7

Molecular Markers and Quantitative Traits in Gossypium hirsutum *L.*

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Many traits of importance in cotton breeding are controlled by several genes each with small effects and are thus quantitative traits. For many years scientists have focused on identifying and controlling quantitative traits for the improvement of crops. The recent developments and advancements made in the use of molecular markers should hasten the realization of these goals. The association of molecular markers with desirable quantitative traits should contribute to the discovery of genetic variability and aid in the selection of desirable parents and progeny through marker assisted breeding. Uses of molecular markers include loci mapping, linkage studies, and as aids in breeding programs (Paterson *et al.* 1991). Environmental influences on molecular markers are minimal, which increases their usefulness in breeding programs. Association of several molecular markers with quantitative traits would provide an indication of the location of major quantitative loci that influence the traits and also provides additional evidence for the quantitative nature of these traits.

Restriction Fragment Length Polymorphism (RFLP), Random Amplified Polymorphic DNA (RAPD), Amplified Fragment Length Polymorphism (AFLP) and Simple Sequence Repeat (SSR) markers are being used in our research. Molecular markers have been employed to further the knowledge of parental relationships, to map genes for insect and disease resistance, and for the identification of quantitative trait loci (QTL), (Paterson *et al.* 1991; Dudley *et al.* 1992; Giese *et al.* 1993; Schon *et al.* 1993; Lee *et al.* 1996; Shappley *et al.* 1998a, b). Some molecular markers allow the identification of homozygous and heterozygous plants at an early stage of growth. This increases their usefulness in marker assisted selection and breeding.

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Several statistical programs have been used for interval mapping of molecular markers. The MAPMAKER\EXP (Lander *et al.* 1987) program has often been used to construct linkage maps of molecular markers. The MAPMAKER\QTL method (Paterson *et al.* 1988) was the standard for interval mapping for several years. The method of composite interval mapping that Zeng (1993, 1994) developed includes marker information for controlling background noise while searching for the QTL. The marker effects, as well as the QTL effects, in this model are treated as fixed effects. Therefore, the estimated QTL effects could be affected by the markers included in the model. Zhu and Weir (1998) proposed a new method that used a mixed model approach for composite interval mapping of QTLs. In their mixed model approach, QTLs are fixed variables while molecular makers are random variables. Thus, the estimates of QTLs do not depend upon a particular fixed set of markers being in the model. This model also provides important estimates of additive and dominance effects of QTLs.

The first report of RFLP evaluations in *Gossypium hirsutum* L., upland cotton, was by Meredith (1992) in a study of heterosis and variety origins. A RFLP map of cotton with 41 linkage groups was developed by Reinisch *et al.* (1994) by using an interspecific F_2 population from the cross of *G. hirsutum* L. race 'palmeri' by *G. barbadense* L. accession K101. Jiang *et al.* (1998) developed a QTL map showing the location of several fiber traits in a cross of *G. barbadense* × *G. hirsutum*. Interspecific incompatibility usually complicates segregation in interspecific hybrids. Shappley (1994) and Shappley *et al.* (1996) established five linkage groups in a cross of two upland *G. hirsutum* L. cottons. Shappley (1996) and Shappley *et al.* (1998a) also developed a genetic linkage map with 31 linkage groups in upland cotton from the cross of two upland lines. Shappley *et al.* (1998b) associated 100 QTLs with 60 maximum likelihood positions in 24 of these linkage groups and this was the first linkage map of QTLs in a cross of two upland cottons. Such maps should be valuable for analysis and detection of variability in *G. hirsutum*, including the elite germplasm. Since upland cultivars comprise the large majority of cotton in the world, identification of QTLs and their association with molecular markers in segregating generations following crosses of upland cotton lines is of great interest to cotton breeders.

The identification of QTLs controlling traits of interest to breeders of upland cotton and their association with molecular makers has been the focus of our research. We used the breeding line MARCABUCAG8US-1-88 as a male with the cultivar HS 46 as female for our research in the development of linkage maps of molecular markers and their association with important QTL. The HS 46 cultivar traces to a cross of AZ7209 × Acala 90 and MARCABUCAG8US-1-88 is from the Multiple Adversity

Resistance (MAR) program of Texas Agricultural Experiment Station (Calhoun *et al.* 1997), thus they are not closely related.

