Theor Appl Genet (2001) 103:1232–1242

G.E. Hart · K.F. Schertz · Y. Peng · N.H. Syed

Genetic mapping of *Sorghum bicolor* (L.) Moench QTLs that control variation in tillering and other morphological characters

Received: 20 September 2000 / Accepted: 26 October 2000

Abstract Grain yield of *Sorghum bicolor* (L.) Moench is significantly influenced by genetically controlled variation in the number of tillers, plant height, time of anthesis, and various other morphological and physiological characters. In this study, a minimum of 27 unique QTLs that control variation in nine morphological traits, including the presence versus the absence and the height of basal tillers, were mapped, and the percentage of additive genetic variance explained by the QTLs was determined in a population of 137 recombinant inbred lines in two environments. Four QTLs explained from 86.3% to 48.9% (depending upon the environment) of the additive genetic variance in the number of basal tillers with heads, and seven QTLs explained from 85.9% to 47.9% of the additive genetic variance in panicle width. It is unlikely that different alleles were segregating in the mapping population at any of the major dwarfing loci, but five QTLs that explained from 65.8% to 52.0% of the additive genetic variance in main-culm height were mapped. QTLs controlling variation in height of the tallest basal tiller, number of basal tillers per basal-tillered plant, panicle length, leaf angle, maturity, and awn length also were mapped. Three or more QTLs were mapped in linkage groups A, E, G, and I, while none were mapped in linkage groups B and D. Several of the QTLs mapped in this study are likely candidates for marker-assisted selection in breeding programs.

Communicated by P.M.A. Tigerstedt

G.E. Hart (✉) · Y. Peng · N.H. Syed
Department of Soil and Crop Sciences, Texas A&M University, College Station, TX, 77843, USA
e-mail: g-hart@tamu.edu
Fax: +1-409-8456483

K.F. Schertz
USDA-ARS, Texas A&M University, College Station, TX 77843, USA

Present address:
Y. Peng, Synergene Seed & Technology, 1147 Madison Lane, Salinas, CA, 93907, USA

Keywords *Sorghum bicolor* · QTLs · Recombinant inbred lines · Genetic mapping

Introduction

Sorghum is one of the world's most important crop plants, ranking fifth in acreage among the cereals (Doggett 1988). It is used for forage, as a source of sugar and syrup, as feed for livestock, and to make numerous food products. Grown principally in the semi-arid regions of the tropics and subtropics, it has numerous characteristics, including drought tolerance and resistance to various diseases and pests, that adapt it to the harsh environments of these regions. *Sorghum bicolor* ssp. *bicolor* ($2n=2x=20$) is the most important taxon agronomically in that it includes the cultivated grain races, but wild races in other *S. bicolor* subspecies that grow in proximity to the cultivated races in Africa and are usually interfertile with them also possess a significant amount of genetic diversity in traits of agronomic importance.

It is well-known that genetically controlled variation in height, maturity (time of anthesis), number of tillers, panicle length, and other morphological and physiological characters influence grain yield, the most important agronomic trait of sorghum. Classical genetic studies have contributed information regarding the manner of genetic control of some of these characters. For example, variation in maturity is known to be controlled principally by six genes (Quinby and Karper 1945; Quinby 1966; Rooney and Aydin 1999) and dwarfism is principally controlled by four genes (Quinby and Karper 1954; Hadley 1957). The recent construction of molecular marker maps of sorghum has made it possible to map these and other genetic loci that control variation in quantitative traits. Among the traits for which controlling regions of the sorghum genome have been identified using molecular markers are plant height (Lin et al. 1995; Pereira and Lee 1995; Rami et al. 1998; Klein et al. 2001), maturity (Lin et al. 1995; Crasta et al. 1999), number of tillers (Paterson et al. 1995a), seed weight

(Pereira et al. 1995), panicle characteristics (Pereira et al. 1995; Rami et al. 1998), 'stay green' (Tuinstra et al. 1996, 1997; Xu et al. 2000; Tao et al. 2000), and disease resistance (Klein et al. 2001). Many of the genes with major effect that control plant height and maturity are unmapped as yet, however, as are the genes that control many other quantitative characters that are of agronomic importance.

The objectives of the research reported in this paper were to genetically map quantitative trait loci (QTLs) that control variation in the number and height of basal tillers and several other morphological characters and to quantify the amount of variation controlled by the mapped QTLs. Mapping was performed in a recombinant inbred population derived from *S. bicolor* accession BTx623, which lacks basal tillers, and IS3620C, which produces them. A linkage map composed of 470 restriction fragment length polymorphisms (RFLPs) and simple sequence repeats (SSRs) is available for the population (Peng et al. 1999; Kong et al. 2000; Bhattramakki et al. 2000), and a physical map of sorghum is being constructed with a bacteria artificial chromosome (BAC) library constructed from BTx623 (Klein et al. 2000). Reported here are the map locations and quantitative effects of genetic loci that control basal-tiller number and height, main-culm height, panicle length and width, leaf angle, awn length, and maturity. Also reported are the map locations of genes that control the presence versus the absence of awns and dry versus juicy midribs.

Materials and methods

Plant materials

Mapping was performed in a population of 137 F_{6-8} recombinant inbred lines (RILs) developed by Dr. K. F. Schertz from a cross of BTx623×IS3620C (Peng et al. 1999). BTx623 is an agronomically important inbred line that was derived from a cross between BTx3197, a kafir line, and SC170–6, a zera zera line. IS3620 C is a guinea line. It was originally collected in Gamjin, Nigeria, where it was known as 'KO37 Canjin.' The donor parent used for conversion of IS3620C to a short-day photoperiod was BTx406.

