

# New Virescent Cotton Mutant Linked with the Marker Gene Yellow Petals<sup>1</sup>

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## ABSTRACT

A new virescent mutant in cotton (*Gossypium hirsutum* L.), which occurred spontaneously, was found in the Cotton Breeding Nursery at the Texas Agric. Exp. Stn., College Station, Texas. Inheritance tests indicate that the phenotypic expression of this mutant is controlled by a recessive gene at a single locus that, when homozygous, results in virescent foliage. The degree of yellowness expressed by this mutant during its period of maximum expression is comparable to the expression of virescent-1 (*v*<sub>1</sub>) or yellow green (*yg*, *yg*<sub>2</sub>) at College Station. Linkage tests indicate that this new virescent mutant is linked about five crossover units from Yellow petals (*Y*<sub>1</sub>), comprising a new linkage group in cotton, and it is independent from 16 other marker mutants tested. It is proposed that this new virescent mutant be named virescent-10 and assigned gene symbol *v*<sub>10</sub>, and the new linkage group be designated Linkage Group XII.

**Additional index words:** *Gossypium hirsutum* L.

A cotton (*Gossypium hirsutum* L.) seedling with yellow foliage was observed in the Cotton Breeding Nursery at the Texas Agric. Exp. Stn., College Station, Texas, in 1969. This aberrant plant appeared spontaneously in an otherwise normal row. The mutant seedling was transplanted to a greenhouse, and as the plant developed, its leaves became normal green in color. This behavior typifies virescent yellow mutant plants, and the aberrant plant was provisionally designated V5-69. Self-pollinated seeds were obtained from this virescent yellow plant, and progeny tests proved that the virescent characteristic was heritable.

## MATERIALS AND METHODS

Plants of V5-69 and their subsequent progeny were crossed with appropriate test lines to furnish seeds needed for F<sub>1</sub>, F<sub>2</sub>, and backcross populations to test for allelism, inheritance, and linkage. Texas Marker-1 (TM-1) (Kohel et al., 1970) was used in this study as the normal green stock. The inbred line (TM-1) was developed from the cultivar 'Deltapine 14' to serve as a standard tester. Allelism tests were conducted to determine the relationship between the V5-69 mutant and the eight previously identified virescent mutants of *G. hirsutum* L. The test consisted of classifying the F<sub>1</sub> progeny of the crosses, V5-69 × identified virescent mutants, V5-69 × V5-69 (virescent check), and V5-69 × TM-1 (green check) for virescent and green leaf color expression. Virescent-7 (*v*<sub>7</sub>) from *G. barbadense* L., the homoeolog of virescent-1 (*v*<sub>1</sub>) in *G. hirsutum* L. (Turcotte and Feaster, 1973), was not included in the test.

Inheritance tests consisted of classifying progeny of the [(V5-69 × TM-1) × V5-69] backcross and (V5-69 × TM-1)F<sub>2</sub> populations for the virescent and green leaf color character expression. In addition, backcross and F<sub>2</sub> segregating populations of the linkage tests were included in the inheritance tests.

The V5-69 mutant was tested for linkage with 17 known monogenic markers; 12 of these markers are included in the multiple marker lines Texas 586 (T586) and a selection of Texas 582 (T582), which were synthesized for this purpose (Kohel, 1972). T586 contains dominant genes at eight loci representing

five linkage groups: Linkage Group I—Brown lint (*LC*<sub>1</sub>) and Petal spot (*R*<sub>2</sub>); Linkage Group II—Okra leaf (*L*<sup>o</sup>); Linkage Group III—Red plant (*R*<sub>1</sub>); Linkage Group IV—Pilose (*H*<sub>2</sub>); Linkage Group XI—Pollen color (*P*<sub>1</sub>); and Independent—Yellow petals (*Y*<sub>1</sub>) and Naked seed (*N*<sub>1</sub>). The T582 selection contains four recessive genes and represents two linkage groups: Linkage Group III—cluster-1 (*cl*<sub>1</sub>); Linkage Group VI—frégo bract (*fg*); and Independent—glandless boll (*gl*<sub>1</sub>) and cup leaf (*cu*). Linkage Group V—glandless plant-2 (*gl*<sub>2</sub>) and Linkage Group IX—glandless plant-3 (*gl*<sub>3</sub>) (McMichael, 1960) were tested together, because they are a recessive duplicate pair for gossypol gland density on cotyledons (segregation ratio 13:3). The remaining two dominant and one recessive marker loci were tested individually: Linkage Group VII—Laciniate Leaf (*L*<sup>l</sup>) (Hutchinson, 1934); Linkage Group VIII—mosaic leaf (*ml*) (Lewis, 1958); and Linkage Group X—Ragged leaf (*Rg*) (Kohel and Lewis, 1962). Backcross populations were primarily used to test dominant loci and F<sub>2</sub> populations were used to test recessive loci. These data were analyzed for linkage by the methods outlined by Mather (1951).