One F_1 plant from this cross was self-pollinated. We grew 96 F_2 derived F_3 lines ($F_2.F_3$). Bulk samples of leaves were collected from each of these F_3 lines and RFLP analysis was contracted from Biogenetic Services Inc., Brookings, South Dakota. The probes they used were prepared from a cDNA library using leaf material collected from six diverse upland cotton cultivars. Since these were from a cDNA library, the QTL identified using these materials should have a high probability of being associated with actual genes in cotton. Detailed protocols used in the RFLP research are given in Shappley *et al.* (1998a).

The RFLP data from the $F_2.F_3$ families was analyzed with MAPMAKER\EXP 3.0 (Lander *et al.* 1987) for linkage associations. There were 129 probe enzyme combinations resulting in 138 loci. Thus, some probe enzyme combinations revealed more than one locus. Based upon single locus chi-square analyses, the majority of the progeny arrays fit the expected 3:1 (dominant) or 1:2:1 (co-dominant) genotypic ratios. We found 84 co-dominant loci with 76 of these segregating normally and 54 dominant loci with 50 segregating normally. These 138 loci were arranged into 31 linkage groups containing 120 loci with 18 loci not mapped in any linkage group (Shappley, 1996; Shappley *et al.* 1998a). We found 3 of 31 linkage groups that showed abnormal segregation for more than one locus. The 12 of 138 loci that showed abnormal segregation may have been due to such things as gametophyte selection, genetic drift, or cytological attributes. We have found in other research with cytogenetic deficiency lines that 3 of these same 12 loci also show abnormal segregation in these stocks.

Some abnormal segregation is not unusual with molecular markers. Schon *et al.* (1993) reported 18 of 87 markers in an F_2 population in corn showed significant deviation from the expected 1:2:1 ratios with abnormal segregation of molecular markers on each of the 10 chromosomes in corn. Saha (1989) found an excess of heterozygotes in his isozyme alleles in cotton. Bernardo *et al.* (1997) found a wide deviation in the proportion of the genome derived from each parent in the development of inbreds in corn. Thus, our observation of abnormal segregation of 12 loci should not be surprising.

In our research (Shappley *et al.* 1998a), linkage groups contained from two to ten loci. Map distances ranged from 0.0 to 45.7 cM as determined by two-point analyses and measured in probability, by LOD scores. The average distance between loci was 7.0 cM. Linkage group arrangements were determined by using multi-point analyses and the gene orders are given in Shappley *et al.* (1998a). Reinisch *et al.* (1994) estimated that the cotton genome has a map distance of 4600 cM. Our linkage map with 865 cM thus covers 18.6% of the minimum map.

A beginning linkage map in segregating generations following a cross of two *G. hirsutum* L. lines of upland cotton has been established; however further mapping work will be needed to resolve the order of some closely linked markers. Some of these linkage groups have been assigned to specific chromosomes in cotton (Saha *et al.* 1998).

The $F_2.F_3$ families were advanced by bulk self pollination within a family to the F_5 generation. In these $F_2.F_5$ families we grew two rows of each family and measured agronomic and fiber traits. Twenty-five plants in each family were measured for fiber traits. For number of nodes, plant height, and node of first fruiting branch all plants in each F_5 row were measured and the mean was calculated for each family. White bloom counts in the F_3 were taken one time each week for a 4-week period. The percentage of the plants flowering at a given date was calculated for each family. Cottonseeds for seed index were collected from hand-picked boll samples from each family in the F_4 generation. One-hundred fuzzy seeds were counted and weighed to determine an average seed weight for each family.

Agronomic and fiber traits are listed in Table 1. Samples for lint percentage measurements and all measurements of fiber traits were made from hand-picked boll samples, ginned on a 10 saw gin. Conventional and arealometer fiber measurements were conducted by Starlab Inc.,

Table 1. Phenotypic data of agronomic and fiber traits for F_2 derived families from a cross of MARCABUCAG8US-1-88 as parent 1 × HS 46 as parent 2. (From Shappley *et al.* 1998b)

