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Registration of 17 Upland (*Gossypium hirsutum*) Cotton Germplasm Lines Disomic for Different *G. barbadense* Chromosome or Arm Substitutions

Seventeen germplasm lines (Reg. no. GP-836 through GP-852, PI 636346 to PI 636362) were each developed by hypoaneuploid-based backcross substitution of a different *Gossypium barbadense* L. chromosome or chromosome segment into a *G. hirsutum* L. genetic background by the Texas Agricultural Experiment Station, the USDA-ARS, and the Mississippi Agricultural and Forestry Experiment Station and released in 2004. The substitution lines are genetically similar to TM-1, an Upland cotton (*G. hirsutum*) genetic standard, and to each other, except that each line differs by the replacement of a specific homologous pair of chromosomes or chromosome segments from the donor line 3–79, a *G. barbadense* genetic standard. TM-1 is an inbred line extracted from the commercial cultivar Deltapine 14 and has been maintained over 40 generations by self-pollination (Kohel et al., 2001). Line 3–79 originated as a doubled-haploid from *G. barbadense* (Endrizzi et al., 1985). The high fiber quality and genetic uniformity of 3–79 and the availability of hypoaneuploid stocks in TM-1 background influenced the choice of parental lines.

These germplasm lines were released because attempts to incorporate genes from *G. barbadense* for exceptional fiber length, strength, and fineness into Upland cotton have generally not achieved stable introgression. Poor agronomic qualities of most progeny, distorted segregation, sterility, and limited recombination due to incompatibility between the genomes have been associated with previous attempts to incorporate genes from *G. barbadense*. By hypoaneuploid-based backcross chromosome substitution, we have replaced individual TM-1 chromosome pairs with their respective 3–79 chromosome pairs and largely reconstituted the other *G. hirsutum* chromosomes (Table 1). The lines are designated as CS–B lines to reflect their origin through chromosome substitution from a *G. barbadense* donor. Collectively, this set of substitutions accounts for about half of the 26 chromosomes in the cotton genome, and thus offers breeders facile access to the respective portion of the *G. barbadense* 3–79 genome. At the BC_5F_1 generation, nonsubstituted chromosomes or arms would be expected under random recovery to contain only about 1.6% alien sequences, and after extraction of a euploid self-progeny, only about 0.8%. When one of these CS-B lines is crossed with *G. hirsutum*, the entire substituted *G. barbadense* chromosome or arm is expected to pair and recombine with the same chromosome or arm from *G. hirsutum*. The strong differential representation between substituted (1.0) and non-substituted chromosomes (0.008) offers a number of additional analytical and genetic advantages.

The development of the CS-B lines was based on principles of cotton cytogenetic behavior, transmission, and inheritance (Endrizzi et al., 1985). Key among these is that whereas transmission of hypoaneuploidy through the ovule parent is common (up to 50%), transmission through pollen is totally lacking or rare for all whole-chromosomes and most large-segment deletions, including those associated with telosomy. The backcrossing process used in cotton is facilitated by this differential transmission between mega- versus microgametophytes.

Development of each *G. barbadense* chromosome substitution line involved three stages: (i) development of a TM-1–like hypoaneuploid stock that served as the TM-1–like recurrent parent (Table 1), (ii) selective introgression of a *G. barbadense* chromosome or segment per line to create a monosomic substitution stock, and (iii) recovery of the euploid, disomic substitution line, by inbreeding. Monosomic and monotelodisomic cytogenetic stocks nearly isogenic to TM-1 and each other were created by intraspecific hybridization and backcrossing the respective cytologically identified aneuploids at each generation to TM-1, up through the BC_5 generation or higher, before making an interspecific cross. Development of the hemizygous monosomic substitutions was initiated by crossing each type of TM-1–like hypoaneuploid as the seed parent with the line 3–79 as pollen parent. Whereas euploid *G. hirsutum* × *G. barbadense* F_1 and BC_nF_1 hybrids typically form 26 ring bivalents (II) at metaphase I, monosomic and monotelodisomic hybrids form diagnostic metaphase I meiotic configurations that include a univalent (I) or a rod bivalent (Ii), 25 II + I or 25 II + Ii, respectively. For each line, an interspecific F_1

Table 1. Interspecific chromosome substitution lines of Upland cotton in TM-1 *G. hirsutum* background containing a *G. barbadense* chromosome or segment.

