Published March, 1971

Isolines in Cotton: Effects of Nine Dominant Genes[1]

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ABSTRACT

Nine isolines of cotton, *Gossypium hirsutum* L., were developed by backcrossing lines carrying dominant marker alleles at a total of nine loci to Texas Marker-1. The marker alleles were isolated into separate lines and backcrossed for eight generations to provide a common genetic background in which the effects of the individual marker loci could be evaluated and characterized. The effects of the qualitative characters on quantitative agronomic traits were determined for each line in a replicated performance test and compared to the performance of the recurrent parent. Certain quantitative traits of the isolines remained variable, and this variability was interpreted to be caused by linkage associations.

***Additional index words:* *Gossypium hirsutum*, Genetic markers, Pleiotropy, Linkage.**

IN 1956, a program was initiated to develop isogenic lines (isolines) in cotton, *Gossypium hirsutum* L. involving certain qualitative genetic markers. The program was established to provide a series of lines with a common genetic background in which the effects of individual marker loci could be evaluated and characterized. The isolines were developed to provide the basis for more effective use of these markers in genetic and agronomic studies.

The results of a performance test of the nine isolines are reported. The isolines were characterized by measurement of the effects of the qualitative characters on quantitative traits and by comparison with the performance of the recurrent parent. The variability of lines within each isoline was also determined.

MATERIALS AND METHODS

Two lines, carrying dominant marker alleles at a total of nine loci, were crossed to Texas Marker-1 (TM-1) in 1956. These marker alleles were then backcrossed eight generations to TM-1. During the backcrossing period, the alleles were isolated into individual lines and maintained as heterozygotes. After eight backcross generations, the nine lines were self-fertilized for two to four generations to establish true breeding lines and to provide a seed increase for a performance test.

The recurrent parent for the isoline program (TM-1) had been inbred nine generations when the program began. This line was maintained by individual-plant-pedigree inbreeding during the backcrossing period. TM-1 was selected from 'Deltapine 14,' and it was developed as a genetic standard for *Gossypium hirsutum* L. Kohel, Richmond, and Lewis (1970) described the origin, development, and properties of TM-1.

The performance test, carried out in 1968, included the nine isolines (genotypes) with three entries within each one (lines within genotypes), and two commercial varieties. Three plants from each isoline were chosen at random and increased for the replicated test. The three lines were identified separately to provide an estimate of the variability within each isoline. TM-1 was included in the randomization, but a limited amount of seed prevented its inclusion in the test proper. Seven entries of TM-1, representing seven of the inbred generations used in the development of the isolines, were included to estimate the mean performance of TM-1 for purposes of comparison.

Each entry (line within genotype and TM-1 generation sample) was grown in a single-row plot (1 × 10m) in a test replicated four times and located on the Texas A&M Brazos Valley Farm. The following characters were measured: *(1) Yield* —lint (g) per plot; *(2) Boll size*—seed cotton (g) per boll; *(3) Earliness*—percentage of the total yield harvested at the first picking; *(4) Seed index*—weight (g) of 100 seeds; *(5) Lint index* —weight (g) of lint from 100 seeds; *(6) Lint percentage*—percentage of seed cotton that is lint; *(7)* Fiber *Fineness*—expressed in micronaire units (the units increase with increasing coarseness); *(8) Strength*—breaking strength (g per tex) of a fiber bundle measured with 3.2 mm gage stelometer; *(9) 50% span length*—and *(10) 2.5% span length*—length (in.) at which the given percentage of fibers in an array are that long or longer; and *(11) Elongation*—percentage elongation of the fiber bundle before breakage.

The isolines represented the following markers: *(1)* Red Plant Body *(R_1)*, *(2)* Okra Leaf *(L^o)*, *(3)* Petal Spot *(R_2)*, *(4)* Brown Lint *(Lc_1)*, *(5)* Naked Seed *(N)*, *(6)* Hirsute *(H_2)*, *(7)* Green Lint *(Lg)*, *(8)* Yellow Pollen *(P_1)*, and *(9)* Yellow Petals *(Y_1)*. The markers numbered 1 to 5 originated from one multiple dominant marker line and the remainder from another. Petal Spot and Brown Lint are located in Linkage Group I, Okra Leaf and Green Lint are located in Linkage Group II but are more than 50 units apart; and the remainder of the loci are independently associated (Kohel, Lewis and Richmond, 1965, and Wilson and Kohel, 1970). The commercial varieties in the test (each with three entries) were 'Deltapine 15' and 'Stoneville 213,' designated "D" and "S" respectively.

RESULTS

The analysis of the experimental results revealed that the genotypes differed significantly for each of the 11 characters measured (Table 1). The mean values of the characters measured for each of the isolines and the results of Duncan's test are shown in Table 2. TM-1 is included in this table for comparison. Figure 1 provides a ready comparison of the isolines with TM-1. This figure expresses the values of the isolines as (isoline-TM-1) ($S.E._{\bar{x}}^{-1}$). Only those values greater than $\pm L.S.D._{.05}$ are presented.

