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A Laboratory Technique to Evaluate Boll Weevil Oviposition Preference Among Cotton Lines[1]

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ABSTRACT

A technique was developed to evaluate a large number of cotton lines in a minimum of time as oviposition sites for the boll weevil. Mississippi field weevils and the A & M laboratory strain were tested for a comparison of their response to various cotton lines. We concluded that: *(1)* Evaluating cotton lines with the laboratory strain of weevile would closely approximate the cotton varietal differences evident in evaluating with field weevils; *(2)* Boll weevil oviposition was an insect biological response which could be modified by the host plant genotype; and *(3)* 'Triple Hallmark Sea Island,' 'Seaberry Sea Island,' their F_1 hybrid, 'Brown Egyptian', and Seaberry Sea Island × Brown Egyptian were less preferred for oviposition by the boll weevil than 'Deltapine Smooth Leaf.'

Additional index word: resistance.

THE boll weevil, *Anthonomus grandis* Boh., is a destructive insect on cotton, *Gossypium hirsutum* L., in the raingrown cotton belt. The weevil first entered this country from Mexico in 1892 and by 1922 had covered the entire cotton belt. Knipling (9) estimated a $300 million annual loss from the boll weevil plus insecticide costs estimated at $70 million annually.

Control of this insect has depended upon rapid-fruiting, early maturing cotton varieties combined with an adequate insecticidal control program. Recently we began research in host plant resistance to the boll weevil. If a cotton variety resistant to the boll weevil could be developed, it would lower the cost of production.

Cotton is the only cultivated crop plant on which the boll weevil feeds and oviposits; however, there are several plants which have been classified as alternate hosts (Lukefahr and Martin, 10). All of these occur in the wild state and are of no major economic importance.

This close association between an insect and a single cultivated crop plant has stimulated numerous research projects. Cushman (3) presented data on oviposition period, total eggs deposited, daily oviposition, and time from emergence to oviposition. Everett and Ray (5) described oviposition as "a series of sequential reflex actions each triggered by specific stimuli." The weevil oviposits eggs in the cotton bud (square). Fenton and Dunham (6) found that squares 6 days old up to 3 days prior to bloom were preferred for oviposition sites.

Since oviposition is basic to population dynamics, a line of cotton which is less preferred by the boll weevil for oviposition purposes should reduce the boll weevil problem. Early work suggests that this type of non-preference might exist in some lines of cotton. Hunter and Hinds (7,8) collected more weevils from Sea Island cotton than from American Upland. Smith (11) found Upland squares were preferred over Sea Island for oviposition; however, the reverse was true for bolls.

Cook and Doyle (2) attributed the susceptibility of Sea Island to the thin, tender carpel of the bolls. Black and Leigh (1) in a study of five widely variable cotton types—'Deltapine 15,' 'Hopi Moencopi,' 'Sea Island Short Sympodia,' *Gossypium arboreum,* and *G. herbaceum* found a significant difference in feeding and oviposition. The oviposition rate was higher on Hopi Moencopi and Sea Island Short Sympodia.

The results obtained by these workers, plus field observations made in the past, prompted research to develop an adequate test for evaluating cotton lines for oviposition suppression qualities.

MATERIALS AND METHODS

Buds (squares) for the laboratory tests were collected from field plots of cotton. Both plots in Experiment I were planted at the same time the first year. All plots in Experiment II were planted at the same time the second year. All plots in both experiments were handled alike during the growing season. Organic phosphate insecticides, which provide quick kill and short residual action, were used to control natural boll weevil infestations. The applications were timed so that insecticide residue would not interefer with the tests.