Design of field trials and phenotypes analyzed

The RILs and parental lines were evaluated in field trials at College Station (CS) and Lubbock (LBK), Texas, in 1994, with the RILs distributed in a randomized complete block design with two replications (plots) at each location and with six replications of each parent at each location. Each plot consisted of a row 5.5 m in length, with a spacing of 0.076 m between plants and 0.76 m between rows at CS and 0.97 m between rows at LBK. The quantitative traits analyzed were: height of the main culm (CUH, in centimeters, average of 10 plants/plot), height of the tallest basal tiller on basal-tillered plants (TIH, in centimeters, average of up to 10 plants/plot), number of basal tillers with heads (TINA; 10 plants/plot), number of basal tillers/basal-tillered plant (TINB; up to 10 plants/plot), angle of the third leaf-blade-junction relative to the main culm (LEA; 1–5, 1=parallel, 2=approximately 22.5° angle, 3=approximately 45° angle, 4=approximately 67.5° angle and 5=90° angle, 1 rating/CS plot and 1 rating/plot of 1 LBK replication), panicle width (PAW; in centimeters, 3 plants/plot), panicle length (PAL; in centimeters, 3 plants/plot), maturity (MA; number of days from planting until 50% of plants flower, 1 rating/plot), and length of awns of awned plants (AWL; in millimeters, 1 rating/CS plot). Means over replications were used in data analyses of these traits. Two qualitative traits, presence versus absence of awns (1 rating/CS plot) and dry versus juicy midribs (1 rating/CS plot), also were analyzed.

Table 1 Characteristics of the molecular marker map that was used for QTL and gene mapping

Linkage group	Length (cM)	Number of markers	Average distance between markers (cM)
A	163.5	19	9.1
B	167.6	18	9.9
C	154.9	19	8.3
D	138.5	13	11.5
E	133.7	14	10.3
F	127.1	15	9.1
G	119.4	14	9.2
H	112.3	14	8.6
I	83.0	11	8.3
J	78.8	8	11.3
Total	1278.8	145	9.5

Linkage map

QTLs and gene loci were placed on a linkage map composed of a subset of the 323 RFLPs and 147 SSRs mapped in the BTx623×IS3620C RI population by Peng et al. (1999), Kong et al. (2000), and Bhattramakki et al. (2000). Criteria for the map, which was constructed using the computer program MAPMAKER Macintosh v2.0, included spacing of markers at intervals of 5–15 cM to the maximum extent possible, a minimum LOD score ≥3.0 for terminal triplets, and a minimum LOD score ≥5.0 for non-terminal triplets. Recombination frequencies were converted to centiMorgans using the Kosambi function (Kosambi 1944). The map consisted of 145 markers spaced at an average interval of 9.5 cM (Table 1, Fig. 1).

Data analyses

Trait means and standard deviations were calculated using Microsoft Excel (Microsoft, Tacoma, Wash.). QTL analyses were performed with the marker-regression method of Kearsey and Hyne (1994), using software obtained from the web site – http://web.bham.ac.uk/G.G.Seaton/. ANOVAs were performed to test for the presence of one or more QTLs, and 1,000 simulations were conducted to disclose the probabilities associated with ANOVA *F*-values and confidence intervals for the estimated QTL positions. With this method, deviations from normality in the distribution of traits do not disturb QTL detection. Also, because the *F* tests are assessed by simulation, the normal significance levels of 5%, 1%, and 0.1% are applicable. The percentage of additive genetic variance explained by each QTL was estimated as

$$\%\ V_A \text{ explained} = a^2 \times 100/2V_A,$$

where a is the additive effect of the QTL. Exceptions were QTLs controlling the same trait that were located in the same linkage group (LG). For these, a combined '% V_A explained' was estimated, with

$$a^2 = a_{xy}^2 = \frac{a_x^2}{2} + \frac{a_y^2}{2} + \delta_{xy}(1 - 2R_{xy})a_x a_y$$

where x and y are the QTLs, δ=–1 with linkage in repulsion and +1 with linkage in coupling, and R is the recombination frequency (Kearsey and Pooni 1996). The QTL data also were analyzed with the computer program QTL CARTOGRAPHER (Basten et al. 1994, 1997). Similar findings were obtained, but they are not reported here.

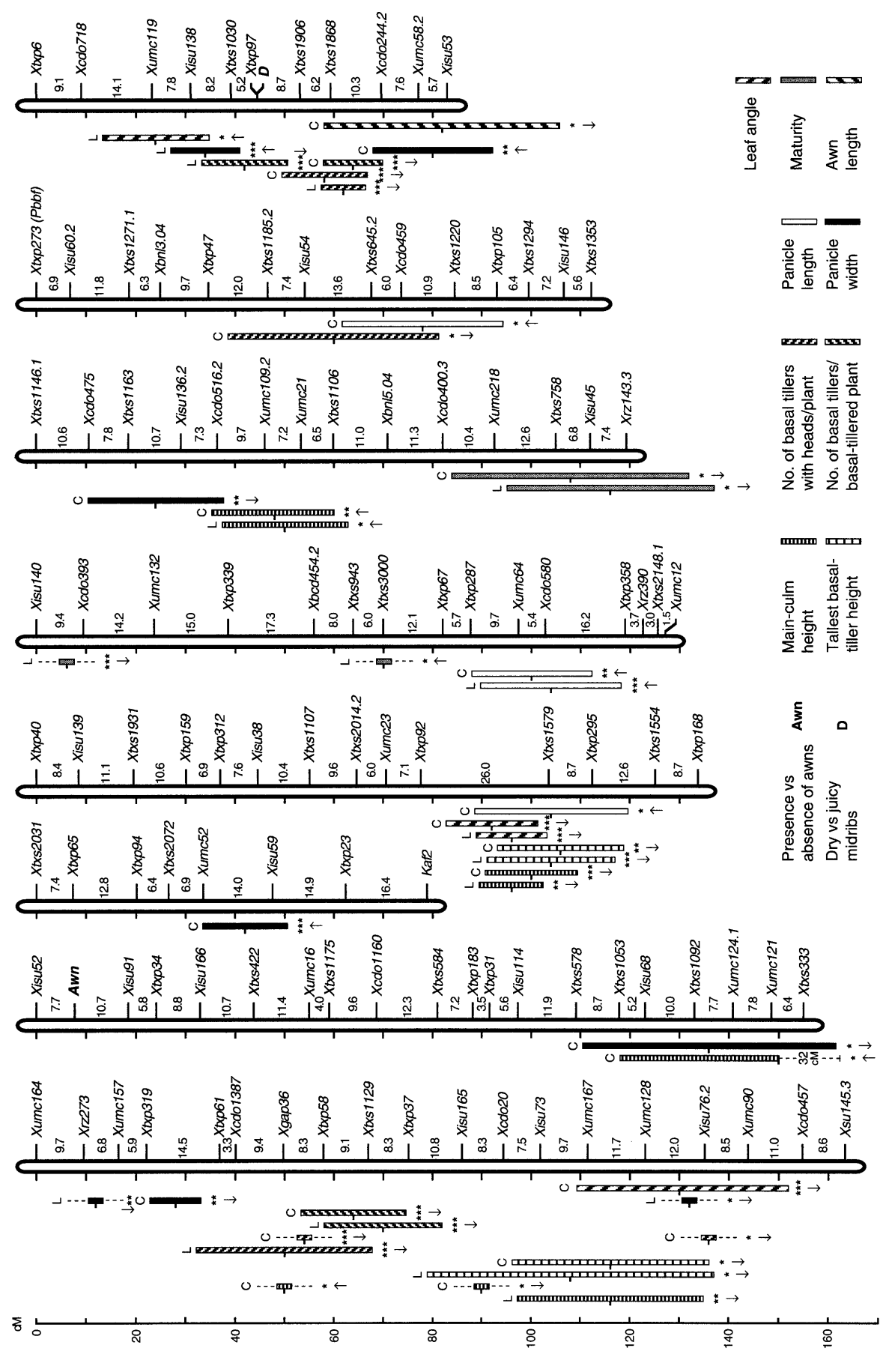
A
C
J
E
F
G
H
I
cM
Presence vs absence of awns
Awn
Dry vs juicy midribs
D
Main-culm height
Tallest basal-tiller height
No. of basal tillers with heads/plant
No. of basal tillers/ basal-tillered plant
Panicle length
Panicle width
Leaf angle
Maturity
Awn length