Trait	Mean	SD	Maximum	Minimum	Skewness	Kurtosis
Seed index, g	11.10	1.00	13.50	8.20	−0.10	−0.17
Lint fraction, %	35.70	1.30	39.40	33.10	0.33	−0.20
Micronaire	4.24	0.36	4.95	3.29	−0.45	−0.08
Elongation, %	6.72	0.48	7.79	5.57	−0.34	−0.25
Strength, kgNm kg^{-1}	20.30	1.00	22.60	17.60	0.20	−0.12
50% Span length, mm	13.97	0.51	14.99	12.95	−0.04	−0.70
2.5% Span length, mm	28.45	0.76	30.23	26.42	−0.07	−0.40
Ah	498.00	36.00	602.00	424.00	0.48	0.02
A	471.00	31.00	556.00	409.00	0.49	−0.02
Immaturity	1.68	0.12	2.04	1.43	0.28	−0.02
Maturity, %	86.00	5.00	95.00	72.00	−0.30	0.02
Perimeter, µm	44.80	1.70	49.40	40.20	0.02	−0.42
Weight fineness	3.73	0.28	4.34	3.06	−0.20	−0.17
Wall thickness, µm	2.68	0.22	3.25	2.14	0.06	−0.08
Nodes	18.40	1.30	22.30	14.60	0.11	1.59
Node 1st fruiting branch	6.90	0.40	7.90	6.20	0.59	0.25
Height, cm	77.80	6.00	92.10	64.90	−0.09	−0.50
Height/node ratio	4.30	0.40	5.40	3.40	0.14	−0.05
Bloom rate, %	48.90	17.10	92.10	11.70	−0.07	−0.03

Knoxville, TN, on samples from 25 individual F_5 plants per family. Cottonseed for seed index measurements were collected from hand picked boll samples from each family in the F_4 generation. One-hundred fuzzy seeds were counted and weighed to determine an average seed weight for each family.

Seed index (Sdx) is the weight of 100 ginned, but not delinted, seed and is an indicator of seed size or density. Lint percent (Lp) or lint fraction, is the ratio of lint to the total weight of unginned seed cotton expressed as a percentage. Micronaire (Mic) is a measure of the fineness of the sample of fibers and is reported in standard micronaire units. Elongation (El) is a measure of the elasticity of the fiber sample. The value is determined at the break point in the strength determination and is defined as a percent stretch of the fiber sample at the breaking point. Strength (T1) is the fiber strength of a bundle of fibers measured with two stelometer jaws holding the fiber bundle separated by 0.3175 cm (one-eighth inch) and is measured in kN m kg^{-1}.

The digital fibrograph is an instrument for measuring fiber length. Span length is the distance spanned by a specific percentage of the fibers in the test specimen when the initial starting point of the scanning in the test is considered 100%. The 50% span length (SL50) is the length on the test specimen spanned by 50% of the fibers scanned at the initial starting point. The 2.5% span length (SL2.5) is the length on the test specimen spanned by the longest 2.5% of the cotton fibers scanned at the initial starting point. The 2.5% span length approximates the classer's staple.

The arealometer instrument measures fiber fineness and shape by measuring the resistance a given mass of fibers offers to the flow of air. Fineness and shape measurements are used to calculate immaturity ratio, percentage maturity, perimeter, weight fineness, and wall thickness. The measurement, A, describes the external surface of the fibers of a given volume of fiberous material under standard pressure, expressed in terms of square millimeters per cubic millimeter of fiberous material. The measurement Ah measures the same fibers as the A measurement, but under high pressure. The difference between A and Ah is an estimate of the flatness of the fiber ribbon. The greater the difference the more ribbon-like are the fibers. The immaturity ratio (Im) is a dimensionless number that describes a physical characteristic of the fiber cross section. It is defined as the ratio of the area that the fiber cross section would have, if its perimeter enclosed a circle compared to the area that the perimeter actually encloses.

Measurement of fiber maturity (Mat) is based on the simple linear regression prediction of the caustic soda percent maturity method (Hertel and Craven 1951). The prediction equation is $M=150.5 - 38.1I$, where $I=$ the immaturity value calculated. The perimeter (Per) is defined as the

distance around the outside wall of the fiber section in microns. The weight fineness (Wtfn), or linear density, is defined as the mass per unit length of fiber expressed in micrograms per inch. The fiber wall thickness (Wall) is the measurement in microns of the width of the wall of the cotton fiber. Equations for calculations of each of these traits and their relationships are given in the National Cotton Variety Test Report by Rayburn *et al.* (1996).

To determine if trait data were normally distributed, the skewness and kurtosis values were calculated for each trait (Table 1). The traits segregated continuously and both skewness and kurtosis values, except for number of nodes (kurtosis value 1.59), suggested that the agronomic and fiber traits in the study were normally distributed and thus suitable for QTL analysis. Several of the traits are significantly correlated (Table 2).

