The *Journal of Cotton Science* 6:115-118 (2002)
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NOTE

Inheritance and Linkage Analysis of the Yellow Pulvinus Mutant of Cotton

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INTERPRETIVE SUMMARY

Morphological mutants of cotton are used in studies of plant development and genetic mapping, and occasionally have proven to have agronomic value. We discovered a spontaneous mutant in a cotton breeding nursery, which is the source of more than half the mutants in cotton. Inheritance studies established that the mutant was controlled by recessive alleles at a single locus. Linkage analysis showed weak linkage with cluster-1, which would place the mutant in linkage group III on chromosome 16. No linkage was associated with the other 12 markers tested, including the other marker located distally on chromosome 16, which should place the mutant near the centromere region of chromosome 16. This mutant adds an additional morphological mutant for genetics research on cotton.

ABSTRACT

Morphological mutants of cotton are used in genetic mapping studies and in studies of plant development, and occasionally have proven to have agronomic value. The objectives of this study were to determine the inheritance and linkage relations of a spontaneous mutant found in a cotton breeding nursery. Seeds of the mutant plant were grown and crossed with the genetic standard, TM-1, and multiple marker tester lines, T582 and T586, to determine the inheritance and linkage relations. Inheritance studies established that the mutant was controlled by recessive alleles at a single locus. Linkage analysis showed weak linkage with cluster-1, which would place the mutant in linkage group III on chromosome 16. No linkage associations were found with the 12 markers tested, including the other marker located distally on chromosome 16, which should place the mutant near the centromere region of chromosome 16. These results have identified a new recessive mutant located on chromosome 16, which we designate yellow pulvinus (*yp*). This mutant adds an additional morphological mutant for genetics research on cotton.

Our program to identify and maintain morphological mutants in cotton (*Gossypium* spp.) seeks mutants as genetic markers and as aberrant forms to study basic plant processes and development. With few exceptions, the most useful mutants have been those found as spontaneous mutants in breeding populations. In general, they are more vigorous and easier to maintain and to manipulate in experimental conditions than induced mutants. Of the 146 mutants in cotton, only 2 were recovered from irradiation; the remaining are natural variants, introduced by introgression, or were found as variants (77) in breeding nurseries or production fields (Percy and Kohel, 1999). The ability of cotton to tolerate duplications and deficiencies complicates the recovery of mutants from high-energy radiation treatments (R.J. Kohel, unpublished data, 1960-1980).

A mutant was found in L. S. Bird's upland cotton breeding nursery in 1973. Open-pollinated seed was harvested, but the recovered mutant plants were unproductive because the mutants failed to survive long enough to recover seed. In 1977, plants with the same mutant phenotype were observed again in his breeding nursery. Based on previous experience, we took greater care and precautions in its growth and culture. Self-pollinated seeds were produced, and crosses were made to initiate the inheritance study.

MATERIALS AND METHODS

The mutant phenotype has general depauperate growth with reduced glands on the stem, no or reduced normal reddening of the pulvinus area, a

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Fig. 1. Young yellow pulvinus plant exhibiting the general depauperate growth with reduced normal reddening of the pulvinus area, a gathering of veins at the base of the leaf, and a distinctive yellowing of the pulvinus area.

gathering of veins at the base of the leaf and bract, and a distinctive yellowing of the pulvinus area (Fig. 1). The mutant phenotype is not expressed in the seedling stage and begins in the pre-flowering growth period. E. L. Turcotte indicated that he had a mutant with a similar phenotype in pima cotton, *Gossypium barbadense* L. He conducted allelic tests that showed that it was genetically independent (Turcotte and Feaster, 1983). When both mutants were grown at College Station, TX, they were visually distinct. The mutant from *Gossypium hirsutum* L. has a more extreme mutant phenotype, and plant growth is more debilitating than *golden veins* mutant of *G. barbadense*.

The genetic standard, Texas Marker-1 (TM-1) (Kohel et al., 1970), was used in crosses with the mutant for inheritance studies. For the analysis of genetic linkage relations, the mutant line was crossed with T582, multiple recessive marker line, and T586, multiple dominant marker line. T582 includes the following recessive marker loci: virescent-1, v_1, linkage group XVII on chromosome 20; cup leaf, *cu*, location unknown; glandless-1, gl_1, location unknown; frego bract, *fg*, linkage group VI on chromosome 3; and cluster-1, cl_1, linkage group III on chromosome 16. T586 includes the following dominant marker loci: red plant, R_1, linkage group III on chromosome 16; okra leaf, L_2^o, linkage group II on chromosome 15; tomentum, T_1, linkage group IV on chromosome 6; petal spot, R_2, linkage group I on chromosome 7; yellow pollen, P_1, linkage group XI on chromosome 5; yellow petals, Y_1, linkage group XII on an unknown A chromosome; brown lint, Lc_1, linkage group I on chromosome 7; green lint, *Lg*, linkage group II on chromosome 15; and naked seed, N_1, linkage group V on chromosome 12 (Percy and Kohel, 1999).