Table 2 Trait means ($\bar{x}$) and standard deviations (SD) of BTx623, IS3620C, and the recombinant inbred lines (RILs), and trait ranges and coefficients of variation (cv) of the RILs

Traits[a]	BT×623				IS3620C				Recombinant inbred lines							
	College Station		Lubbock		College Station		Lubbock		College Station				Lubbock			
	$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD	Range	cv (%)	$\bar{X}$	SD	Range	cv (%)
CUH	140±4.6		119±4.7		99±4.7		134±10		140±40		77–239	29	134±41		67–220	31
TIH	–[b]		–		127±14		144±14		137±40		62–219	29	143±41		71–223	29
TINA	–		–		1.4±0.4		1.5±0.5		0.4±0.4		0–1.6	10	0.7±0.5		0–2.3	71
TINB	–		–		1.4±0.4		1.5±0.5		1.3±0.4		1–2.2	31	1.5±0.4		1–2.8	27
PAL	34±3.4		32±2.1		28±2.5		30±4.0		36±5.2		21–51	14	30±4.7		21–47	16
PAW	7.2±1.2		7.5±0.9		13.2±3.2		12.4±2.8		14.3±5.2		5.9–29	36	13.7±4.2		6.7–23	31
LEA	2.8±0.4		3.0±0.0		4.0±0.0		3.7±0.6		3.4±0.7		2–5	21	3.5±0.9		1–5	26
MA	66±3.0		59±3.6		61±1.5		69±4.7		68±6.3		55–90	09	64±9.3		46–85	15
AWL	–[c]		dnr[d]		10±0.0		dnr		7.3±3.2		1–13.5	44	dnr			

[a] CUH, main-culm height; TIH, tallest basal-tiller height; TINA number of basal tillers with heads per plant; TINB, number of basal tillers per basal-tillered plant; PAL, panicle length; PAW, panicle width; LEA, leaf angle; MA, maturity; AWL, awn length

[b] Basil tillers not produced

[c] Awnless

[d] dnr, Data not recorded

MAPMAKER Mcintosh v2.0 was used to assign map positions to genes controlling the presence versus absence of awns and dry versus juicy midribs.

Genetic nomenclature

The designations *Awn* and *D* were assigned to genes controlling the presence versus absence of awns and dry versus juicy midribs, respectively. QTLs were designated with italicized symbols consisting of a *Q*, a trait designator (composed of a capital Roman letter and one or more lower case letters), a period, *txs* as a laboratory designator, a hyphen, the symbol for the LG in which the QTL is located, and, in instances where two loci controlling a trait were detected in the same LG, an Arabic numeral. For example, *QCuh.txs-A1* designates one of the main-culm-height QTLs that was mapped in LG A.

◀ **Fig. 1** Molecular marker map of *Sorghum bicolor* showing the locations of QTLs and genes that were mapped in the BTx623×IS3620C recombinant inbred population. CentiMorgan distances between markers and the locations of mapped genes are shown to the *right* of the linkage groups (LGs). Locations of QTLs are indicated by *bars* to the *left* of the LGs. *C* and *L* designate the College Station and Lubbock trials, respectively. The most likely location of each QTL is indicated by the *horizontal line* attached to the *left side* of each bar. The lengths of the bars delineate 95% confidence intervals for the QTL locations, except for QTLs controlling the same trait that are located in the same LG. 95% confidence intervals for these QTLs, which are delineated by *bars* that have *vertical dashed lines above* and *below* them, could not be determined with the marker regression software program that was utilized. *Asterisks* indicate levels of significance associated with QTLs, with single (*), double (**), and triple asterisks (***) indicating the 0.05, 0.01, and 0.001 levels, respectively (see Table 3). An *upward-pointing arrow below a bar* indicates that the BTx623 allele conditions a larger value for the parameter analyzed than the IS3620C allele, and a *downward-pointing arrow*, that it conditions a smaller value. A 170-cM-long scale is located at the *left side* of the figure, and 10-cM-long intervals are indicated by *horizontal lines attached* to the *left sides* of the LGs

Linkage group designations

The designations assigned to sorghum LGs by Peng et al. (1999) are used in this paper. When reference is made to a sorghum LG developed by other investigators, the designation assigned to it by the other investigators, if different (see Table 1 of Peng et al. 1999), is also listed in parentheses.

Results and discussion

The trait means of the parents and RILs indicated substantial differences between BTx623 and IS3620 C for most of the traits analyzed (Table 2). A high degree of genotype-environment interaction was expressed for main-culm height and maturity, however, with BTx623 being much taller and maturing slightly later than IS3620 C at College Station (CS) and marginally shorter and maturing slightly earlier than IS3620C at Lubbock (LBK). Panicle length varied significantly between the parents at CS but not at LBK.