With the RFLP linkage data from the $F_2.F_3$ families and the agronomic and fiber data from the $F_2.F_5$ families we were able to determine the association of molecular markers with these agronomic and fiber traits, i.e. we could determine the linkage of RFLP markers with QTLs. We used the mixed model approach where the effects of QTLs are considered fixed and the molecular markers are random (Zhu and Weir, 1998) to calculate the relationship of molecular markers and QTL.

Programs for the mixed model equation approach were written in C. The mixed model equation approach program calculates the likelihood ratio value for testing the presence of a QTL within linkage groups. The approach searches for QTLs along the whole genome by a step of 2.0 cM and also gives estimates of the likelihood ratio value as well as estimates of genetic additive and dominance effects. Since the likelihood ratio is closely approximated by the chi-square distribution, this statistic can be used to test for levels of significance in the likelihood ratio. Likelihood ratio values of 6.63, 7.88, and 10.83 show a QTL significantly associated with the molecular marker at the 0.01, 0.005, and 0.001 level, respectively. Estimated genetic additive and dominance effects were tested for significance by using the standard normal distribution. Additive and dominance effects are defined in these data with respect to the MAR allele. Thus, negative genetic effect values indicate that the MAR allele decreases the phenotypic value of the trait, and a positive value indicates an increase in the phenotype with MAR allele. The HS allele has the opposite effect, i.e. a negative genetic effect indicates that the HS allele increases the phenotypic value of the trait.

Using the mixed model approach a total of 100 QTLs mapped to 60 maximum likelihood locations in 24 linkage groups (Table 3, Fig. 1). Additive and dominance genetic effects are also shown in Table 3 for each QTL. These can be used to determine the relative importance of various QTLs for any given trait. For most traits, alleles at different QTLs from

Table 2. Correlation coefficients among fiber traits in F_5 generation (seed index in F_4). (From Shappley *et al.* 1998b)

	Seed index	Lint fraction	Micro-naire	Elon-gation	Strength	Span length 50%	Span length 25%	Ah	A	Imma-turity	Maturity	Peri-meter	Weight fineness
Micronaire	0.50*	0.13											
Elongation	-0.2	0.12	-0.41**										
Strength	-0.26**	-0.27**	0.36**	-0.52**									
50% Span length	0.29**	-0.31**	0.19	-0.15	0.31**								
2.5% Span length	0.31**	-0.38**	-0.04	-0.20*	0.20*	0.64**							
Ah	-0.54**	-0.08	-0.94**	0.43**	-0.39**	-0.14	0.02						
A	-0.55**	-0.11	-0.95**	0.40**	-0.35**	-0.13	-0.05	0.99*					
Immaturity	-0.35**	0.04	-0.76**	0.53**	-0.55**	-0.16	-0.10	0.89**	0.85*				
Maturity	0.36**	-0.04	0.76**	-0.53**	0.54**	0.15	0.10	-0.89**	-0.85**	-0.99*			
Perimeter	0.23*	0.25**	0.15	0.34**	-0.45**	-0.06	-0.25**	0.03	-0.05	0.48**	-0.48**		
Weight fineness	0.51**	0.23*	0.86**	-0.16	0.06	0.06	-0.21*	-0.80**	-0.85**	-0.45**	0.45**	0.57**	
Wall thickness	0.23*	-0.09	0.91**	-0.46**	0.42**	0.08	-0.10	-0.98**	-0.97**	-0.92**	0.92**	0.13	0.74

*,** Significantly different from zero at the 0.05 and 0.01 levels of probability, respectively.

Linkage Group 3

POS	MM or QTL
0.0	C41F5RI
3.9	C24A4RI
18.5	*sl50*
49.1	C81A5RI

Linkage Group 11

POS	MM or QTL
0.0	F3B9RV
0.5	*el*
2.5	*mic*
27.8	C65C2RI
40.9	C43E4RV
83.0	C19B3RV
86.5	*sdx*
92.8	C26B5RV

Linkage Group 4

POS	MM or QTL
0.0	C116C6RI
30.5	*el*
32.0	C121B1RV
32.5	*sdx*
34.4	C44B4RV
36.5	*lp*
40.6	C46F1RI
57.9	C34E3RV
88.2	C108C5RV
100.6	C42E4RI
102.2	C56E1RIB
103.8	C112E4RI
107.7	C111A6RI

