analysis but presented in original scale.

^bData transformed square root of X + 0.5 before analysis but presented in the original scale.

 $^{^{\}circ}$ Means in columns followed by different letters differ significantly at the P = 0.05 level by Duncan's multiple range test.

TABLE 2. WEEKLY CAPTURES OF ADULT PEPPER WEEVILS, ANTHONOMUS EUGENII CANO, USING MODIFIED PMS BOLL WEEVIL TRAPS BAITED WITH ONE ADULT MALE OR FEMALE PEPPER WEEVIL PLUS PEPPER FLOWERS AND FRUIT OR WHEN BAITED WITH PEPPER FLOWERS AND FRUIT ONLY. MANATEE COUNTY, FLORIDA. 1984.

Date	Male baited trap		Female baited trap		Control trap	
	Males	Females	Males	Females	Males	Females
3/22-3/28	0	2	0	0	0	0
3/29-4/4	0	0	0	0	0	Ô
4/5-4/11	0	0	0	0	1	0
4/12-4/18	1	2	1	0	$\bar{1}$	Õ
4/19-4/26	0	1	0	0	0	Õ
4/27-5/2	1	4	0	0	0	Ô
5/3-5/9	3	4	1	3	1	2
5/10-5/16	0	4	0	0	0	$\overline{0}$
5/17-5/22	3	7	3	2	1	4
5/23-5/30	5	9	3	3	3	5
5/31-6/6	14	21	3	2	4	5
6/7-6/13	15	1	2	3	Ō	5
6/14-6/20	6	6	0	3	2	$\overset{\circ}{2}$
Total	4 6	61	11	16	12	$\overline{21}$

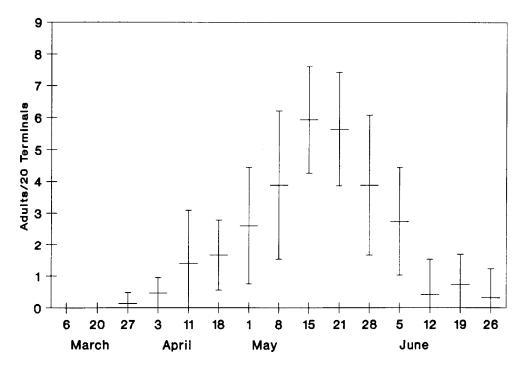


Fig. 1. The number of pepper weevil adults sampled weekly on terminals of bell pepper grown in the field. Manatee County, Florida. 1984.

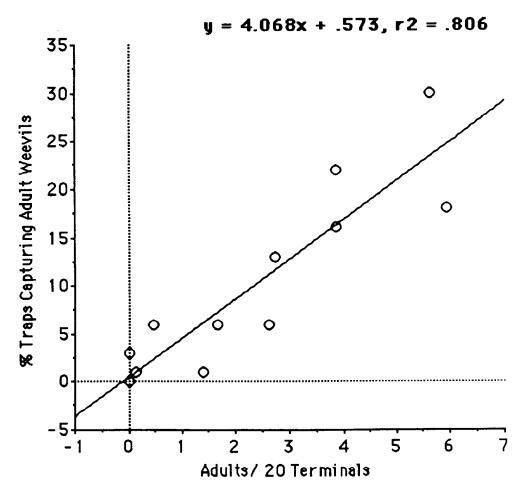


Fig. 2. The relationship between the number of pepper weevil adults observed per 20 pepper terminals (x) and the percentage (y) of modified PMS boll weevil traps capturing pepper weevil adults.

returned to the trap base. This observation was made on a male-baited trap. In all cases, the insects flew away after a short period. Adults were seen walking on the inside of the yellow base as well as on the stick supporting the trap. These observations formed the basis for drilling the holes in the top part of the yellow base as suggested by Dickerson et al. (1972). In laboratory cages holding various numbers of adult pepper weevils, adults were seen moving in and out of the inspection dome. While this behavior was not observed in the field, it is likely that it did occur to some degree.