ANOVAs revealed differences among the RILs at the 0.001 level of significance in both environments for the traits analyzed with only one exception, namely, the number of basal tillers per basal-tillered plant in the LBK environment, where P=0.005 (Table 3). Significant differences among replications were detected in five instances, namely, for main-culm height, number of basal tillers with heads, and maturity in the CS environment, and for panicle width and maturity in the LBK environment.

Height of main-culms and tallest basal tillers

Evidence for three main-culm-height (CUH) QTLs located in LGs A, E, and G was obtained in both environments, and the 95% confidence interval for one of the

Table 3 Results of ANOVAs of the data obtained for the nine quantitative traits that were analyzed together with estimates of genetical and environmental components

Source	College Station				Lubbock			
	df	MS	*P*	ems	*df*	MS	*P*	ems
Main-culm height								
RILs	136	3,368.3	<0.001	$\sigma^2+2\sigma^2b$	136	2,538.0	<0.001	$\sigma^2+2\sigma^2b$
Reps	1	469.3	0.002	$\sigma^2+137\sigma^2b$	1	1,395.0	0.176	$\sigma^2+137\sigma^2b$
Error	136	46.6		σ^2	136	756.0		σ^2
				Estimates				
V_A (=1/2σ^2b)		830.4				445.5		
V_E (=σ^2)		46.6				756.0		
Tallest basal-tiller height								
RILs	81	2,755.0	<0.001	$\sigma^2+2\sigma^2b$	112	2,735.0	<0.001	$\sigma^2+2\sigma^2b$
Reps	1	720.0	0.222	$\sigma^2+82\sigma^2b$	1	1,514.0	0.131	$\sigma^2+113\sigma^2b$
Error	81	476.0		σ^2	112	654.0		σ^2
				Estimates				
V_A (=1/2σ^2b)		569.8				520.3		
V_E (=σ^2)		476.0				654.0		
Number of basal tillers with heads per plant								
RILs	136	0.191	<0.001	$\sigma^2+2\sigma^2b$	136	0.398	<0.001	$\sigma^2+2\sigma^2b$
Reps	1	0.900	0.004	$\sigma^2+137\sigma^2b$	1	0.008	0.815	$\sigma^2+137\sigma^2b$
Error	136	0.102		σ^2	136	0.149		σ^2
				Estimates				
V_A (=1/2σ^2b)		0.022				0.062		
V_E (=σ^2)		0.102				0.149		
Number of basal tillers per basal-tillered plant								
RILs	93	0.186	0.001	$\sigma^2+2\sigma^2b$	114	0.231	0.005	$\sigma^2+2\sigma^2b$
Reps	1	0.048	0.478	$\sigma^2+94\sigma^2b$	1	0.141	0.319	$\sigma^2+115\sigma^2b$
Error	93	0.095		σ^2	114	0.141		σ^2
				Estimates				
V_A (=1/2σ^2b)		0.023				0.023		
*V*E (=σ^2)		0.095				0.141		
Panicle length								
RILs	136	50.0	<0.001	$\sigma^2+2\sigma^2b$	136	31.1	<0.001	$\sigma^2+2\sigma^2b$
Reps	1	1.0	0.641	$\sigma^2+137\sigma^2b$	1	38.5	0.079	$\sigma^2+137\sigma^2b$
Error	136	4.4		σ^2	136	12.3		σ^2
				Estimates				
V_A (=1/2σ^2b)		11.4				4.7		
V_E (=σ^2)		4.4				12.3		
Panicle width								
RILs	136	48.2	<0.001	$\sigma^2+2\sigma^2b$	136	24.0	<0.001	$\sigma^2+2\sigma^2b$
Reps	1	4.1	0.393	$\sigma^2+137\sigma^2b$	1	50.8	0.033	$\sigma^2+137\sigma^2b$
Error	136	5.6		σ^2	136	10.9		σ^2
				Estimates				
V_A (=1/2σ^2b)		10.6				3.3		
V_E (=σ^2)		5.6				10.9		
Leaf angle								
RILs	136	0.764	<0.001	$\sigma^2+2\sigma^2b$	(Leaf-angle data were collected from only one of the two Lubbock replications			
Reps	1	0.131	0.441	$\sigma^2+137\sigma^2b$				
Error	136	0.220		σ^2				
				Estimates				
V_A (=1/2σ^2b)		0.136						
V_E (=σ^2)		0.220						
Maturity								
RILs	136	73.4	<0.001	$\sigma^2+2\sigma^2b$	136	134.3	<0.001	$\sigma^2+2\sigma^2b$
Reps	1	39.5	0.007	$\sigma^2+137\sigma^2b$	1	176.6	0.033	$\sigma^2+137\sigma^2b$
Error	136	5.4		σ^2	136	37.9		σ^2
				Estimates				
V_A (=1/2σ^2b)		17.0				24.1		
V_E (=σ^2)		5.4				37.9		

Table 3 (continued)

Source	College Station				Lubbock			
	df	MS	*P*	ems	*df*	MS	*P*	ems
Awn length								
RILs	78	11.1	<0.001	$\sigma^2+2\sigma^2b$	(Length-of-awn data were not collected at Lubbock)			
Reps	1	2.3	0.393	$\sigma^2+137\sigma^2b$				
Error	78	3.1		σ^2				
				Estimates				
V_A (=1/2σ^2b)		4.0						
V_E (=σ^2)		3.1						

Table 4 Probabilities associated with ANOVA regression and residual mean squares, based on one QTL, for the linkage groups for which significant evidence for a QTL was detected in either trial

