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Linkage Tests in Upland Cotton, *Gossypium hirsutum* L. II.[1]

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ABSTRACT

Results of linkage analyses of cotton performed at College Station, Texas are presented, and linkage information reported by other workers is summarized. The mutants withering bract, accessory involucre, abnormal palisades, Ragged Leaf, round leaf, Red Dwarf, male-sterile-1, male-sterile-2, Male-sterile-4, and Ligon Lintless were tested for linkage with several marker loci and in various combinations among themselves.

Ragged Leaf and round leaf were found to be linked (R.C. = 32%), and thus form a new linkage group, number X. Data are presented to document the simple dominant inheritance of Ligon Lintless; the symbol *Li* is assigned to the mutant allele.

Additional index words: **Linkage Group X, Ligon Lintless inheritance.**

THIS paper reviews the efforts by researchers at several locations to identify the 26 potential linkage groups of *Gossypium hirsutum* L., and it reports the progress made since the last published linkage report (34).

In the past, researchers at the Texas Station have relied heavily upon our multiple marker lines to expedite tests for linkage associations. These lines are still useful for testing new mutants, although most known mutants have been tested with the multiple marker stocks. Often, as in this report, the only remaining untested combinations in many of our linkage tests involve digenic combinations.

MATERIALS

Our most useful multiple marker stocks are T 582 (*cu, fg, cl_1, gl_1,* and *v_1*), T 586 (*R_2, Lc_1, L^o, R_1, H_2, Y_1, N,* and *P_a*), and T 588 (*R_2, L^o, Lg, R_1, H_2, Y_1, N,* and *P_a*). Summary information concerning the mutant alleles in the multiple marker lines and the other lines involved in the linkage analyses is presented in Table 1. In this table and also in the text, the conventional method of numbering the chromosomes of *G. hirsutum* is followed in which numbers 1 to 13 are assigned to the A subgenome chromosomes and numbers 14 to 26 are assigned to the D subgenome chromosomes.

The mutants in the linkage tests with which 1 was primarily concerned were withering bract, accessory involucre, abnormal palisades, Ragged Leaf, round leaf, Red Dwarf, male-sterile-1, male-sterile-2, Male-sterile-4, and Ligon Lintless.

Withering bract, initially called deciduous bract, was discovered in 'Stoneville 2B.' The inheritance of this mutant was determined by Knight (35) and it was found to be linked to *gl_2* by Rhyne (46). Subsequent studies of withering bract revealed that it was controlled by duplicate loci (47). All *G. hirsutum* lines tested were recessive at the second locus (*bw_2*), which accounts for the apparent monogenic control of this mutant. The *bw_1* locus is located on a chromosome of the A subgenome, and *bw_2* is located on the homeologous D chromosome.

Accessory involucre is one of a series of mutants (Heritable Abnormality #7) found by H. C. McNamara. It is simply inherited and linked with frego-bract (34). Frego-bract is thought to reside on an A subgenome chromosome.

Abnormal palisades is controlled by duplicate recessive genes (31). The amphidiploid nature of *G. hirsutum* and the experiences of cotton geneticists with other duplicate genes suggest that the controlling loci reside on homeologous chromosomes. Convention places the lower numbered locus (*lp_1*) in the A subgenome and the other (*lp_2*) in the D subgenome.

Ragged Leaf is a simply inherited dominant in which the homozygote is usually lethal (33). Round leaf (3, 51) and crenate (58) are merely two names for the same condition.

Red Dwarf (*Rd*) has a phenotype similar to Red Plant (*R_1*) except that, as its name implies, plants with mutant alleles at the Red Dwarf locus have short stature associated with their red pigmentation (25, 40).

Male-sterile-1 and -2 are simply inherited recessives, whereas Male-sterile-4 is a simply inherited dominant. The chromosomal or subgenomic location of these genes is not known. All three male-steriles differ in their expression. Male-sterile-1 is conditionally male-sterile in that the mutant expression (non-dehiscent anthers) is influenced by environmental conditions. At some locations, it can be identified with a high degree of accuracy, but at College Station the degree of expression varies from year to year. Both male-sterile-2 and Male-sterile-4 impart

Table 1. Name, symbol, and chromosome or subgenome location of mutants involved in linkage tests (references in parentheses).