Designation of released line	Recurrent parent hypoaneuploid: monosomic (H) or monotelodisomic (Te)				Chromosome or segment substituted from *G. barbadense* 3–79†	Germplasm registration number	Accession
	Designation	Chromosome	Short arm	Long arm			
CS-B01	H01	1	hemizygous	hemizygous	1	GP-836	PI 636346
CS-B02	H02	2	hemizygous	hemizygous	2	GP-837	PI 636347
CS-B04	H04	4	hemizygous	hemizygous	4	GP-838	PI 636348
CS-B05sh	Te05Lo	5	hemizygous	disomic	5, short arm	GP-845	PI 636355
CS-B06	H06	6	hemizygous	hemizygous	6	GP-839	PI 636349
CS-B07	H07	7	hemizygous	hemizygous	7	GP-840	PI 636350
CS-B11sh	Te11Lo	11	hemizygous	disomic	11, short arm	GP-846	PI 636356
CS-B12sh	Te12Lo	12	hemizygous	disomic	12, short arm	GP-847	PI 636357
CS-B14sh	Te14Lo	14	hemizygous	disomic	14, short arm	GP-848	PI 636358
CS-B15sh	Te15Lo	15	hemizygous	disomic	15, short arm	GP-849	PI 636359
CS-B16	H16	16	hemizygous	hemizygous	16	GP-841	PI 636351
CS-B17	H17	17	hemizygous	hemizygous	17	GP-842	PI 636352
CS-B18‡	H18	18	hemizygous	hemizygous	18	GP-843	PI 636353
CS-B22Lo	Te22sh	22	disomic	hemizygous	22, long arm	GP-850	PI 636360
CS-B22sh	Te22Lo	22	hemizygous	disomic	22, short arm	GP-851	PI 636361
CS-B25	H25	25	hemizygous	hemizygous	25	GP-844	PI 636354
CS-B26Lo	Te26sh	26	disomic	hemizygous	26, long arm	GP-852	PI 636362

† Caution should be exercised when drawing conclusions about positions of the centromere, genes, and molecular markers based on BC_5S_1 chromosome arm substitution analysis. Users should recognize that while it is likely that the centromere is close to the respective junction between *G. hirsutum* and *G. barbadense* chromatin, the exact position of the junction is constrained by acrocentricity of the telosome, but ultimately dependent on the point of crossing-over closest to the acrocentric telomere. That site is unknown at this time. Users are thus encouraged to share and post data that will help define the positions of these junctions on integrated chromosome maps.

‡ Homozygous for *open-bud* mutant conditioned by a double recessive gene with one gene in each genome.

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plant deficient for the *G. hirsutum* chromosome or chromosome arm was selected based on plant morphological features and metaphase I analysis of microsporocytes. The interspecific F_1 hypoaneuploid plants, each hemizygous only for a specific *G. barbadense* chromosome or segment, were then backcrossed as pollen parents onto the respective types of TM-1–like aneuploid plants. As in the interspecific F_1 generation, the aneuploid BC_1F_1 plants were selected based on phenotype and metaphase I analysis to confirm that each contained the *G. barbadense* chromosome or telosome of interest. The process was repeated through the BC_5F_1 generation at which point efforts were undertaken to recover a corresponding euploid disomic substitution. Extraction of each homozygous disomic substitution line was initiated by self-pollination of a monosomic BC_5F_1 plant hemizygous for the desired *G. barbadense* chromosome or telosome. A euploid BC_5S_1 26II plant was selected on the basis of phenotype and cytological analysis of the metaphase microspores, and selfed to establish the corresponding euploid BC_5S_1 line. Lines were then increased and evaluated as sources of beneficial alleles governing multigenic traits, including fiber properties (Saha et al., 2004a, 2004b).