Seeds of each population were germinated in the greenhouse in peat pellets following the normal procedures of the genetics program at College Station, TX. At age 3 wk, the greenhouse-grown seedlings were transplanted to field plots. Plots consisted of rows with 0.5-m spacing between plants within rows and 1.0-m spacing between rows. This procedure requires fewer seeds, which is important when studying mutants of low productivity. It further allows for stress-free and competition-free observation among segregating individuals. Inheritance and linkage tests were conducted over several years between 1986 and 1996.

RESULTS AND DISCUSSION

Crosses of the mutant with Texas Marker-1 produced normal F_1 progeny, which indicated that the mutant was recessive. Subsequent segregating generations followed the pattern of segregation of a single recessive allele. Segregation of F_2's from reciprocal crosses of TM-1 with the mutant and T582 with the mutant did not deviate significantly from a 3:1 ratio of normal to mutant segregation (Table 1). Heterogeneity among these populations was not significant. Progeny from backcrosses of reciprocal F_1 populations to the mutant did not deviate significantly from an expected 1:1 segregation (Table 1). Heterogeneity was not significant among the three backcross populations. The uniformity within the F_2 and backcross populations of reciprocal crosses showed that there were no maternal effects.

Table 1. Segregation and inheritance analysis of the yellow pulvinus cotton mutant in F_2 and backcross populations.

Population	Total	Normal	Mutant	df	$\chi^2_{(3:1)}$
		no.†			
(TM-1 × mutant)F_2	98	76	22	1	0.34
(mutant × TM-1)F_2	97	75	22	1	0.28
(TM-1 × mutant)F_2	130	99	31	1	0.09
(T582 × mutant)F_2	56	43	13	1	0.10
(T582 × mutant)F_2	90	64	26	1	0.73
Sum	471	357	114	6	1.53
Pooled				1	0.16
Heterogeneity				5	1.37
					$\chi^2_{(1:1)}$
(mutant × TM-1) × mutant	96	44	52	1	0.67
(TM-1 × mutant) × mutant	99	44	55	1	1.22
(TM-1 × mutant) × mutant	96	52	44	1	0.67
Sum	291	140	151	3	2.56
Pooled				1	0.42
Heterogeneity				2	2.14

† Total, normal, and mutant number of plants observed in each population.

Results of the linkage analysis of the five markers of T582 with the mutant are shown in Table 2. The marker loci *fg* and cl_1 showed significant linkage deviations. The mutant-marker combinations were in repulsion and *fg* showed significant excessive recombination (RC = 68%) with the mutant because the mutant phenotype showed over expression of the *fg* phenotype. Heterozygous *fg* has a slight twisting of the bract, and on the mutant background, some heterozygotes were not distinguishable from the homozygotes. Linkage of cl_1 and the mutant was not significant in the first F_2, but it was significant in the second F_2 population. Averaged over the two F_2's, the linkage was significant and heterogeneity was not significant. The linkage was weak, 29 cM, in the F_2 of 146 plants. In the backcross population with the nine dominant markers of T586, no linkage combinations were significant (Table 3). Since cl_1 and R_1 are in Linkage group III on chromosome 16 with R_1 17 map units distal to cl_1, the lack of linkage between them means that the mutant is located proximally near the centromere (Percy and Kohel, 1999).

Table 3. Linkage analysis of backcross segregation of the mutant with dominant markers of T586. The population size, chi-squared for linkage deviations, and recombination values are given.

Marker	n†	χ^2_L‡	RC%§
R_1	89	0.28	47
L_2^o	89	1.36	56
T_1	89	0.55	54
R_2	89	0.55	54
P_1	89	0.91	55
Y_1	89	0.01	49
Lc_1	89	1.90	43
N_1	89	0.10	48
Lg	89	1.36	44

† Population size.
‡ Chi-squared for linkage deviations.
§ Recombination values.

The most distinctive feature of this mutant is the yellow coloration of the pulvinus region, so we propose the name yellow pulvinus with the gene symbol, *yp*, which follows the established convention (Kohel, 1973).

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Table 2. Linkage analysis of F_2 segregation of the yellow pulvinus cotton mutant with recessive markers of T582 in two test populations.

	Population 1			Population 2			Total				
Marker	n†	χ^2_L‡	RC%§	n	χ^2_L	RC%	n	χ^2_L	RC%	χ^2_T	χ^2_H
fg	56	18.29**	84	90	3.6	58	146	17.12**	68	21.89	4.77
cl_1	56	1.14	38	90	11.86**	22	146	11.33**	29	13.00	1.67
v_1	56	0.79	39	90	0.00	49	146	0.37	46	0.79	0.42
cu	56	0.51	43	90	1.43	57	146	0.25	52	1.94	1.69
gl_1	56	0.51	41	90	0.00	49	146	0.025	47	0.51	0.26

**** Chi-squared, $p > 0.01$.**
† Population size.
‡ Chi-squared for linkage deviations.
§ Recombination values.

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