TARNISHED PLANT BUG PHEROMONE - OLFACTOMETER STUDIES

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Abstract

The research described here has been directed toward the development of an olfactometer to bioassay tarnished plant bug (*Lygus lineolaris* Palisot de Beauvois) (TPB) sex attractant. Plans for the olfactometer, conditions under which the TPBs are held, considerations in the development of the bioassay procedure, and results from bioassays are discussed. We developed a reliable olfactometer for the project and established a bioassay procedure that generates positive male TPB olfactory responses to females.

Introduction

The effects of tarnished plant bugs (*Lygus lineolaris* Palisot de Beauvois) (TPBs) on cotton plants and on cotton production have been well documented by Scales & Furr (1968); Hanney et al. (1977); and others. The isolation, identification, and synthesis of pheromones has been an important development in the control of insects such as the gypsy moth (Burgess 1964), pine sawfly (Coppel et al. 1960), and cotton boll weevil (Hardee 1967). The identification of a TPB pheromone for use in traps should be of great benefit for a pest management program directed at this insect.

This research effort has been directed toward the development of a reliable olfactometer to bioassay the TPB sex attractant. Considerations in the development of the bioassay procedure, plans for the Olfactometer, and data from its use are presented.

Procedures and Results

There are a number of considerations concerned with the development of a bioassay procedure other than the olfactometer. These include temperature, light, time-of-day, age of test insects, noise, human presence, and perhaps isolation from other insects of their own species and sex.

Conditions under which the TPBs are held prior to testing are important for consistent results from olfactometer tests. They are sexed as ca 1-day- old adults and placed in cardboard containers at 26.5°C., 80% RH, 16:8 LD cycle, and are fed green beans daily. The green beans are purchased from a local supermarket and are washed in distilled water before use.

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In our laboratory we observed that TPBs begin to lay eggs at ca 7 days of adult age and live for ca 40 days. We also observed that they produce a noticeable alarm pheromone when they are disturbed. Because of these factors we set the following conditions for pheromone tests: 1) tests are conducted on adults that are 10 days of adult age or older; and 2) they are placed in the olfactometer 30 minutes before the tests begin and are not disturbed by humans during the test.

Figure 1 shows the dimensions of the olfactometer. It is constructed from one-eighth inch plate glass and glued with silicone sealant. After construction the unit was allowed to cure for ca 1 week and was cleaned with alcohol and distilled water before use. Likewise, the unit was cleaned in the same manner before each test.

Figure 2 shows the video camera above the olfactometer. It is connected to a monitor in an adjacent room. This video camera/monitor arrangement allows observation of the test insects without human interference. Figure 3 shows a view of the olfactometer. The cages that are used to hold females in these tests are attached to the sides (Fig. 4). Air flow goes through the filter/dryers (Fig. 5) which are connected to the ends of the female cages, through the part of the olfactometer containing males, and to the tube in the top center of the olfactometer. This tube is connected to a vacuum pump. Air flow is set at 5 liters per minute going into the vacuum pump.

In the initial tests it was important to determine that male insects respond to females under our conditions and that the olfactometer produces consistent results. The insects are placed in the test apparatus 30 minutes before the beginning of the test with the vacuum pump in operation. They are not disturbed after this. During a test one of the cages contains the female and the other cage is empty. Two males are placed in the central part of the unit and generate a positive response by moving to the cage containing the female. A negative response consists of movement to the empty cage. The males have equal chance to move to the empty cage. Figure 4 shows a male approaching the cage containing the female. If a male crawls within 1 cm of the front of a cage and stops for 15 seconds, or crawls onto the front of the cage itself, this is considered a response.

Figure 6 shows the video monitor in an adjoining room where a technician records test results. Insects are placed in the olfactometer two hours after lights in the holding incubator are switched on. The test is then conducted for 4 hours.

Eight tests were conducted with 40 positive responses and 20 negative responses. Analysis by t test showed this to be a highly significant response by males to females ($p < 0.01$).

These results indicated to us that male plant bugs are responding to pheromone released by females in our

olfactometer. We are now ready to trap the pheromone for chemical identification. For this effort, females will be placed in the cages as in the previous tests, but no males will be used. A column of active substrate to trap the pheromone will be placed in the tubing going from the olfactometer to the vacuum pump. A solenoid (Fig. 5) will be used to switch the airflow through the substrate if a test is in progress, or around it if the olfactometer is being vented. This solenoid can be controlled at the video monitor.

Discussion

Herein we have described results in our continuing effort to isolate and identify the TPB sex pheromone. We have developed an olfactometer for the project, established a test procedure that generates positive responses from male test insects to females, and are now ready to trap volatile compounds released by females.

Acknowledgement

The authors wish to acknowledge the contributions of Mr. John R. McCoy in the development of the olfactometer used in these projects.

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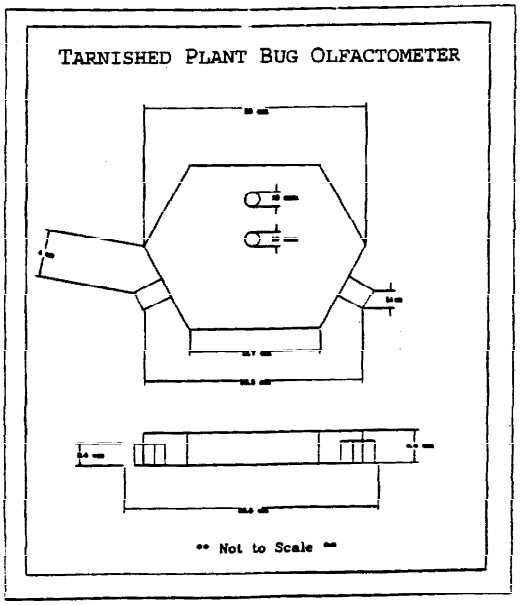

Figure 1. Schematic drawing of TPB olfactometer.