Yellow Pollen (P_1), *Petal Spot* (R_2), and *Yellow Petals* (Y_1) isolines deviated only in a minor way from TM-1. These characters have no obvious effects that would be expected to alter plant growth and development.

[1] Contribution of the Crops Research Division, Agricultural Research Service, U.S. Department of Agriculture in cooperation with the Texas Agricultural Experiment Station. Received Sep. 30, 1970.

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Table 1. Mean squares from the analysis of variance of the entries for each of the 11 characters measured.

Source	df	Yield	Boll size	Earliness	Seed index	Lint index	Lint percentage	Fiber properties: Fineness	Strength	50% span length	2.5% span length	Elongation
Genotypes	10	1,571,250**	3.98**	1,910**	9.47**	42.53**	988.59**	9.12**	22.16**	.0272**	.1062**	4.70**
Reps	3	30,247	.31	142	.88	.16	.31	.09	2.08	.0011	.0014	.87
G × R	30	19,248	.07	121**	.49	.21	2.84	.03	.67**	.0003**	.0004**	.24
Lines/(G)	22	18,882	.12**	85*	.50	.19	5.02	.08**	.40*	.0003**	.0013**	.19
L/(G) × R	66	14,842	.05	46	.26	.15	2.22	.02	.20	.0001	.0002	.19

*, ** significance at the .05 and .01 levels of probability, respectively.

Table 2. Mean performance of the entries and TM-1 and the results of Duncan's test.

Line	Yield*	Boll size	Earliness	Seed index	Lint index	Lint percentage	Fiber properties				
							Fineness	Strength	50% span length	2.5% span length	Elongation
R_2	1,000 de	6.02 a	51 bc	13.0 abc	6.54 b	32.9 d	4.93 c	19.04 a	.56 ab	1.15 b	7.94 b
P_1	1,159 c	5.59 bc	53 bc	12.2 def	6.32 b	33.4 d	5.13 b	19.01 a	.54 cd	1.13 c	7.72 bc
Y_1	1,093 cd	5.44 cd	54 bc	12.2 def	6.36 b	34.2 c	4.97 bc	19.04 a	.56 ab	1.14 bc	7.58 bc
R_d	923 e	5.77 b	46 cd	12.7 cd	6.47 b	33.7 cd	5.06 bc	19.04 a	.55 bc	1.09 e	7.60 bc
L	1,096 cd	5.32 de	82 a	11.8 fgh	5.82 c	33.1 d	4.94 c	18.80 ab	.54 cd	1.08 e	7.41 d
H_2	895 e	5.18 e	38 c	13.2 ab	7.76 a	36.3 b	6.08 a	18.14 b	.49 e	.95 f	7.62 bc
N	134 g	4.00 h	51 bc	12.4 cdef	.90 f	6.8 g	4.91 c	18.06 b	.57 a	1.19 a	7.80 bc
Lc_1	601 f	4.50 g	37 c	13.4 a	5.08 d	27.9 e	3.92 e	14.92 d	.42 g	.87 g	9.10 a
Lg	656 f	4.87 f	41 c	12.6 cde	3.78 e	23.2 f	2.58 f	16.01 c	.46 f	1.09 e	8.97 a
D	1,300 b	5.31 e	60 b	10.3 i	6.67 b	39.3 a	4.50 d	18.25 ab	.53 d	1.11 d	8.77 a
S	1,423 a	5.47 cd	55 bc	11.3 h	6.73 b	37.3 b	4.94 c	18.32 ab	.55 bc	1.14 bc	7.51 c
TM-1	1,051	5.74	48	12.8	6.41	33.1	5.09	19.24	.56	1.14	7.52

* Means identified with same letter indicate nonsignificant group.

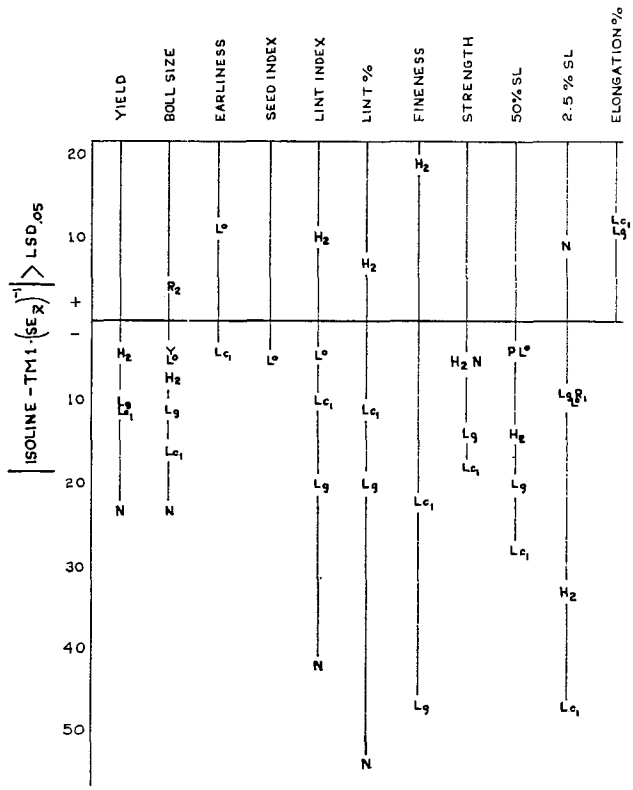

Fig. 1. The performance of the nine isolines for each of the 11 characters measured compared with TM-1.

