HYBRIDS PRODUCED VERY ECONOMICALLY WITH TRANSGENIC COTTON J. B. Weaver, Jr. Professor Emeritus Athens, GA

<u>Abstract</u>

Seed harvested from a herbicide susceptible genotype open pollinated with a transgenic herbicide resistant strain can be planted and sprayed with the herbicide to kill all nonhybrids. A 3:1 ratio of rows of resistant to susceptible strains was used to increase the percentage of open pollinated hybrids obtained from the female parent. Roundup was sprayed during the flowering period on RR cultivars which caused a high level of male sterility along with slight female sterility when the plants were under drought stressed conditions.

Introduction

When scientists inserted various genes for herbicide resistance into cotton, they had no idea that they were also making it practical to produce hybrid cotton. All of the herbicide resistant genes are inherited as a dominant character. Thus, seed harvested from a herbicide susceptible genotype that has been cross pollinated by a resistant genotype can be planted, then sprayed with the appropriate herbicide, and all of the non-hybrids will be killed. The susceptible parent could be (1) a conventional fertile genotype, (2) a fertile genotype sprayed with a male gemetocide or (3) a genetic male sterile segregating in a 1:1 ratio of male fertile to male sterile plants.

In all three systems, the F_2 generation could be grown on a commercial scale. If only one herbicide resistance gene is involved, the F_2 would segregate in a 3:1 ratio of resistant to susceptible plants. The farmer would have to decide whether or not he wanted to randomally kill one-fourth of his plants along with the weed control expected with the herbicide. For many farmers, it would be desirable to reduce the plant population once a good stand was established.

Roundup applied to Roundup Ready (RR) genotypes has been found to be a far more satisfactory male gemetocide than previously used chemicals. A RR strain could be treated with Roundup and cross pollinated with a genotype that is RR and Buctril resistant. The F_2 would segregate 3:1 for Buctril resistance but would be homozygous RR.

The genetic male sterile $m_{s_5}m_{s_6}$ is being extensively used in India to produce F_1 hybrids. In the United States, it has not been practical to remove the fertile plants, so that no sib

crosses would occur between sterile and fertile plants. If most of the fertile plants were removed in a once over operation about one week after first flowering, the percentage of desired F_1 's could be 90 to 95%. When Buctril resistance is used as the pollinator parent, the seed harvested from the mostly male sterile rows could be treated with Buctril. Two or three days after emergence, the 5 to 10% sib crosses would die. The F_2 population would segregate 15:1 for fertile and male sterile plants. The male sterile plants would be adequately cross pollinated in perhaps 60% of the cotton acreage in the United States. Several years ago, it was found that male sterile plants when they were adequately cross pollinated.

Hybrid cotton (or use of heterosis) has been researched for 40 years. DeKalb Agricultural Research used a system of ten strains exposed for two years to high natural crossing by bumble bees. The "strain cross" that was marketed had about 55% heterozygosity which gave excellent seedling vigor and consistent superior yield. In 1961, a double recessive genetic male sterile (ms₅ms₆) was discovered. Although no 100% F₁ hybrid had been found that produced a higher yield than the "strain cross", DeKalb management insisted on having a 100% hybrid. The research operations were moved from Athens, Georgia to Arizona where it was intended to produce F₁ hybrids on rows of "stubbed" male sterile plants. Immediately the Department of Agriculture in Arizona ruled that stubbing cotton could no longer be practiced in Arizona. DeKalb management "threw out the baby with the wash water" and terminated all cotton research. The gemetocide FW-450 had been extensively tested but found to be unsatisfactory.

With the development of cytoplasmic male sterile (CMS) cotton and a fertility restorer enhancer factor, it appeared that hybrid cotton would be extensively grown. Although high yielding hybrids were found when grown under conditions where the plants reached a height of about two feet, these same hybrids performed poorly when the plants grew to about five feet in a "middle lapping" environment. Reciprocal crosses between plants with *G. hirsutum* cytoplasm and *G. harknessii* cytoplasm indicated that the *harknessii* cytoplasm reduced yield by about 12%.