Linkage Group 6

POS	MM or QTL
0.0	C84B3RI
6.5	*mic,t1,wall*
7.1	C16A1RI
8.5	*el*
41.0	*hgt*
41.7	C56A6RIB
44.2	C79F2RIB
46.5	*sl50*
46.6	C56E1RIA
48.6	C56E1RV

Linkage Group 7

POS	MM or QTL
0	C15A4RV
14.5	*bl*
18.4	C50D1RV
18.5	*mic,el*
22.5	C120F1RV
38.5	*bl*
40.2	C45B6RV

Linkage Group 10

POS	MM or QTL
0.0	C50C1RIB
1.6	C61A1RIB
2.5	*mic, el, wtfn*
8.5	*hnrat*
12.2	C34F5RV
14.9	C26D5RV
22.5	*nfb*
22.5	C5F4RI
23.0	C112E6RV
34.7	C116D3RV
52.5	C57D3RI
54.5	*a, wall*
66.5	*lp, t1*
67.3	C89B4RV

Linkage Group 13

POS	MM or QTL
0.0	C38E2RI
1.1	C87F1RV
2.5	*t1*
12.0	C112F3RI

Linkage Group 12

POS	MM or QTL
0.0	C78C3RI
0.5	*sl2.5*
7.7	C17A6RI

Linkage Group 14

POS	MM or QTL
0.0	F4B4RV
2.5	*mic,el, ah, wall*
4.5	*a*
38.5	*sdx*
39.3	C104A1RI
41.4	C100A2RV
41.9	C116D6RI
42.5	*a, im*
53.3	C117C5RV
54.2	C117C5RI
54.5	*sdx, mic*
54.7	F2E6RI

Linkage Group 9

POS	MM or QTL
0.0	C50C1RIA
0.5	*mic, ah, a, wall*
10.9	C81F4RIA
20.5	*wtfn*
28.6	C61A1RIA

POS	MM or QTL
0.0	C107B2RIA
0.5	*el*
6.3	C52E2RV
7.9	C107B2RIB
10.1	C107B2RV
10.5	*lp*
38.9	C55B1RI

POS	MM or QTL
0.0	F3A9RV
10.3	C47B1RV
12.4	C17F2RV
14.5	*mic, el, wtfn*
16.5	*wtfn*
47.8	F3B2RI
61.1	F4B6RV
61.6	F2C11RV
62.5	*sl2.5*
66.0	C86B3RI

POS	MM or QTL
0.0	C56A6RIA
22.3	C111C3RV
27.3	C27B6RI
27.8	F2A4RV
29.4	F9D3RV
47.6	C80F1RV
48.5	*el*
50.5	*mic, a*
52.5	*ah, wall*
54.5	*im, mat*
56.5	*t1*
73.9	C71E2RI

POS	MM or QTL
0.0	C44D2RI
0.5	*node, hgt*
6.5	*hnrat*
8.9	C87E4RI

POS	MM or QTL
0.0	C116E4RI
0.5	*mic, el*
4.5	*t1*
5.6	C116E4RV

Linkage Group 16

POS	MM or QTL
0.0	C13F3RV
0.5	*el*
10.5	*lp*
14.5	*sl2.5*
16.5	*sl50*
29.2	C26D2RI
32.8	C16F4RV

Linkage Group 18

POS	MM or QTL
0.0	C18ARI
0.5	*el*
3.0	C18A4RV

Linkage Group 20

POS	MM or QTL
0.0	C28C1RV
2.5	*t1*
8.5	*mic, el*
8.5	F3F7RI

Linkage Group 21

POS	MM or QTL
0.0	C33A6RV
0.5	*el*
1.0	C33A6RI
1.9	F6D4RV

Linkage Group 31

POS	MM or QTL
0.0	F7C8RI
0.5	*node*
7.5	C118C4RVA

Linkage Group 24

POS	MM or QTL
0.0	C113C4RI
0.5	*mic*
6.5	*el*
6.8	C113F6RI
38.9	C79F2RIA
50.5	*mic, ah, a, im, mat, , wtfn, wall*
50.6	C114B1RI