Name	Symbol	Chromosome or subgenome
abnormal palisades	lp_1, lp_2 (31)	1, 15 D (10)
accessory involucre	*Ia* (32)	
cluster fruiting	cl_1 (24, 59)	16 (8, 12)
cup leaf	*cu* (38)	A (13)
frego bract	*fg* (14)	
glandless boll	gl_1 (41)	D (12, 48)
male-sterile-1	ms_1 (26)	
male-sterile-2	ms_2 (52)	
round leaf	*rl* (3, 51)	
virescent-1	v_1 (24, 30)	A (12, 13)
withering bract	bw_1, bw_2 (35)	A, D (47)
Brown Lint	Lc_1 (22, 24, 50, 61)	7 (11, 12, 27)
Green Lint	Lg^+(19, 24, 50)	15 (8)
Laciniate Leaf	L^L	1 (62)
Ligon Lintless	*Li* (4, text)	D (42)
Male-sterile-4	Ms_4 (1)	
Naked Seeds	*N* (5, 15, 19, 24, 29, 59, 60)	A (12)
Okra Leaf	L^o(20, 24, 53)	15 (8, 12)
Petal Spot	R_2 (17, 19, 20, 24)	7 (11, 12)
Pilose	H_2 (55)	6 (6, 12)
Ragged Leaf	*Rg* (33)	
Red Dwarf	*Rd* (25, 40)	D (12, 42)
Red Plant	R_1 (20, 24, 30, 39, 59)	16 (12)
Yellow Petals	Y_1 (16, 24)	A (57)
Yellow Pollen	P_a (18, 24)	A (56)

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complete male-sterility; however, male-sterile-2 results in normal-appearing, but indehiscent anthers, that contain aberrant pollen, whereas Male-sterile-4 varies in its expression from indehiscent anthers to the formation of only rudimentary filaments and anthers.

Ligon Lintless is a mutant that has been known since the 1930's (4) and it has been used extensively as a genetic marker at the Texas Station and elsewhere. It is known to be inherited as a simple complete dominant; however, its inheritance has never been documented in print. This paper will present data to document the inheritance and to assign gene symbols to Ligon Lintless in addition to reporting its performance in linkage tests.

RESULTS

Results of the linkage tests are presented in Table 2. The table contains the recombination values and the population sizes in the various test combinations. The dashes indicate previous tests with independent associations. The associations of *Rg* with *rl* and *fg* with *ia* were the only significant linkage associations detected among the test combinations.

The linkage between *rl* and *Rg* represents a new linkage group, number X, in *G. hirsutum*. Data from the initial test populations indicated that *Rd*, a D genome marker, was linked to *Rg*. However, information from a larger population failed to indicate linkage.

Kohel, Lewis and Richmond (34) identified linkage group VI (*ia-fg*). My data from additional populations of this combination provided a slightly lower estimate of the recombination value than the original one.

Although geneticists agree that Ligon Lintless is a simply inherited dominant mutant, this fact has not been documented, and a gene symbol has not been assigned. Table 3 summarizes the segregation data from those linkage populations reported in this paper and a previous one (34), and the symbol *Li* is assigned to the mutant allele.

Results verified that *Li* is inherited as a simple dominant and also showed that Ligon Lintless plants are less vigorous than normal reflected by the deficient number of plants in the mutant class. The variation in goodness-of-fit to the single gene model was related in part to whether the plants were classified in the greenhouse soon after germination or in the field after they had been transplanted.

Ligon Lintless mutants are characterized by contorted leaf laminae and a twisted appearance of the branches and stems. This expression is first evident in the cotyledonary seedling stage. Another feature characteristic of this mutant is its short lint (approximately 0.2 cm long compared with lint approximately 2.5 cm long in normal cotton). In addition, the homozygous mutant plants appear slightly smaller than the heterozygous ones.

DISCUSSION AND REVIEW

The results of linkage analyses in *G. hirsutum* were summarized in a recent report (2). However, subsequent papers have provided new information; and this information has been incorporated in the current linkage map of *G. hirsutum* presented in Table 4.

Endrizzi and Taylor (11) and Poisson (43) isolated the monosome for chromosome 7, and found that it is associated with linkage group I. As a result of these

Table 2. Summary of linkage tests (the upper number is the recombination percent, and the lower number is the number of plants classified in the test combination).