Trait	Linkage group	College Station		Lubbock	
		Regression	Residual	Regression	Residual
Main-culm height	A	0.027	0.025	0.01	0.09
	C	0.023	0.043	0.19	0.051
	E	<0.001	0.70	0.01	0.41
	G	0.008	0.52	0.02	0.20
Tallest basal-tiller height	A	0.03	0.44	0.04	0.11
	E	0.01	0.17	<0.001	0.28
Number of basal tillers with heads per plant	A	<0.001	0.03	<0.001	0.051
	H	0.04	0.86	0.08	0.92
	I	<0.001	0.20	<0.001	0.24
Number of basal tillers per basal-tillered plant	A	<0.001	0.08	<0.001	0.14
	I	<0.001	0.12	<0.001	0.64
Panicle length	E	0.012	0.07	0.17	0.01
	F	0.008	0.14	<0.001	0.30
	H	0.02	0.40	0.12	0.47
Panicle width	A	0.002	0.18	0.01	0.03
	C	0.02	0.044	0.19	0.01
	G	0.002	0.25	0.18	0.05
	I	0.007	0.13	<0.001	0.18
	J	0.001	0.40	0.19	0.34
Leaf angle	A	<0.001	0.30	0.46	0.31
	E	<0.001	0.20	<0.001	0.36
	I	0.03	0.99	0.02	0.31
Maturity	F	<0.001	0.01	<0.001	0.03
	G	0.028	0.94	0.05	0.38
Awn length	I	0.03	0.09	dnr[a]	dnr

[a] dnr, Data not recorded

QTLs, *XCuh.txs-E*, was only 18.6 cM long in the CS environment and 13.1 cM long in the LBK environment. The residual mean squares probability for LG A in the CS environment, based on one QTL, was significant, and a second QTL was mapped in the LG, resulting in a non-significant residual. The regression and residual probabilities, based on one QTL, also were significant for LG C in the CS environment (*P*=0.023 and 0.043, respectively; Table 4), but only one QTL was mapped in the LG. Tallest-basal-tiller-height (TIH) QTLs were mapped in LGs A and E in both environments (Table 5, Fig. 1). Their locations are highly similar to those of *QCuh.txs-A2* and *QCuh.txs-E*, suggesting that they are the same QTLs. Evidence for a TIH QTL corresponding to *QCuh.txs-G* was not obtained, however.

The CUH QTLs explained 52% and 65.8% of the additive genetic variance (V_A) at CS and LBK, respectively (or, approximately 21 cm and 10 cm of the difference between the parental strains in main-culm height in the two environments, respectively), and the TIH QTLs explained 29.9% and 30.5% of the V_A in the two environments, respectively (Table 5).

The dwarfing-loci genotype of BTx623 is *dw1 Dw2 dw3 dw4*, and that of IS3620C is unknown. The phenotype of IS3620C is suggestive of the *dw1 Dw2 dw3 dw4* genotype, however, and thus it is unlikely that differing alleles were segregating at any of these dwarfing loci in the BTx623×IS3620C RI population. Lin et al. (1995) mapped six plant-height QTLs, including *Dw2* in LG I (LG D of their map)(see also Ulanch 1999), Klein

Table 5 QTL map locations, additive effects, and percentage additive genetic variance (V_A) explained

Trait/ QTL	Map location (cM)		Additive effect (*a*)		Percentage of V_A explained	
	College Station	Lubbock	College Station	Lubbock	College Station	Lubbock
Main-culm height						
QCuh.txs-A1	50	–[a]	↓[b] (+15.3)	–	↓	–
QCuh.txs-A2	90	116	13.5 (–17.6)	–12.0	11.0	16.2
QCuh.txs-C	150	–	+11.2[c]	–	7.6	–
QCuh.txs-E	100	96	–20.1	–18.9	24.3	40.1
QCuh.txs-G	48	50	+12.3	+9.2	9.1	9.5
		$\sum a^2$=	863.0	585.9		
		Exp $\sum a^2$ (=$2V_A$)=	1,660.8	891.0		
		% of total V_A explained=	52.0	65.8	52.0	65.8
Tallest basal-tiller height						
QTih.txs-A	116	108	–10.8	–10.6	10.2	10.8
QTih.txs-E	106	104	–15.0	–14.3	19.7	19.7
		$\sum a^2$=	341.6	316.9		
		Exp $\sum a^2$ (=$2V_A$)=	1,139.6	1040.6		
		% of total V_A explained=	29.9	30.5	29.9	30.5
Number of basal tillers with heads, per plant						
QTina.txs-A1	54	50	↓ (–0.13)	–0.120	↓	11.7
QTina.txs-A2	136	–	–0.148 (–0.08)	–	48.7	–
QTina.txs-H	60	–	–0.066	–	9.7	–
QTina.txs-I	58	62	–0.112	–0.215	27.9	37.2
		$\sum a^2$=	0.039	0.061		
		Exp $\sum a^2$ ($2V_A$)=	0.045	0.124		
		% of total V_A explained=	86.3	48.9	86.3	48.9
Number of basal-tillers per basal-tillered plant						
QTinb.txs-A	64	70	–0.121	–0.125	31.8	34.0
QTinb.txs-I1	–	42	–	–0.103	–	23.0
QTinb-txs-I2	64	–	–0.134	–	39.0	–
		$\sum a^2$=	0.033	0.026		
		Exp$\sum a^2$ (=$2V_A$)=	0.046	0.046		
		% of total V_A explained=	70.8	57.0	70.8	57.0
Panicle length						
QPal.txs-E	104	–	+1.67	–	12.2	–
QPal.txs-F	100	104	+1.57	+1.39	10.8	20.6
QPal.txs-H	78	–	+1.40	–	8.6	–
		$\sum a^2$=	7.2			
		Exp $\sum a^2$ (=$2V_A$)=	22.8	9.4		
		% of total V_A explained=	+31.6	20.6	31.6	20.6
Panicle width						
QPaw.txs.A1	28	12	–2.64	↓ (–1.22)	32.9	↓
QPaw.txs-A2	–	132	–	1.32 (–1.01)	–	26.4
QPaw.txs-C	136	–	–1.42	–	9.5	–
QPaw.txs.G	24	–	–1.77	–	14.8	–
QPaw.txs-I1	–	34	–	+1.19	–	21.5
QPaw.txs.I2	80	–	+1.55	–	11.3	–
QPaw.txs.J	42	–	+1.92	–	17.4	–
		$\sum a^2$=	18.2	3.16		
		Exp $\sum a^2$ (=$2V_A$)=	21.2	6.6		
		% of total V_A explained=	85.9	47.9	85.9	47.9
Leaf angle						
QLea.txs-A	130	–	–0.229	–	19.2	–
QLea.txs-E	92	96	–0.351	–0.471	45.3	28.4[d]
QLea.txs-I	–	24	–	+0.240	–	7.4[d]
		$\sum a^2$=	0.176	0.279		
		Exp $\sum a^2$(=$2V_A$)=	0.272	0.781[d]		
		% of total V_A explained=	64.5	35.8[d]	64.5	35.8[d]