The results of this study and that of Coudriet & Kishaba (1988) indicate that the male pepper weevil produces an aggregation pheromone. The response of wild pepper weevils to the traps in this study was substantially less than that reported by Coudriet & Kishaba (1988) who used laboratory-reared weevils in field cages from which all vegetation had been removed. This difference probably is the result of many factors, including the larger number of bait weevils used in that study (5 vs. 1), and the openness of the environment.

ACKNOWLEDGMENT

Published as Florida Agricultural Experiment Station Journal Series No. R-01390.

REFERENCES CITED

- AGEE, H. R. 1964. Characters for determination of sex of the boll weevil. J. Econ. Entomol. 57: 500-501.
- Anderson, D. M. 1968. Observations on the pupae of Anthonomus grandis grandis Boheman and A. grandis thurberiae Pierce (Coleoptera:Curculionidae). Ann. Entomol. Soc. Amer. 61: 125-129.
- Andrews, K. L., A. Rueda, G. Gandini, S. Evans, A. Arango, and M. Avedillo. 1986. A supervised control programme for the pepper weevil, *Anthonomus eugenii* Cano, in Honduras, Central America. Tropical Pest Management 32: 1-4.
- BRADLEY, J. R., D. F. CLOWER, AND J. B. GRAVES. 1968. Field studies of sex attraction in the boll weevil. J. Econ. Entomol. 61: 1457-1458.
- CARTWRIGHT, B., T. G. TEAGUE, L. D. CHANDLER, J. V. EDELSON, AND G. BENTSEN. 1990. An action threshold for management of the pepper weevil (Coleoptera:Curculionidae) on bell peppers. J. Econ. Entomol. 83: 2003-2007.
- CLARK, W. E. 1988. Revision of the *furcatus* species group of the weevil genus *Anthonomus* Germar (Coleoptera: Curculionidae). Coleopterists Bull. 42: 359-377.
- COUDRIET, D. L., AND A. N. KISHABA. 1988. Bioassay procedure for an attractant of the pepper weevil, (Coleoptera: Curculionidae). J. Econ. Entomol. 81: 1499-1502.
- DICKERSON, W. A., G. H. MCKIBBEN, E. P. LLOYD, J. F. KEARNEY, J. J. LAM, JR., AND W. H. CROSS. 1981. Field evaluation of a modified in-field boll trap. J. Econ. Entomol. 74: 280-282.
- GENUNG, W. G., AND H. Y. OZAKI. 1972. Pepper weevil on the Florida East Coast. Univ. Fla. AREC Belle Glade Mimeo Rept. EV-1972-2, Belle Glade. 15 pp.
- HARDEE, D. D. 1972. A review of literature on the pheromone of the boll weevil *Anthonomus grandis* Boheman (Coleoptera: Curculionidae). USDA Coop. Econ. Insect Rep. 22(14): 200-207.
- KLASSEN, W., R. L. RIDGEWAY, AND M. INSCOE. 1982. Chemical attractant in integrated pest management programs, pp. 13-130 in A. F. Kydonieus and M. Beroza, [eds.], Insect suppression with controlled release pheromone systems. Vol. II, C.R.C. Press, Boca Raton.
- RAY, A. A. 1982. SAS user's guide: Statistics. SAS Institute Inc., Cary, NC. 584 pp.
- ROELOFS, W. L. [ed.]. 1979. Establishing efficacy of sex attractants and disruptants for insect control. Entomological Society of America, College Park, MD. 97 pp.
- SEGARRA-CARMONA, A. E., AND A. PANTOJA. 1988. Evaluation of relative sampling methods for application estimation of the pepper weevil, *Anthonomus eugenii* Cano (Coleoptera: Curculionidae). J. Agric. Univ. P.R. 72: 387-393.
- VAN CLEAVE, H. W., AND J. J. HARP. 1971. The pecan weevil: present status and future prospective. Proc. Southeast Pecan Growers Assoc. 64: 9-111.