	Mutants tested									
Testers	bw_1	ia	lp_1 lp_2	Rg	rl	Rd	ms_1	ms_2	Ms_4	Li
R_2	53 100	-	50 176	-	-	-	54 93	-	67 98	
Lc_1				-	-		48 94	-	42 99	
L^a	47 102	-	56 176	-	-	-	53 95	-	50 98	
lg	43 99	-	57 176	-	-	-	55 95		56 99	
cl_1	45 192	53 178		-	45 193	56 112		-	44 64	
R_1	51 102	-	49 176	-	-		58 95	-	55 98	
H_2	53 102	-	47 176	-	-	-	50 95	-	58 98	
fg	58 192	29, 89 572		-	55 196	48 230		-	44 64	
L^L				43 106	44 98	52 103		47 97		
rl			32, 45 607			55 433		45 88		58 112
Rg						54 433		54 93		56 77
Rd								51 95		48 92
Y_1	44 100	-	48 176	-	-	-		-	46 98	
N	50 99		53 176						60 99	
v_1	58 193	52 192	50 186	-	43 197	52 112		-	58 64	
cu	57 195	43 192	63 186	-	60 197	55 112		-	53 64	
gl_1	52 194	58 192	57 186	-	53 197	52 112		-	50 64	
P_a	54 100	-		-	-	-	55 92	-	54 50	
Li				56 77	58 112	48 92		52 98		

Table 3. Segregation of Ligon Lintless in backcross and F_2 populations.

	Number of plants			
Population	Ligon Lintless (Li-)	Normal (lili)	Chi-square value	P
Backcross (1:1)				
lili(lili·LiLi)-1	31	45	2.19	.2-.1
-2	45	47	0.10	.5-.7
-3	45	53	0.65	.5-.3
(LiLi·lili)lili-1	75	112	7.32	<.01
-2	48	64	2.28	.2-.1
(lili·LiLi)lili	88	105	1.50	.3-.2
Pooled	332	426	11.66	<.01
Heterogeneity			2.38	.8-.7
F_2 (3:1)				
(lili·LiLi)⊗ -1	133	60	3.82	.1-.05
-2	127	44	0.05	.9-.8
-3	219	86	1.66	.2-.1
Pooled	479	150	.44	.7-.5
Heterogeneity			5.09	.05-.02

Table 4. Linkage map of *Gossypium hirsutum*.

Linkage group	Mutant loci and recombination values	Chromosome or genome location	Reference
I	R_2-16-cl_2-4-yg_2-32-Lc_1	7	11, 21, 27, 43, 44, 45, 54, 58
II	lp_2-?-L^a-32-c*-44-Lg-7-cv	15	9, 10, 58, 63
III	Dw-29-yg_1-14-R_1-15-cl_1-25-c ms_3-20-R_1 ms_3-2-yg_1	16	9, 21, 44, 45 28
IV	H_2-4-c-18-Lc_2	6	9, 58
V	gl_2-33-ne_1 gl_2-15-bw_1	A	23, 49 46, 47
VI	ia-30-fg	A	34
VII	L^L-?-lp_1	1	10, 62
VIII	ml-41-st_1	4	7, 62
IX	gl_3-39-ne_2 gl_3-20-bw_2	D	23, 49 37, 47
X	rl-32-Rg		

* c centromere of chromosomes 6, 15 and 16.

tests, Endrizzi and Taylor (ibid.) and Kammacher (27) found that the Naked Seed locus was not on chromosome 7 as had been previously reported (58). However, the evidence remains that Naked Seed is an A subgenome mutant (12).

Wilson and Kohel (63) used a translocation stock to provide additional information about the location of L° on chromosome 15 (linkage group II). Results suggested an increase in the overall length of the linkage group. The tentative assignment of lp_2 to this linkage group increases its length even more (see discussion below).

Kammacher and Schwendiman (28) found ms_3 linked to R_1 and yg_1. This finding adds another identified locus to linkage group III. The homozygous mutant alleles ms_3 ms_3 were reported to produce partial male sterility, but Kammacher was able to increase the degree of male sterility through selection (personal communication).

The homeologous linkage groups V (gl_2-bw_1) and IX (gl_3-bw_2) were identified by Lee (36) and by Rhyne (47). Subsequently, Holder, Jenkins and Maxwell (23) found that the duplicate factors for nectariless (ne_1 and ne_2) were also members of these homeologous groups.

Monosomic tests by Endrizzi and Stith (10) have placed lp_1 on chromosome 1 along with L^{L} (linkage group VII). Chromosomes 1 and 15 are homeologues; thus, lp_2 should reside on chromosome 15. However, the results of the present study indicate no linkage of lp_2 with L° or Lg. L° and Lg are in opposite arms of chromosome 15 (9), and if lp_2 were in the proximal region of 15, the tests should have detected linkages with one or the other of these marker loci. Therefore, lp_2 must be distal to L° or Lg.

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