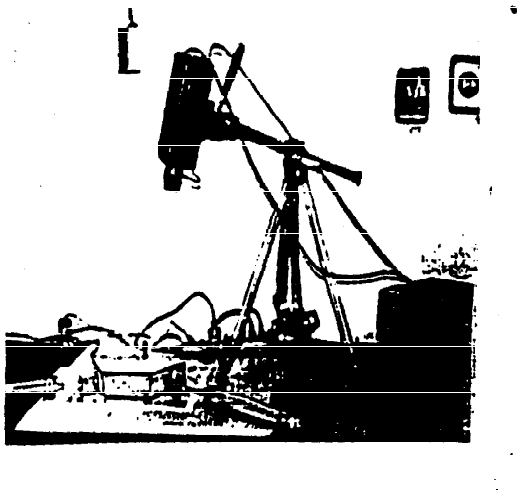

Figure 2. Olfactometer with video camera.

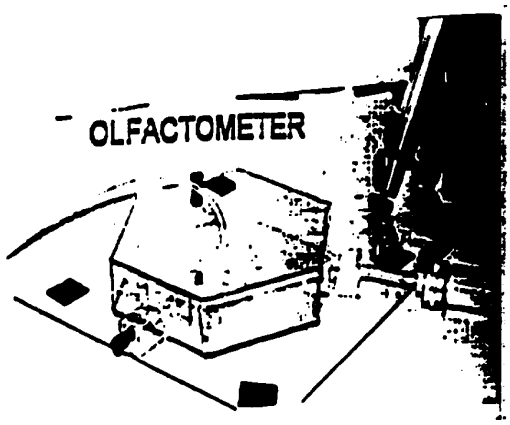

Figure 3. Close up view of olfactometer.

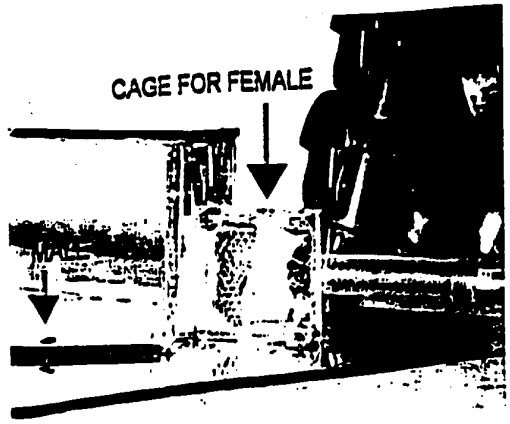

Figure 4. Close up view of female cage with female inside and male approaching.

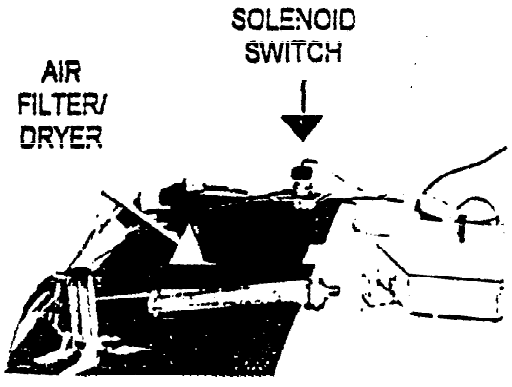

Figure 5. View of solenoid switch and air filter/dryer.

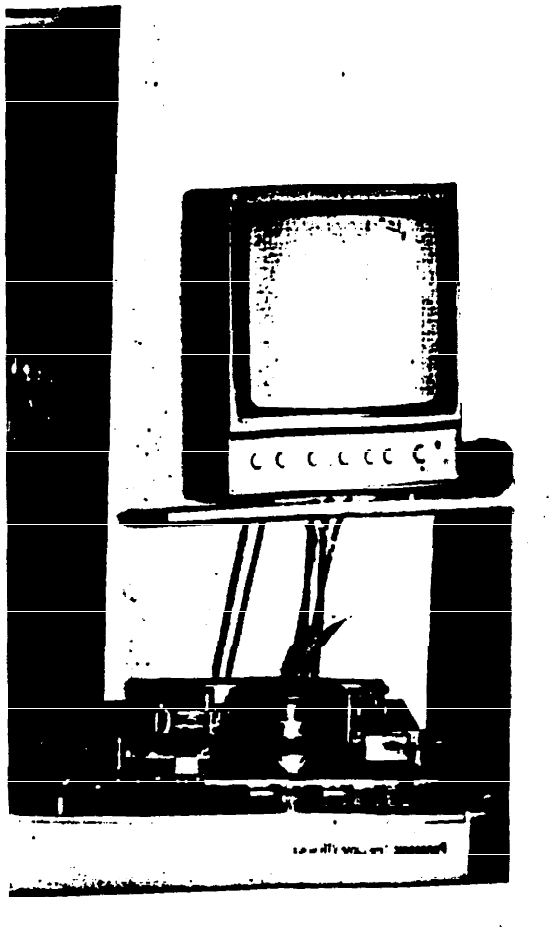

Figure 6. View of video monitor.