Table 5 (continued)

Trait/ QTL	Map location (cM)		Additive effect (*a*)		Percentage of V_A explained	
	College Station	Lubbock	College Station	Lubbock	College Station	Lubbock
Maturity						
QMa.txs-F1	–	6	–	↓ (–4.54)	–	↓
QMa.txs-F2	–	70	–	3.26 (+1.73)	–	22.0
QMa.txs-G	108	116	–1.67	–2.31	8.2	11.1
		$\sum a^2$=	2.79	15.96		
		Exp $\sum a^2$ (=2V_A)=	34.0	48.2		
		% of total V_A explained=	8.2	33.1	8.2	33.1
Awn length						
QAwl.txs-I	82	na	–0.73	na[e]	6.7	na
		$\sum a^2$=	0.53			
		Exp $\sum a^2$ (=2V_A)=	8.0			
		% of total V_A explained=	6.7	na	6.7	na

[a] – Indicates that a QTL was not detected at the 5% level of significance

[b] The values below the arrowheads in the 4th through 7th columns are the 'combined additive effect' and the 'combined % additive genetic variance explained', respectively, of the QTL in the row with the arrowhead and the QTL in the row below it (see Materials and methods). The additive effects of the individual QTLs are shown in parentheses

[c] A + indicates that the BTx623 allele conditions a larger value than the IS3620C allele for the parameter analyzed and a '–' that it conditions a smaller value

[d] True leaf-angle 'Exp Σa^2' estimates could not be calculated for the Lubbock trial because leaf-angle data were collected from only one of the two replications. To obtain 'Exp Σa^2' and '% of total V_A explained' values for comparison purposes, an ANOVA was performed in which the values in the single dataset were assumed to be the means of two data sets (results not shown)

[e] na, Not applicable

et al. (2000) mapped two plant-height QTLs, including *Dw3* in LG D, and Pereira et al. (1995) and Rami et al. (1998) mapped four and three plant-height QTLs, respectively. The similar map positions of the plant-height QTLs mapped in LGs E (A) and G (H) by Pereira et al. (1995) and in LG E by Klein et al. (2001) to those mapped in these LGs in this study indicate that they may be the same QTLs.

Number of basal tillers

Similar data were obtained for the number of basal tillers with heads per plant (TINA) and the number of basal tillers per basal-tillered plant (TINB). For both, strong evidence was obtained for the presence of QTLs in LGs A and I (P <0.001, Table 4). The 95% confidence intervals for the LG I TINB QTLs detected at CS and LBK do not overlap (Fig. 1), but the interval for *QTina.txs-I*, as determined at both CS and LBK, is closely similar to that of *QTinb.txs-I2* (Table 5, Fig. 1). Likewise, the 95% confidence intervals for *QTina.txs-A1* and *QTinb.txs-A* overlap (Table 5, Fig. 1). Two TINA QTLs were mapped in LG A at CS and only one at LBK (the residual mean squares probabilities, based on one QTL, were 0.03 and 0.051 at CS and LBK, respectively; see Table 4), but the minimum residual mean squares value at LBK was at approximately 146 cM (data not shown), close to the most likely location of the QTL detected at CS, namely, 136 cM (Fig. 1). Evidence for a LG H TINA QTL also was obtained at College Station.

Two of the TINA QTLs, namely, *QTina.txs-A1* and *QTina.txs-I*, explained 65.5% of the V_A at CS and 48.9% of the V_A at LBK (Table 5). Also, all of the mapped TINA QTLs combined explained 86.3% of the V_A at CS (1.2 basal tillers out of the 1.4 difference in the number of basal tillers between the parental lines) and 48.9% of it at LBK (0.7 tillers out of the 1.5 basal-tiller difference), while the mapped TINB QTLs explained 70.8% of the V_A at CS (1.0 out of the 1.4 basal-tiller-number difference) and 57.0% of it at LBK (0.9 out of the 1.5 basal-tiller-number difference)(Table 5).

Paterson et al. (1995b) identified four genomic regions that control the number of tillers in *Sorghum halepense* (L.) Pers (johnsongrass, 2n=2x=40), a perennial that is thought to be an interspecific hybrid derived from *S. bicolor* and *S. propinquum* (Kunth.) Hitchc. (2n=2x=20). One of these, located in LG I (D), partially overlaps the 95% confidence interval for *QTinb.txs-I1* (Table 5 and Fig. 1), and thus may be orthologous with it.

Panicle length and width

Three panicle-length (PAL) QTLs were detected (Tables 4 and 5, Fig. 1), but evidence was obtained in both environments and with P values ≤0.01 only for *QPal.txs-F*.

The other two QTLs, detected in the CS environment and with P=0.012 and 0.02, were mapped in LGs E and H, respectively. *QPal.txs-F* explained 20.6% of the V_A in the LBK environment, and the three QTLs detected at CS explained 31.6% of the V_A in that environment (Table 5). Pereira et al. (1995) and Rami et al. (1998) mapped panicle-length QTLs in five and six LGs, respectively. On the basis of the map positions of these QTLs, only one of them is a candidate for being the same as a PAL QTL mapped in this study, namely, the one mapped by Pereira et al. (1995) in LG E (A) may be the same as *QPal.txs-E*.

Seven panicle-width (PAW) QTLs were mapped in LGs A, C, G, I and J, (Tables 4 and 5, Fig. 1), six with $P \leq 0.01$, but only one locus, *QPaw.txs-A1*, was mapped in both environments, and the most probable map location for the locus differed by 16 cM in the two environments. [The residual mean squares probability for LG C in the CS environment, based on one QTL, was 0.044 (Table 4), but it was not possible to map a second PAW QTL in the LG.] The five PAW QTLs mapped at CS explained 85.9% of the V_A in that environment (Table 5), or 5.1 cm of the 6.0 cm difference in PAW between the parental lines (Table 2), and the three mapped at LBK explained 47.9% of the V_A in that environment, or 2.3 cm of the 4.9 cm difference between the parental lines.