A laboratory strain and a Mississippi field strain of weevils were used to develop the test procedure. The laboratory weevils were supplied by the mass rearing section at the Boll Weevil Research Laboratory. The strain, developed at Texas A & M University, is very useful in laboratory screening. Phenotypically, the weevil closely resembles its undomesticated counterpart. It differs mainly in being much more adaptable to laboratory use, being easily reared, and ovipositing more eggs. The Mississippi field strain was from a general field collection of weevils. Fallen, infested squares were collected and placed in a large emergence chamber and the newly emerged weevils collected daily. Mississippi field weevils, though not as well adapted as the laboratory strain, were used for laboratory testing to compare the field strain's response to various varieties tested. Both strains were conditioned for 5 to 7 days on 'Deltapine Smooth Leaf' (DPSL) squares before they were used for testing. Weevils tested before this age and conditioning period were often extremely erratic in their behavior during test periods. The 5- to 7-day-old, conditioned weevils gave a much more uniform response.

All oviposition testing was conducted in the laboratory. The test cage was a ¼-liter (½ pt.) glass jar sealed with screen wire lid. Five female and one male weevil were placed in each cage containing five squares. Squares from each cotton line were collected, brought into the laboratory, the bracts removed, and roughly sized to reduce variability resulting from variable size squares. Squares were changed in the cages at 10 AM and 3 PM. At each change the cages were carefully checked for the full complement of weevils. Each cage of insects was thus exposed to 10 fresh squares each day. Data consisted of the number of eggs in the squares. The two readings were added to give a total oviposition value per cage per day.

Preliminary records were taken by dissecting each square under a microscope and counting the eggs. This procedure was laborious, especially when a large number of lines were being tested. Everett et al. (4) had proposed a method for estimating

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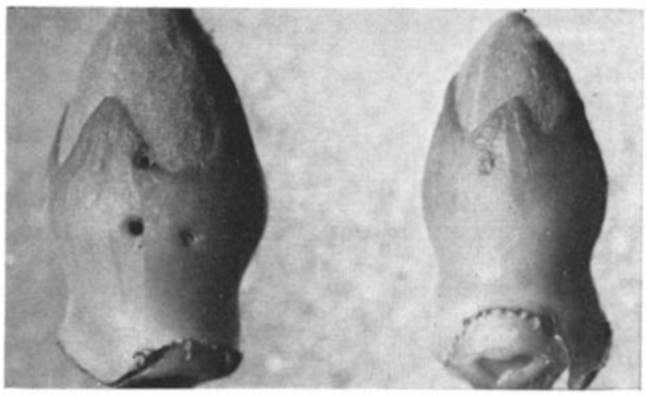

Figure 1. The square on the left shows unsealed feeding punctures. The square on the right shows a sealed oviposition puncture.

eggs. The female boll weevil secretes a substance which seals her egg puncture in the square (Fig. 1). Utilizing this characteristic for increased counting speed, one could count the sealed punctures on the square and arrive at a rapid estimation of oviposition while sacrificing only a small deviation from the actual number of eggs oviposited. Everett's counting technique was evaluated and verified as adequate for our oviposition testing.

The test procedure we developed was nsed in Experiment I in a comparison of two lines of cotton using the two strains of weevils. Four cages of insects were used for each cotton line and the data were collected for 6 days. 'Pima S-2' (*G. barbadense*) and Deltapine Smooth Leaf (*G. hirsutum*) were chosen initially in order to test the reported preference of weevils for *G. barbadnse* over *G. hirsutum*. Hunter and Hinds (7,8) stated that Sea Island cotton was preferred over Upland for oviposition. Although the early writings did not classify the cotton further than the broad term, Sea Island, early writers used the term to encompass all *G. barbadense* types grown on the Southeastern sea coast islands. This preference for Sea Island should be verified by comparing Pima S-2 with Deltapine Smooth Leaf in our test.