Linkage Group 25

POS	MM or QTL
0.0	C101D1RV
0.5	*mic, el, wtfn*
2.5	*lp*
16.7	C106D2RV

Linkage Group 28

POS	MM or QTL
0.0	C88C2RI
0.5	*el, 2.5sl*
1.3	C101B1RI
4.0	C118C3RI
4.5	*ah, im, mat*
6.5	*mic,, wall*
6.7	C117B3RI

Linkage Group 30

POS	MM or QTL
0.0	C61A6RV
0.5	*el*
7.4	C117F1RI
9.6	C115A6RI

Fig. 1. *Linkage map of molecular markers and maximum likelihood estimates of location of QTLs. Molecular markers shown in upper case and QTL loci shown in lower case italics. From Shappley et al. Journal of Cotton Science 4, 153-163.*

Table 3. Maximum likelihood locations of agronomic and fiber trait QTLs, likelihood ratio values, and estimates for additive and dominance effects relative to MAR base phenotype. Mixed model analysis of an F_2 derived population of 96 families from a cross of MARCABUCAG8US-1-88 × HS 46. (From Shappley *et al.* 1998b)

QTL trait	Linkage group	Map dis† (cM)	LR‡	Add. effect	± SE	Dom. effect	± SE
Seed index, g	4	32.5	7.64 **	–1.18 *	±0.51	–0.44	±0.41
Seed index, g	11	86.5	7.79 **	–2.00 *	±0.83	–0.91	±0.61
Seed index, g	14	38.5	12.03****	–0.28 *	±0.13	0.46	±0.25
Seed index, g	14	54.5	9.7 ***	–0.26 *	±0.12	0.38 *	±0.18
Lint fraction, %	4	36.5	9.54***	0.58 **	±0.19	0.79	±0.78
Lint fraction, %	10	66.5	8.82***	–0.40 *	±0.18	0.36	±0.34
Lint fraction, %	15	10.5	7.41 **	0.43 **	±0.16	0.46	±0.34
Lint fraction, %	16	10.5	12.16****	1.32 **	±0.38	1.08 *	±0.42
Lint fraction, %	25	2.5	8.55***	1.95 *	±0.87	0.63	±0.63
Micronaire	6	6.5	7.97***	0.95 *	±0.39	0.74 **	±0.26
Micronaire	7	18.5	7.03 **	0.99 **	±0.38	0.63 *	±0.26
Micronaire	9	0.5	8.25***	–0.13 **	±0.05	–0.16	±0.09
Micronaire	10	2.5	7.76 **	0.30 *	±0.13	0.75 **	±0.28
Micronaire	11	2.5	7.86 **	0.37 **	±0.13	0.65 *	±0.26
Micronaire	14	2.5	16.00****	1.07 **	±0.30	0.54 *	±0.22
Micronaire	14	54.5	19.75****	0.11 *	±0.04	–0.22 **	±0.07
Micronaire	17	14.5	8.66***	0.40 **	±0.14	0.81 **	±0.28
Micronaire	19	50.5	7.49 **	0.33 *	±0.14	0.69 **	±0.26
Micronaire	20	8.5	7.03 **	0.35 **	±0.13	0.65 *	±0.26
Micronaire	24	0.5	7.76 **	1.03 **	±0.37	0.70 **	±0.26
Micronaire	24	50.5	10.58***	–0.01	±0.05	0.26 **	±0.09
Micronaire	25	0.5	9.05***	1.00 *	±0.38	0.51	±0.26
Micronaire	27	0.5	7.14 **	0.31 *	±0.13	0.68 *	±0.26
Micronaire	28	6.5	6.75 **	–0.02	±0.04	0.23 *	±0.10
Elongation, %	4	30.5	9.44***	–1.61 **	±0.53	–1.08 **	±0.37
Elongation, %	6	8.5	11.28****	–1.40 **	±0.47	–1.13 **	±0.34
Elongation, %	7	18.5	13.44****	–1.58 **	±0.50	–0.82 *	±0.34
Elongation, %	10	2.5	10.75***	–0.54 **	±0.17	–0.93 *	±0.36
Elongation, %	11	0.5	12.88****	–0.59 **	±0.17	–1.00 **	±0.34
Elongation, %	14	2.5	16.93****	–1.63 **	±0.41	–0.93 **	±0.30
Elongation, %	15	0.5	8.40***	–0.45 *	±0.17	–1.03 **	±0.36
Elongation, %	16	0.5	7.63 **	–1.39 **	±0.51	–0.96 **	±0.35
Elongation, %	17	14.5	10.48***	–0.50 **	±0.18	–1.13 **	±0.36
Elongation, %	18	0.5	7.98***	–0.45 **	±0.16	–0.99 **	±0.35
Elongation, %	19	48.5	11.61****	–0.48 **	±0.18	–1.11 **	±0.35
Elongation, %	20	8.5	9.57***	–0.53 **	±0.17	–1.06 **	±0.35
Elongation, %	21	0.5	9.63***	–0.69 **	±0.26	–3.43 **	±1.11
Elongation, %	24	6.5	11.00****	–1.56 **	±0.52	–1.18 **	±0.36
Elongation, %	25	0.5	10.61***	–1.23 *	±0.51	–1.04 **	±0.35
Elongation, %	27	0.5	9.70***	–0.45 *	±0.18	–1.03 **	±0.34
Elongation, %	28	0.5	6.84 **	–0.14	±0.14	–0.41 **	±0.16
Elongation, %	30	0.5	7.31***	–0.44 *	±0.17	–0.95 **	±0.35
Strength, kgNmkg^{-1}	6	6.5	6.82 **	17	±10.3	16 *	±7.1