More recently, Chem Brad attempted to use a gemetocide that was not satisfactory. They obtained about 50 to 60% F_1 and, after being advanced to the F_2 generation, there was only 25 to 30% heterozygosity.

Materials and Methods

In a six-acre field near Athens, Georgia in 1997, seed of DPL 90 RR (Deltapine Seeds) were placed in hoppers 1, 2, 5, 6, 7 and 8 of an eight row planter. Seed of SG 501 (Sure Grow Seeds) were placed in hoppers 3 and 4. This 3:1 ratio was chosen because the farmer had a two-row harvester and also because as the cross pollinating insects moved among

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the plants it was desired that they be carrying predominantly DPL 90 RR pollen. On one edge of the field, small plots of several susceptible strains were planted instead of SG 501.

Seed from the SG 501 and JBW 33 open pollinated with DPL 90 RR were sent to a winter nursery where they were sprayed with Roundup soon after emergence to kill the nonhybrids.

On another farm, six rows of BXN 47 were grown along with two rows planted with several Buctril susceptible strains, cultivars and lines segregating 1:1 for genetic male sterility. The fertile plants in the 1:1 populations were removed early in the flowering season. The cultivars DPL 90 RR, DPL 5690 RR and DPL 5415 RR were sprayed once per week with Roundup beginning at the early squaring stage and continuing until the end of August.

Seed harvested from both fields were grown in the greenhouse and sprayed with either Roundup or Buctril to determine the percent hybrids.

Results

A yield test with six replications was planted in 1998 on the University of Georgia Plant Sciences Farm with the cooperation of Dr. Hugh Earl. The seeding rate of the open pollinated genotypes was increased by 50 to 300 percent above the normal rate depending on the percentage of F_1 's previously observed in the greenhouse.

Open pollinated seedlings were sprayed at two weeks after emergence with either Roundup or Buctril. All plots were later hand thinned to a uniform stand.

The F_1 's and F_2 's showed excellent seedling vigor when compared to the cultivars. The 1998 growing season was much warmer than average with low rainfall. The yield test was irrigated three times. There was no significant insect damage.

One replication was hand harvested when about 85% of the cotton had opened in order to get a measurement of earliness. All other harvesting was done with a spindle picker.

Fiber test data were obtained from two replications.

Table 1 presents the yield and fiber test data. The three F_1 hybrids of DPL 90 RR, DPL 5690 RR and DPL 5415 RR x BXN 47 gave a 14.7% increase in yield over the three DPL cultivars. The fiber length and strength of the three F_1 's were essentially the same as the DPL cultivars. The micronaire values for these three hybrids tended to be lower than the DPL parents.

The SG 501 x DPL 90 RR F_2 gave essentially the same yield as F_1 of the three DPL RR x BXN F_1 .

Due to an oversight in preparing the yield test, the F_1 of SG 501 x DPL 90 RR was omitted from the test.

The JBW 33 x DPL 90 RR F_1 gave the lowest yield in the test. One possible explanation is that following seedling treatment with Roundup some of the leaves showed yellowing or abnormal color. The test site had Treflan applied preemerge and Cotoran postemerge. This discoloration did not occur on the same hybrids in a nearby field where neither Treflan nor Cotoran was used.

Due to the long hot summer, the expression of earliness, especially in some of the hybrids, was unfavorable for yield. In late August, the Chinese hybrid H-55 gave the impression that it would be the highest yielding entry in the test. If this same test had been grown in 1997 with below normal temperatures and an early frost, the earliness of the hybrids would have placed them much higher in yield ranking.