Leaf angle

The ANOVA regression probability for leaf angle (LEA, the angle of the third leaf-blade junction relative to the main culm), based on one QTL, was significant for LG A at CS and for LGs E and I in both environments, and the residual probability was not significant in either environment for any LG (Table 4). A LEA QTL was not mapped in LG I at CS, however, because there was a nonsignificant marker-means association in the LG in the CS environment. Thus, only *QLea.txs-E* was mapped in both environments (Table 5, Fig. 1). It explained 45.3% of the V_A in the CS environment, or about 12° of the approximately 27° difference in LEA between BTx623 and IS3620 C (Table 2), and the 95% confidence interval for the QTL was only 18.7 cM long. The other LEA QTL detected at CS, *QLea.txs-A*, explained an additional 19.2% of the V_A, or an additional 5° of the approximately 27° difference in LEA between the parental lines.

Leaf-angle data were collected for only one of the two Lubbock replications, thus true LEA Exp Σa^2 and percentage of total V_A explained values could not be calculated for the Lubbock trial. Calculated as described in footnote d of Table 5, *QLea.txs-E* explained 28.4% of the V_A in the LBK environment, and the 95% confidence interval for the QTL was only 14.4 cM long.

Maturity

Three maturity (MA) loci were mapped, one in LG G in both environments and two in LG F in the LBK environment only (Tables 4 and 5, Fig. 1). [The regression probability for LG F, based on one QTL, was significant in the CS environment (Table 4), but as there was a nonsignificant marker means association in the LG, a MA QTL was not mapped in the LG.]. The LG G QTL explained 8.2% of the V_A at CS and 11.1% of it at LBK, while the two LG F QTLs explained 22.0% of the V_A at LBK (Table 5).

Both BTx623 and IS3620C are known to be recessive at the *Ma1* locus, but their genotypes at the other major maturity loci are unknown. *Ma1* is located in LG I [Lin et al. 1995 (D); Ulanch 1999], *Ma3* in LG A (Childs et al. 1997), and *Ma4* in LG G, near *Xtxs1163* in the upper part of the LG (K.L. Childs and J.E. Mullet, personal communication). Other loci controlling maturity were mapped in LGs B and F (G) by Lin et al. (1995) and LGs B (G) and G (B) by Crasta et al. (1999). None of these loci occupy map positions similar to those of the maturity loci mapped in this study.

Awn length

A QTL controlling the length of the awns of awned plants was detected with P=0.03 (Table 4) at the bottom of LG I (Fig. 1). It controlled only 6.7% of the V_A (Table 5), indicating that the genetic control of awn length of awned plants in the BTx623×IS3620C RIL population is by numerous loci that individually have small effects.

Mapping of *Awn* and *D*

A gene that controls the presence versus the absence of awns, designated *Awn*, was mapped at a LOD score of 5.1 near the top of LG C, 7.7 cM from *Xisu52*, the terminal marker (Fig. 1). The genetic control of awning is known to vary as a function of the strains being analyzed (for review, see Rooney 2000), and whether or not *Awn* is among the genes controlling awning that were characterized by Sieglinger et al. (1934) and Kullaiswamy and Goud (1983) is unknown. *Awn* is probably the same as *AW*, however, a gene controlling the presence versus the absence of awns that was mapped by Tao et al. (1998, 2000) in LG C (A) at a position that, within the limits of resolution of their more than 435-cM-long map of the LG, approximates that of *Awn*. *D*, a gene that controls dry versus juicy midribs, was mapped at a LOD score of 5.2 near the middle of LG I, co-segregating with *Xtxp97* [which is immediately adjacent to *Xumc34* (Bhattramakki et al. 2000)] and 5.2 cM from *Xtxs1030*, confirming the location reported for the gene by Xu et al. (1998).

Concluding remarks

Thirty-one named QTLs were mapped. *QCuh.txs-A2* may be the same as *QTih.txs-A*, *QCuh.txs-E* may be the

same as *QTih.txs-E*, *QTina.txs-A1* may be the same as *QTinb.txs-A*, and *QTina.txs-I* may be the same as *QTinb.txs-I2*, however based on their control of variation in similar characters and their similar map locations. Thus, a minimum of 27 unique QTLs were mapped. Also, two of the aforementioned pairs of QTLs, namely, *QCuh.txs-E* and *QTih.txs-E* and *QTina.txs-I* and *QTinb.txs-I2*, and two other QTLs, *QCuh.txs-G* and *QPal.txs-E*, occupy map locations similar to those of QTLs controlling these characters that were mapped previously by others. Consequently, a minimum of 23 previously unreported QTLs were mapped in this study.

With the exception of three characters, [MA in the CS environment, PAL at LBK, and AWL at CS (the only environment in which data was recorded for AWL)], the mapped QTLs explained a minimum of 30% of the V_A for the characters in both environments. The range of % V_A explained was from 86.3% for TINA (in the CS environment) to only 6.7% for AWL.

QTLs that control the number of basal tillers were among the most significant of the loci that were mapped. BTx623, one of the parents of the mapping population, lacks basal tillers; IS3620C, the other parent, produces them, and two QTLs that explained 65.5% of the TINA V_A at CS and 48.9% of it at LBK were mapped in LGs A and I with $P<0.001$ in both environments. Similar findings were obtained for TINB, the other tiller-number parameter that was analyzed. These and the other mapped QTLs that displayed little genotype-environment interaction should be readily amenable to marker-assisted selection.

Mapping QTLs that control variation in traits of agronomic importance is a key part of the process of using molecular markers in plant improvement and in elucidating the manner in which the loci control variation in the traits. The results obtained in this study should facilitate marker-assisted selection for sorghum improvement and future studies of the functional genomics of the QTLs.

Acknowledgments We thank Dr. C. Du for performing the QTL CARTOGRAPHER analyses and Ms. Monica Menz and Drs. Robert Klein and William Rooney for comments on the manuscript This research was supported by the Texas Agricultural Experiment Station, the USDA Agricultural Research Service, and a grant to G.E. Hart and K.F. Schertz from the United States Department of Agriculture National Research Initiative Competitive Research Grants Program.