(*Contd.*)

Table 3. (*Contd.*)

QTL trait	Linkage group	Map dis†(cM)	LR‡	Add. effect	± SE	Dom. effect	± SE
Strength, kgNmkg^{-1}	10	66.5	6.72 **	3 *	±1.4	−1	±2.6
Strength, kgNmkg^{-1}	13	2.5	12.21****	5 **	±1.5	−0	±3.2
Strength, kgNmkg^{-1}	19	56.5	6.97 **	6 *	±3.2	14 *	±5.7
Strength, kgNmkg^{-1}	20	2.5	7.15 **	8 *	±3.2	13 *	±6.3
Strength, kgNmkg^{-1}	27	4.5	7.96***	5	±3.5	14 *	±6.7
50% Span length	6	46.5	8.60***	0.03 *	±0.00	−0.00	±0.03
50% Span length	16	16.5	7.45 **	−0.03 *	±0.00	−0.03 *	±0.03
2.5% Span length	3	18.5	9.85***	0.03 *	±0.03	−0.00	±0.03
2.5% Span length	12	0.5	7.08 **	0.03 *	±0.00	−0.00	±0.03
2.5% Span length	16	14.5	7.38 **	−0.05 *	±0.03	−0.05 *	±0.03
2.5% Span length	17	62.5	9.40***	0.03 *	±0.00	0.05 *	±0.03
2.5% Span length	28	0.5	6.80 **	0.05 *	±0.03	0.05	±0.03
Ah	9	0.5	7.38 **	11.99 *	±4.79	17.38	±9.08
Ah	14	2.5	13.84****	−95.88 **	±30.16	−45.55 *	±21.71
Ah	14	54.5	18.63****	−10.66 *	±4.32	21.58 **	±6.54
Ah	19	52.5	7.03 **	−25.27	±13.51	−58.86 *	±24.91
Ah	24	50.5	12.01****	2.47	±4.64	−27.05 **	±8.89
Ah	28	4.5	6.66 **	2.86	±4.33	−22.13 *	±10.24
A	9	0.5	8.09****	10.92 **	±4.08	14.62	±7.74
A	10	54.5	6.83**	−20.05	±10.71	−51.72 *	±21.60
A	14	4.5	13.34****	−55.73**	±18.75	−20.80	±14.92
A	14	42.5	16.77****	−9.92*	±4.14	17.02 *	±8.41
A	19	50.5	6.66 **	−23.60*	±11.65	−52.86 *	±22.13
A	24	50.5	12.28****	1.89	±3.95	−23.49 **	±7.57
Immaturity	14	42.5	12.68****	−0.04 *	±0.02	0.05	±0.03
Immaturity	19	54.5	8.65***	−0.06	±0.04	−0.17 *	±0.08
Immaturity	24	50.5	7.69 **	0.01	±0.02	−0.07 *	±0.03
Immaturity	28	4.5	8.03***	0.01	±0.01	−0.08 *	±0.03
Maturity, %	14	54.5	13.82****	1.56 **	±0.57	−1.98 *	±0.87
Maturity, %	19	54.5	8.63***	2.32	±1.68	6.50 *	±3.02
Maturity, %	24	50.5	7.86 **	−0.48	±0.61	2.64 *	±1.16
Maturity, %	28	4.5	8.09***	−0.51	±0.55	2.97 *	±1.31
Weight fineness	9	20.5	10.36****	−0.12 **	±0.04	−0.09	±0.07
Weight fineness	10	2.5	10.76***	0.18	±0.10	0.55 **	±0.21
Weight fineness	14	54.5	13.39****	0.04	±0.03	−0.17 **	±0.05
Weight fineness	17	14.5	7.05 **	0.28 *	±0.11	0.55 **	±0.21
Weight fineness	17	16.5	7.97***	0.28 **	±0.10	0.57 **	±0.20
Weight fineness	24	50.5	10.06***	0.00	±0.04	0.20 **	±0.07
Weight fineness	25	0.5	13.95****	0.73 *	±0.29	0.29	±0.20
Wall thickness, µm	6	6.5	7.07 **	0.61 *	±0.24	0.44 **	±0.17
Wall thickness, µm	9	0.5	7.93***	−0.07 *	±0.03	−0.12 *	±0.06
Wall thickness, µm	10	54.5	7.45 **	0.16 *	±0.08	0.39 *	±0.15
Wall thickness, µm	14	2.5	12.05****	0.57 **	±0.18	0.29 *	±0.13
Wall thickness, µm	14	54.5	20.64****	0.08 **	±0.03	−0.13 **	±0.04
Wall thickness, µm	19	52.5	8.20***	0.15	±0.08	0.37 *	±0.15
Wall thickness, µm	24	50.5	10.82****	−0.02	±0.03	0.15 **	±0.05
Wall thickness, µm	28	6.5	8.03***	−0.03	±0.03	0.12 *	±0.06