In Experiment II, six lines, 'Triple Hallmark Sea Island,' 'Seaberry Sea Island,' their F_1 hybrid, 'Brown Egyptian,' Seaberry Sea Island $\times$ Brown Egyptian, and Deltapine Smooth Leaf were tested with the two strains of weevils to further substantiate the validity of testing with laboratory-reared weevils. The experiment was replicated four times for each test. There were four tests involved in this experiment. Test no. 2 was an 8-day test with data being collected each day. Tests no. 3, 6, and 7 were 8-day tests with data being collected the last 3 days of each test. Tests no. 2, 3, and 6 were with the laboratory strain of weevils and test 7 was with the field weevils. It was verified that collecting data for the last 3 days of an 8-day test measured egg production on a cotton line as accurately as collecting data all 8 days the weevils were on test. These 4 tests were conducted at different times during the summer and the mean eggs per female per day were averaged for tests no. 2, 3, and 6 to give the data reported for the laboratory strain. Because of this, the two strains of weevils should be compared using the T/DPSL ratios rather than mean eggs per female per day. The mean eggs per female per day should be compared only between lines involved in the same tests. The T/DPSL ratio is obtained as follows: mean eggs per female per day on the test line divided by mean eggs per female per day on the DPSL line in the same tests. Thus, setting the mean egg production on DPSL at 1.00 gives a good method for comparisons between cotton lines and weevil strains tested at different times.

RESULTS AND DISCUSSION

Table 1 shows the mean number of eggs per female per day on each strain of cotton in Experiment I. The laboratory weevils had a higher oviposition rate on both cotton strains than the Mississippi field weevils. Mississippi field weevils oviposited 80% more on Pima than on Deltapine Smooth Leaf, whereas the laboratory weevils oviposited only 19% more.

Table 1. A comparison of oviposition by Mississippi field weevils and the laboratory strain of weevils on Deltapine Smooth Leaf and Pima S-2 cotton (4 replications).

		Mean eggs /female /day	Pima/DPSL ratio
Mississippi field weevils,	DPSL	2.72	----
	Pima S-2	4.88	1.80
Laboratory weevils,	DPSL	6.63	----
	Pima S-2	7.92	1.19

Table 2. A comparison of oviposition by Mississippi field weevils and the laboratory strain of weevils on six cotton lines (4 replications per test).

	Laboratory weevils				Mississippi field weevils			
	Mean eggs /female /day				Mean eggs /female /day			
Cotton line	Test no.	Test line	DPSL	T/DPSL ratio	Test no.	Test line	DPSL	T/DPSL ratio
THSI	3,6	9.45	12.41	.76	7	3.72	5.11	.73
SSI	2,3,6	8.54	11.03	.77	7	4.68	5.11	.91
THSI × SSI	2,3,6	8.31	11.03	.75	7	4.04	5.11	.79
BE	2,6	7.42	9.45	.78	7	3.58	5.11	.70
SSI × BE	2,3,6	9.21	11.03	.83	7	4.56	5.11	.89

THSI = Triple Hallmark Sea Island. SSI = Seaberry Sea Island. BE = Brown Egyptian. DPSL = Deltapine Smooth Leaf. T/DPSL = No. of eggs on test line/No. of eggs on DPSL.

Table 2 presents the results of Experiment II with laboratory and Mississippi field weevils. There was a good comparison of the T/DPSL ratio within lines between strains of weevils. The mean oviposition rate when compared between strains of weevils within cotton lines is not as uniform as the T/DPSL ratios. This is due to the higher egg production characteristic of the laboratory weevils and to the tests being conducted at different times. The T/DPSL ratio represents the relative worth of a particular line for oviposition by the weevil as compared to Deltapine Smooth Leaf. When this ratio is employed, a particular line may be compared directly between strains of weevils and with other cotton lines. The ratios revealed a reduced preference for all five cotton lines when compared to DPSL.

These tests led to three conclusions. *(1)* Evaluating cotton lines with the laboratory strain of weevils would closely approximate the cotton varietal differences evident in evaluating with field weevils. *(2)* Boll weevil oviposition was an insect biological response which could be modified by the host plant genotype. *(3)* Triple Hallmark Sea Island, Seaberry Sea Island, their F_1 hybrid, Brown Egyptian, and Seaberry Sea Island $\times$ Brown Egyptian were less preferred for oviposition by the boll weevil than Deltapine Smooth Leaf.

It would be very difficult to supply enough field weevils to carry out an extensive evaluation program but the weevil rearing section of our laboratory is capable of supplying the necessary weevils; therefore, we plan to use the laboratory strain in future mass screening of cotton lines.

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