These CS-B lines are useful as new sources of genetic variation for improvement of Upland cotton through breeding and fundamental genetic research. Evaluation of the CS-B germplasm in replicated trials across years revealed that genetic variation for all measured traits was highly significant among the CS-B lines and corresponding TM-1 × CS-B line F_2 families (Saha et al., 2004a, 2004b). For some traits, the phenotypic range of CS-B lines exceeds the parental range, and some CS-B lines are superior to the recurrent parent TM-1, indicating their prospective value as breeding parents. Bolls of all CS-B lines were intermediate in size to those of the parents, 3–79 (3.6 g boll^{-1}) and TM-1 (5.9 g boll^{-1}), except for CS-B06 and CS-B12sh (about 6 g boll^{-1}). Lint percentages among CS-B lines ranged from lower (29.7) to higher (36.8), and were higher for CS-B05sh, CS-B16, CS-B22Lo, and CS-B22sh than the parents, TM-1 (31.8) and 3–79 (34.2). CS-B25 had reduced micronaire readings (3.7) and increased fiber strength (218 kN m kg^{-1}) relative to TM-1 (4.7, 196 kN m kg^{-1}), without marked yield reduction, 2358 vs. 2519 kg ha^{-1} (Saha et al., 2004b). Fiber lengths (2.5% span length) of CS-B14sh (30.4 mm), CS-B15sh (30.0 mm), and CS-B25 (30.4 mm) were longer than those of TM-1 (29.4 mm), but were shorter than that of 3–79 (34.0 mm). Seedcotton yield was not significantly different in CS-B06 (2584 kg ha^{-1}) and CS-B15sh (2622 kg ha^{-1}) than TM-1 (2519 kg ha^{-1}), but higher in all CS-B lines (1429 to 2622 kg ha^{-1}) than 3–79 (540 kg ha^{-1}). Whereas lint yields of the donor line 3–79 were less than half that of TM-1, and lint yields of most CS-B lines are also significantly lower than that of TM-1, the lint yields of CS-B01, CS-B05sh, and CS-B12sh were not significantly different from TM-1, which could indicate these CS-B lines lack yield-affecting loci from 3–79, or that the collective effects of multiple 3–79 loci are offsetting each other with respect to this trait. Additional trials (unpublished data, 2004) compared CS-B01, CS-B11sh, CS-B12sh, and CS-B26Lo to the parents and other checks. CS-B01 was similar to TM-1 in terms of most fiber yield and property characteristics, but CS-B01 had smaller bolls (4.9 g boll^{-1} vs. 6.0 g boll^{-1}) and nonsignificantly lower micronaire readings (3.95 vs. 4.75). Both CS-B01 and CS-B26Lo had unexpectedly high elongation (10.8%), departing markedly from TM-1 (8.8%) and 3–79 (8.8%), indicating possible transgressive segregation. Bolls of CS-B11sh were not significantly different in size from TM-1, yet had significantly lower lint percent (29.1 versus 34.7) and yielded less seedcotton (66%) and lint (59%). CS-B26Lo had only 80% the boll size of TM-1 and contained only about 40% as much seedcotton and lint. Chromosome-specific QTL analysis of these lines could reveal markers useful for indirect selection against undesirable genes when conducting wide cross introgression into Upland cotton from 3–79 and perhaps other *G. barbadense* germplasm. Conversely, these marker loci or linked ones might be used to introgress beneficial Upland alleles into elite *G. barbadense* germplasm. Relative to TM-1, CS-B12sh was similar in terms of most fiber yield and property characteristics but yielded nonsignificantly more seedcotton and lint, with fiber quality slightly better than or similar to TM-1 fiber. Comparisons of trait means of CS-B lines versus their (TM-1 × CS-B line) F_2 families revealed pairwise incongruity in the direction of departures among the CS-B lines for certain traits, which is congruent with the idea that genetic differences among the CS-B lines differentially lead to epistatic effects in the F_2s (Saha et al., 2004a).

Given a rough estimate of at least 30 000 genes in the cotton genome, we can estimate each of the monosome-derived CS-B lines carries an average of 1000 or more 3–79 genes, some of which may differ significantly from the respective TM-1 alleles. In a minority of instances, the difference might offer a chance for genetic improvement of domesticated Upland cotton. According to random probability, we expect about five-sixths of the differences between CS-B lines and TM-1 to be due to the substituted chromosome or segment, and the remaining sixth due to remnant 3–79 genes in nonhomologous chromosomes that was inadvertently retained during backcrossing and inbreeding. Thus, it was not unexpected that a few BC_5S_1 families segregated for obvious plant traits petal color or pubescence. Their presence should not detract from the value of this source of *G. barbadense* germplasm for breeding purposes.

These germplasm lines were developed, evaluated and released by D.M. Stelly and W. Raska of Texas A&M University, S. Saha, J.N. Jenkins, J.C. McCarty, and O.A. Gutierrez of the USDA-ARS, Mississippi State, MS, in a collaborative research program.

A limited amount of BC_5–derived self-pollinated seeds are available for distribution to cotton geneticist, breeders, and other research personnel on written request to Dr. D. Stelly, Dep. Soil & Crop Sciences, Texas A&M University, College Station, TX 77843–2474 USA or Dr. S. Saha, USDA-ARS, P.O. Box 5367, Mississippi State, MS 39762, USA. Genetic materials of this release will be deposited in the National Plant Germplasm System where these materials will be available for research purposes, including development and commercialization of new materials. It is requested that appropriate recognition of the source be given when these germplasm lines contribute to research or the development of improved line, cultivar, or hybrid.

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References

Endrizzi, J.E., E.L. Turcotte, and R.J. Kohel. 1985. Genetics, cytology, and evolution of *Gossypium*. Adv. Genet. 23:271–375.

Kohel, R.J., J. Yu, Y. Park, and G.R. Lazo. 2001. Molecular mapping and characterization of traits controlling fiber quality in cotton. Euphytica 121:163–172.

Saha, S., D.A. Raska, J.N. Jenkins, J.C. McCarty, O. Gutierrez, R.G. Percy, R.G. Cantrell, J. Wu, J. Zhu, and D.M. Stelly. 2004a. Effect of chromosome on important quantitative traits of agronomic and fiber traits using chromosome specific recombinant lines of *G. hirsutum*. p. 169–174. *In* A. Swanepoel (ed.) Proc. World Cotton

Res. Conf.-3 9–13 March 2003. Cape Town, South Africa. Agricultural Research Council– Institute for Industrial Crops, Rustenberg, SA.

Saha, S., J. Wu, J.N. Jenkins, J.C. McCarty, Jr., O.A. Gutierrez, D.M. Stelly, R.G. Percy, and D.A. Raska. 2004b. Effect of chromosome substitutions from *Gossypium barbadense* L. 3–79 into *G. hirsutum* L. TM-1 on agronomic and fiber traits. J. Cotton Sci. 8:162–169.

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