(*Contd.*)

Table 3. (*Contd.*)

QTL trait	Linkage group	Map dis†(cM)	LR‡	Add. effect	± SE	Dom. effect	± SE
Nodes	14	54.5	7.14 **	0.01	±0.15	−0.63 **	±0.24
Nodes	23	0.5	7.55**	−0.22	±0.16	−0.92**	±0.34
Nodes	31	0.5	6.71 **	−0.08	±0.16	0.69 *	±0.33
Node 1st fruiting branch	10	22.5	9.06***	−0.01	±0.05	0.27 **	±0.09
Height, cm	6	40.5	7.67 **	0.61	±0.72	-3.56 *	±1.65
Height, cm	23	0.5	10.33***	−2.44 **	±0.76	-1.17	±1.64
Height/node ratio	10	8.5	6.71 **	−0.07	±0.06	-0.37 *	±0.15
Height/node ratio	23	6.5	6.83 **	−0.04	±0.05	0.23 *	±0.11
Bloom rate, %	7	14.5	7.79 **	8.88	±9.41	17.09 *	±7.75
Bloom rate, %	7	38.5	9.70***	0.38	±2.22	13.15 **	±4.37

*, **, ***, **** Significant at the 0.05, 0.01, 0.005 and 0.0001 levels of probability, respectively.

† Map distance from first molecular marker in linkage group to the estimated location of the QTL.

‡ LR is likelihood ratio of the QTL.

either parent could contribute to increased performance for the trait. Shappley *et al.* (1998b) show the detailed linkage maps between the molecular markers and the QTL. At least one QTL was identified for each of the 19 agronomic and fiber traits except perimeter of fiber. Fiber traits were measured on 25 individual plants and thus provided an exceptional measurement for the individual family means and variances.

Highly correlated traits (Table 2) show similar QTL results in the mixed model analyses (Shappley *et al.* 1998b). For example a group of highly correlated traits, micronaire, A, Ah, immaturity, maturity, wall thickness, and weight fineness are influenced by fiber fineness and maturity. A few linkage groups have been identified as associated with specific chromosomes (Saha *et al.* 1998). Our putative locations of the QTLs do not necessarily represent physical distances. Thus, a physical map of the linkage groups would be very useful in cloning selected QTLs in cotton. Thus, from the present knowledge we can form a beginning for progress in understanding QTLs in upland cotton and how they are distributed and/or associated among linkage groups. The association of these linkage groups with chromosomes in the A or D genome should prove useful to basic and applied research programs in the future. This knowledge of these associations could target selected chromosomes for further analysis such as the development of chromosome substitution lines with specific chromosomes from other species. We are currently working in this direction.

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