Plant populations were grown from seeds germinated individually in peat pellets in a greenhouse. The seedlings were transplanted within 3 weeks, either to the field or to individual pots that remained in the greenhouse. Field rows were 1 m wide, and plants were spaced 46 cm apart within the row. Seedlings were classified for virescent and mutant marker expression in the greenhouse and in the field throughout the growing season.

The mutant V5-69 has seedlings with cotyledons that vary from pale green to normal green. The first true leaf, and subsequent leaves through the 6 to 8-week stage of development, has a yellow phenotype. After that stage of development the leaves begin to turn green. The greening process is accelerated, with the new leaves progressively less yellow. Newly expanded leaves are green, and the presence of any yellowness is difficult to detect after 10 weeks. The degree of yellowness during the period of maximum expression is comparable to the expression of virescent-1 (*v*<sub>1</sub>) and yellow green (*yg*, *yg*<sub>2</sub>) at College Station. The size and conformation of the V5-69 mutant plants are not appreciably different from those of normal green plants.

## RESULTS AND CONCLUSIONS

The results of the allelism tests show that V5-69 is not an allele of any of the six monogenic or two digenic virescent mutants of *G. hirsutum* (Table 1)

Table 1. Allelism tests of the V5-69 mutant with eight identified virescent mutants, classification of F<sub>1</sub>'s with V5-69 and parental lines in cotton.

Virescent mutant and TM-1	Classification				Reference
	F <sub>1</sub> with V5-69		Parental line		
	Green	Yellow	Green	Yellow	
	no. plants				
<i>v</i> <sub>1</sub>	8	--	--	4	Killough and Horlacher, 1933
<i>v</i> <sub>2</sub>	10	--	--	5	Duncan and Pate, 1967
<i>v</i> <sub>3</sub>	10	--	--	5	Percival and Kohel, 1974
<i>v</i> <sub>4</sub>	10	--	--	5	Quisenberry and Kohel, 1970
<i>v</i> <sub>5</sub> <i>v</i> <sub>6</sub> †	9	--	--	5	Kohel, 1973
<i>v</i> <sub>8</sub>	10	--	--	5	Kohel, 1974
<i>v</i> <sub>9</sub>	10	--	--	5	Percival, 1974
<i>yg</i> <sub>1</sub> <i>yg</i> <sub>2</sub> †	10	--	--	4	Rhyne, 1955
V5-69	--	10	--	10	
TM-1	10	--	10	--	

† Digenic virescent.

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**Table 2.** Classification of backcross and  $F_2$  population segregation of the virescent mutant V5-69 of cotton and chi-square analysis.

Population	Classification		Chi-square analysis	
	Green	Yellow	$\chi^2$ (ratio tested)	P
	no. plants			
Backcross generation			1:1	
[(V5-69 $\times$ TM-1) $\times$ (V5-69)]BC <sub>1</sub>	50	55	0.24	0.50-0.70
[(V5-69 $\times$ T586) $\times$ (V5-69)]BC <sub>1</sub>	70	67	0.07	0.70-0.80
[(V5-69 $\times$ LG III) $\times$ (V5-69)]BC <sub>1</sub>	44	48	0.17	0.50-0.70
[(V5-69 $\times$ LG VII) $\times$ (V5-69)]BC <sub>1</sub>	44	53	0.84	0.30-0.50
[(V5-69 $\times$ LG X) $\times$ (V5-69)]BC <sub>1</sub>	34	39	0.34	0.50-0.70
Total	242	262	0.79	0.30-0.50
Heterogeneity			0.86	0.90-0.95
$F_2$ generation			3:1	
(V5-69 $\times$ TM-1) $F_2$	75	23	0.23	0.70-0.80
(V5-69 $\times$ T582) $F_2$	50	21	0.79	0.30-0.50
(V5-69 $\times$ T582) $F_2$	31	6	1.52	0.20-0.30
(V5-69 $\times$ LG V) $F_2$	62	24	0.39	0.50-0.70
(V5-69 $\times$ LG VI) $F_2$	57	19	0.00	1.00
(V5-69 $\times$ LG VIII) $F_2$	73	20	0.61	0.30-0.50
Total	348	113	0.06	0.80-0.90
Heterogeneity			3.36	0.30-0.50