References

Basten CJ, Weir BS, Zeng Z-B (1994) Zmap-a QTL cartographer. In: Smith C, Gavora JS, Benkel B, Chesnais J, Fairfull W, Gibson JP, Kennedy BW, Burnside EB (eds) Computing strategies and software. Organizing committee, Proc 5th World Cong Genet Appl Livestock Product, vol 22. Guelph, Ont., Canada, pp 65–66

Basten CJ, Weir BS, Zeng Z-B (1997) QTL CARTOGRAPHER: a reference manual and tutorial for QTL mapping. Department of Statistics, North Carolina State University, Raleigh, N.C.

Bhattramakki D, Dong J, Chhabra AK, Hart GE (2000) An integrated SSR and RFLP linkage map of *Sorghum bicolor* (L.) Moench. Genome 43:988–1002

Childs KL, Miller FR, Cordonnier-Pratt M-M, Pratt LH, Morgan PW, Mullet JE (1997) The sorghum photoperiod sensitivity gene, Ma_3, encodes a phytochrome B. Plant Physiol 113:611–619

Crasta OR, Xu W, Rosenow DT, Mullet JE, Nguyen HT (1999) Mapping of post-flowering drought resistance traits in grain sorghum: association of QTLs influencing premature senescence and maturity. Mol Gen Genet 262:579–588

Doggett H (1988) Sorghum, 2nd edn. John Wiley & Sons, New York

Hadley HH (1957) An analysis of variation in height in sorghum. Agron J 49:144–147

Kearsey MJ, Hyne V (1994) QTL analysis: a simple 'marker regression' approach. Theor Appl Genet 89:698–702

Kearsey MJ, Pooni HS (1996) The genetical analysis of quantitative traits. Chapman and Hall, London

Klein PE, Klein RR, Cartinhour SW, Ulanch PE, Dong J, Obert JA, Morishige DT, Schlueter SD, Childs KL, Ale M, Mullet JE (2000) A high-throughput AFLP-based method for constructing integrated genetic and physical maps: progress toward a sorghum genome map. Genome Res 10:789–807

Klein RR, Rodriguez-Herrera R, Schlueter JA, Klein PE, Yu ZH, Rooney WL (2001) Identification of genomic regions that affect grain mold incidence and other traits of agronomic importance in sorghum. Theor Appl Genet 102:307–319

Kong L, Dong J, Hart GE (2000) Characteristics, linkage-map positions, and allelic differentiation of *Sorghum bicolor* (L.) Moench DNA simple-sequence repeats (SSRs). Theor Appl Genet 101:438–448

Kosambi DD (1944) The estimation of map distances from recombination values. Ann Eugen 12:172–175

Kullaiswamy BY, Gould JV (1993) New genes for awning in Sorghum (*Sorghum bicolor* (L.) Moench). Madras Agric J 70: 355–359

Lin Y-R, Schertz KF, Paterson AH (1995) Comparative analysis of QTLs affecting plant height and maturity across the Poaceae, in reference to an interspecific sorghum population. Genetics 141:391–411

Paterson AH, Lin Y-R, Li Z, Schertz KF, Doebley JF, Pinson SRM, Liu S-C, Stansel JW, Irvine JE (1995a) Convergent domestication of cereal crops by independent mutations at corresponding genetic loci. Science 269:1714–1718

Paterson AH, Schertz KG, Lin Y-A, Liu S-C, Chang Y-L (1995b) The weediness of wild plants: molecular analysis of genes influencing dispersal and persistence of johnsongrass, *Sorghum halepense* (L.) Pers. Proc Natl Acad Sci, USA 92:6127–6131

Peng Y, Schertz KF, Cartinhour S, Hart GE (1999) Comparative genome mapping of *Sorghum bicolor* (L.) Moench using a RFLP map constructed in a population of recombinant inbred lines. Plant Breed 118:225–235

Pereira MG, Lee M (1995) Identification of genomic regions affecting plant height in sorghum and maize. Theor Appl Genet 90:380–388

Pereira MG, Ahnert D, Lee M, Klier K (1995) Genetic mapping of quantitative trait loci for panicle characteristics and seed weight in sorghum. Rev Brasil Genet 18:249–257

Quinby JR (1966) Fourth maturity gene locus in sorghum. Crop Sci 6:516

Quinby JR, Karper RE (1945) The inheritance of three genes that influence time of floral initiation and maturity date in Milo. J Am Soc Agron 37:916

Quinby JR, & Karper RE (1954) Inheritance of height in sorghum. Agron J 46:211–216

Rami J-F, Dufour P, Trouche G, Fliedel G, Mestres C, Davrieux F, Blanchard P, Hamon P (1998) Quantitative trait loci for grain quality, productivity, morphological and agronomical traits in sorghum (*Sorghum bicolor* L. Moench). Theor Appl Genet 97:605–616

Rooney WL (2000) Genetics and cytogenetics. In: Smith CW, Frederiksen RA (eds) Sorghum: origin, history, technology, and production. John Wiley and Sons, New York, pp 261–308

Rooney WL, Aydin S (1999) The genetic control of photoperiod sensitive response in sorghum. Crop Sci 39:397–400

Sieglinger JB, Swanson AF, Martin JH (1934) Inheritance of awn development in sorghum. J Agric Res 49:663

Tao YZ, Jordan DR, Henzell RG, McIntyre CL (1998) Construction of a genetic map in a sorghum RIL population using probes from different sources and its comparison with other sorghum maps. Aust J Agric Sci 49:729–736

Tao YZ, Henzell RG, Jordan DR, Butler DG, Kelly AM, McIntyre CL (2000) Identification of genomic regions associated with stay green in sorghum by testing RILs in multiple environments. Theor Appl Genet 100:1225–1232

Tuinstra MR, Grote EM, Goldsbrough PB, Ejeta G (1996) Identification of quantitative trait loci associated with preflowering drought tolerance in sorghum. Crop Sci 36:1337–1344

Tuinstra MR, Grote EM, Goldsbrough PB, Ejeta G (1997) Genetic analysis of post-flowering drought tolerance and components of grain development in *Sorghum bicolor* (L.) Moench. Mol Breed 3:439–448

Ulanch PE (1999) Steps towards map-based cloning Ma_1, a gene controlling photoperiod sensitivity in sorghum. PhD thesis, Texas A&M University, College Station, Tex.

Xu W, Subudhi PK, Crasta OR, Rosenow DT, Mullet JE, Nguyen HT (2000) Molecular mapping of QTLs conferring stay green in grain sorghum. Genome 43:461–469