The *Red Plant Body* (R_1) isoline showed a slight reduction in 2.5% span length and yielded less lint than TM-1. The red pigment in the plant tissue was expected to interfere with the photosynthetic capacity of the plant and therefore, to alter plant growth more than the results indicated. However, the yield-depression effect could assume greater importance at high yield levels.

The *Okra Leaf* ($L°$) isoline was markedly earlier and shorter lint and a lower seed index, lint index, and boll size. Andries et al. (1969) reported the increased earliness of the Okra Leaf genotype in varietal backgrounds.

Hirsute (H_2) causes dense hairs over the plant body and influences the development of the seed hairs (fibers). Hirsute plants had short, coarse fibers. Lint index and lint percentage increased, but boll size and lint yield decreased in the Hirsute isoline. Since seed size (seed index) was not reduced, the reduced boll size and yield must have resulted from a reduction in number of seeds per boll. Simpson (1947), Lee (1964), and Kohel, Lewis and Richmond (1967) have reported similar effects of the H_2 gene on fiber properties.

The *Naked Seeded* (N) genotype results in the absence of seed fuzz, those seed hairs that do not enlongate to form lint. A reduction in the number of fibers produced was associated with the absence of seed fuzz. This reduction was detectable in the large decreases in lint index, lint percentage, and, of course, boll size and lint yield. The energy not expended by producing fewer fibers was utilized in part by producing longer fibers. Low (1968) has reported that number of fibers per seed (as reflected in lint index and lint percentage) can be increased even when the NN genes are present. Our experience was the opposite of Low's, that is, the NN genotype in the isoline had a lower lint percentage and lint index than the original NN source.

Both the *Green Lint* (Lg) and *Brown Lint* (Lc_1) isolines had similar characteristics. The fibers in these mutant types were short, fine, and weak. The total production of cellulose was reduced, which is reflected in a lower lint index, lint percentage, boll size, and yield. The nature of the Green Lint pigment and its influence on fiber development have been studied in detail (Conrad, 1941; Conrad and Neely, 1943; Neely, 1943; and Kohel et al., 1967).

These markers affect the characters studied to varying degrees. With the exception of Hirsute and Green Lint and perhaps Naked Seed, they are found as natural variants in wild *G. hirsutum* populations. Hirsute is present in all known accessions of the wild tetraploid, *G. tomentosum* Nutt. ex Seem.

In addition to the characterization of the isolines, we were interested in the variability that remained within each genotype following eight generations of backcrossing. The comparisons among genotypes and with TM-1 provided the opportunity to measure the pleiotropic or tight linkage effects of the marker loci. The comparison of variability among lines within each genotype provides some measure of the linkages associated with the marker loci. Hanson (1959) calculated that the intact half-segment associated with the locus would be .125 centomorgans, after eight generations of backcrossing in which the locus in question was held heterozygous, as was done in this study.

In the present study, we found that lines within genotypes were significantly variable for boll size, earliness, fiber fineness, strength, and fiber length (Table 1). The variability within each genotype was further analyzed to determine which genotypes contributed significantly to the variability.

Lines within three genotypes were associated with differences in boll size. Yellow Petals and Green Lint each had one line that had a larger boll size, and Naked Seed had one line that was consistently smaller. Green Lint was the only genotype in which the lines varied for earliness, and one line was consistently earlier. Brown Lint lines varied for fiber fineness, but none of the other lines within genotypes varied significantly for fineness. Hirsute and Green Lint lines varied significantly for fiber strength. One Hirsute line had consistently stronger fiber; the Green Lint lines were arrayed for strength. All of the genotypes, except Naked Seed, showed significant differences among lines for either 2.5% or 50% span length. This result would follow from the multiplicity of factors that control fiber length. Genetic analysis of fiber length indicates that it is controlled by many factors with additive gene effects (Lee, Miller and Rawlings, 1967; Al-Rawi and Kohel, 1970).

The significant variability of lines within genotype emphasizes the inherent difficulties involved in attempts to develop a true set of isolines. However, if the observed variability does represent linkages, as it should, it does provide some guidelines in the use of genetic markers for the mapping of agronomic characters. The indicated associations reflect the chromosome location of some loci controlling agronomic traits. They do so to the extent that variability existed in the original material used in the backcrossing. These marker loci came from two multiple marker lines. It is not possible to characterize such multiple marker lines agronomically, but the separate origin of these lines makes it probable that they are different from TM-1.

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