In 1997, the weekly application of Roundup on the three DPL RR cultivars caused continuous male sterility. There was a reduction in the number of seed per boll but excellent boll retention. It is estimated that the actual yield was reduced 20 to 25%. The percentage of hybrids with pollen coming from BXN 47 was 75 to 80%, which was the same as open pollinated seed harvested from genetic male sterile plants. The reason for observing less than 100% hybrids from BXN 47 was that about 25% of the plants in the crossing block did not have the Buctril resistance gene.

In 1998, BXN 47 and DPL 5690 RR were again grown in a crossing block in a 3:1 ratio. Only three applications of Roundup were applied to DPL 5690 RR. Most flowers in the DPL 5690 RR appeared to have no pollen. A slight reduction in number of seed per boll was found only in sections of the rows that were stressed due to inadequate irrigation water.

Monsanto has obtained a patent on the use of Roundup as a gemetocide.

Discussion

Herbicide resistance makes it very economical to produce F_2 's for large scale plantings. Additional research on Roundup as a gemetocide may make it possible for seed companies to produce and sell to farmers F_1 hybrids.

For the past 40 years, there has been a negative attitude among some individuals regarding the use of hybrid vigor in cotton in the United States. Statements such as "hybrids will not yield more than regular varieties," and "hybrids cannot be produced or they would be too expensive" have been made. The first hybrid corn grown in the southeastern US was corn belt hybrids and performed poorly in the southeast. Once hybrid corn Dixie 18 was released, it soon became widely grown in the southeast. A system is now available for utilizing hybrid vigor in cotton. the question now is, "Will the companies that have the patents on the herbicide resistance gene use these genes to produce hybrids; and, if not, will they prevent farmers in areas of high cross pollination from producing hybrids for their own farms?"

<u>Summary</u>

Cotton grown in 1997 in a 1:3 ratio of herbicide susceptible to transgenic herbicide resistant cultivars (BXN 47 or DPL 90 RR) gave over 25% natural cross hybrids in seed harvested from the susceptible rows.

Seed planted from the susceptible rows at 3X normal planting rate/A and treated with Buctril (or Roundup) resulted in all non-hybrids being killed and an adequate uniform plant population of F_1 hybrids.

The herbicide resistance system allows an inexpensive method of producing 100% F_2 plants that are all male fertile.

In a 1998 yield test, an F_2 produced essentially the same amount of lint/A as F_1 's, and both produced more lint than the parents.

Herbicide susceptible genetic male sterile ms_5ms_6 (1 fertile:1 sterile) insures a high percentage of F_1 's but would give some male sterile plants in the F_2 generation.

A very high percentage of F_1 hybrids were found when Roundup Ready cultivars were sprayed weekly with Roundup during the fruiting season. Roundup used as a gemetocide caused some reduction in seed/boll, especially in plants under drought stressed conditions. Table 1. Yield and fiber test data from test conducted on UGA Plant Sciences Farm.

	lbs lint				
Entry	per	% first			2.5%
	acre	pick	Mic	T ₁	SL
DPL90RRxBXN47 F ₁	1075	88	4.35	20.6	1.17
DPL5690RRxBXN47 F ₁	1052	90	4.20	20.8	1.16
SG501xDPL90RR F ₂	1038	85	4.55	20.9	1.16
DPL5415xBXN 47 F ₁	1023	88	4.50	19.2	1.15
PM(1.1MS)xBXN47 F ₁	1008	94	4.45	21.4	1.12
JBW33xDPL90RR F ₂	993	78	4.35	21.2	1.15
BXN47	990	71	4.60	18.8	1.13
SG501	957	79	4.60	22.2	1.14
H-55	944	94	4.40	17.2	1.08
DPL5415RR	936	86	4.50	20.6	1.16
DPL90RR	932	76	4.50	20.6	1.16
DPL5690RR	878	74	4.60	21.8	1.11
JBW(1:1MS)xDPL90RR F ₁	860	86	4.3	21.9	1.15
PM(1:1MS)xDPL90RR F ₁	860	86	4.3	21.9	1.15
Averages	968	84	493	20.69	1.15