**Table 3.** Chi-square linkage deviations and recombination percentages of V5-69 with mutant loci in cotton.

Marker locus	No. plants	Linkage $\chi^2$	P	Ratio tested	Recombination %
[(V5-69 $\times$ T586) $\times$ V5-69]BC <sub>1</sub>					
$Y_1$	90	60.84	<0.01	1:1/1:1	8.89
$Y_1$	132	120.17	<0.01		2.27
$R_1$	137	0.18	0.50-0.70		51.82
$H_2$	136	1.06	0.30-0.50		45.59
$L^0$	137	0.18	0.50-0.70		48.18
$R_2$	132	3.67	0.05-0.10		58.33
$P_1$	132	3.03	0.05-0.10		42.42
$Lc_1$	134	0.12	0.70-0.80		48.51
$N_1$	134	0.75	0.30-0.50		46.27
(V5-69 $\times$ T586) $F_2$					
$Y_1$	79	45.06	<0.01	3:1/3:1	6.84
$R_1$	79	0.32	0.50-0.70		54.38
$H_2$	79	0.00	0.90-0.95		50.14
$L^0$	79	3.38	0.05-0.10		67.90
$R_2$	79	1.53	0.20-0.30		61.13
$P_1$	79	1.54	0.20-0.30		52.29
$Lc_1$	79	1.54	0.20-0.30		56.47
$N_1$	79	1.18	0.20-0.30		60.12
(V5-69 $\times$ T582) $F_2$					
$cl_1$	102	1.34	0.20-0.30	3:1/3:1	40.78
$fg$	103	1.17	0.20-0.30		43.04
$gl_1$	108	3.70	0.05-0.10		35.39
$cu$	108	2.42	0.10-0.20		37.46
[(V5-69 $\times$ LG III) $\times$ (V5-69)]BC <sub>1</sub>					
$R_1$	92	0.54	0.30-0.50	1:1/1:1	46.15
(V5-69 $\times$ LG V) $F_2$					
$gl_2$	86	1.00	0.10-0.20	3:1/13:3	57.90
[(V5-69 $\times$ LG VII) $\times$ (V5-69)]BC <sub>1</sub>					
$L^L$	97	0.26	0.50-0.70	1:1/1:1	52.58
(V5-69 $\times$ LG VIII) $F_2$					
$ml$	93	0.14	0.70-0.80	3:1/3:1	46.21
(V5-69 $\times$ LG IX) $F_2$					
$gl_3$	86	2.21	0.10-0.20	3:1/13:3	58.83
[(V5-69 $\times$ LG X) $\times$ (V5-69)]BC <sub>1</sub>					
$Rg$	71	0.69	0.30-0.50	1:1/1:1	45.07

because all of the  $F_1$  progeny were green. Self-pollinated seeds of the eight virescents used as parents and V5-69 all produced seedlings that expressed a virescent yellow leaf color. These results established V5-69 as a new virescent mutant.

The inheritance tests indicated that the phenotypic expression of V5-69 is controlled by a recessive gene

**Table 4.** Segregation and estimation of the recombination value of the coupling linkage of V5-69 [ $v$  with Yellow petals ( $Y_1$ )] and heterogeneity chi-square.

Data set	Type of population and expected segregation	$V Y_1$	$V y_1$	$v Y_1$	$v y_1$	Recombination %
1	BC <sub>1</sub> (1:1:1:1) (1973)	65	3	0	64	2.27 $\pm$ 1.4
2	BC <sub>1</sub> (1:1:1:1) (1974)	35	7	1	47	8.89 $\pm$ 3.0
3	$F_2$ (9:3:3:1) (1973)	59	2	3	15	6.84 $\pm$ 2.8
Combined populations†						5.00 $\pm$ 1.27

† Heterogeneity (2 df)  $\chi^2 = 5.50$ ,  $P = 0.05-0.10$ .

at a single locus that, when homozygous, results in virescent foliage. This conclusion was made from homogeneous data of five backcross (to the mutant parent) and six  $F_2$  segregating populations totaling 965 plants (Table 2). Backcross (to the mutant parent) and  $F_2$  segregating populations did not deviate significantly from a green:yellow 1:1 and 3:1 ratios, respectively.

The results of linkage tests with known mutants indicated that the V5-69 mutant is linked with the marker, Yellow petals ( $Y_1$ ), a previously independent and unassociated mutant. It was not associated with any of the 16 other markers tested. Recombination between Yellow petals ( $Y_1$ ) and V5-69 in the two backcross populations [(V5-69  $\times$  T586)  $\times$  V5-69] was 8.89 and 2.27%, and the recombination estimated in the (V5-69  $\times$  T586)  $F_2$  population was 6.84% (Table 3). A combined estimate of the recombination value using Fisher's scoring method, as outlined by Allard (1956), yielded a value of  $5.00 \pm 1.27\%$  (Table 4). The linkage of V5-69 with Yellow petals ( $Y_1$ ) establishes a new linkage group in cotton.

It is proposed that this new virescent mutant be named virescent-10 and assigned gene symbol  $v_{10}$ . Since there are 11 previously reported linkage groups, each independent of  $v_{10}$ , the new linkage between Yellow petals ( $Y_1$ ) and virescent-10 ( $v_{10}$ ) is designated Linkage Group XII.

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