BREEDING AND GENETICS

Combining Ability for Neps, Seed Coat Fragments, and Motes in Upland Cotton

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ABSTRACT

Minimizing neppiness, neps, seed coat fragments, and motes in ginned cotton (*Gossypium hirsutum* L.) fibers is one of the keys to keep US fibers competitive in the global market. Forty-eight F_2 hybrids derived from crosses between 12 exotic germplasm lines (male parents), i.e., six Species Polycross (SP) and six John Cotton lines, and four elite germplasm lines (female parents) were evaluated to detect combining ability for the neppiness traits. F_2 hybrids and parents were planted at two locations with four and three replicates each in 2008 and 2009. Neps and seed coat fragments were measured using Advanced Fiber Information System. Significant general combining ability (GCA) effects were observed for most neppiness traits. Additive effects were more important than nonadditive effects for neps, seed coat neps (SCN), and motes. Correlation ($r = 0.52$) between increased nep count and increased short fiber content (SFCn) implied a possibility for simultaneous improvement of these two traits. In contrast, correlation ($r = -0.50$) between nep count and fineness implied an antagonistic relationship between the two properties in selection. MD15 was the best female parent with negative GCA effects for neppiness traits and SFCn. SP156 was the best general combiner with favorable GCA effects for neppiness traits and other fiber properties with positive GCA effect for lint yield. Results provide evidence for the possibility of simultaneous genetic improvement of multiple neppiness traits with lint yield and other fiber properties in Upland cotton using SP156 as parent.

The world demand for cotton imports has been up and down with an overall increasing demand trend in recent years. World cotton imports peaked at 39 million bales in 2007, a 34% increase since 2001 (USDA-FAS, 2008). After a sharp decline in 2008, world cotton imports recovered to 36 million bales in 2010-2011, and were estimated to be 39 million bales in 2011-2012 according to a survey by Cotton Incorporated (2011). The US textile industry consumed only 28% of US-produced cotton; a majority of the produced fibers were exported in 2009-2010 (USDA-ERS, 2011). With a strong demand for raw fibers in the global market and sharp declines in the domestic demand, it is expected that the US cotton market will continue to shift from a domestic market to an export market. Fiber quality will be a crucial factor for continuing success in the export of US cotton. To maintain competitiveness in the global market, it is imperative for the US cotton industry to produce stronger and cleaner fiber with less short fiber content and fewer neps. According to Michael Watson, Vice President of Fiber Quality Research at Cotton Incorporated, "Quality continues to be the key for keeping U.S. cotton competitive in a global market and anything the industry can do to develop better varieties and better production techniques that reduce broken fibers, minimize neps, and avoid contamination will increase potential for market share" (Smith, 2010). The requirement for genetic improvement of fiber quality might be imminent in the mid-south region of US because high yielding cultivars in this region traditionally lack high fiber quality.

A fiber nep is defined as "a tightly tangled knot-like mass of unorganized fibers" (ASTM, 1999). Neps are formed during fiber development, harvesting, ginning, and cleaning; especially if immature fibers or broken fibers are created during ginning and cleaning (Hebert et al., 1988; Mangialardi, 1986). Neps adversely affect textile processing during spinning by lowering strength and uniformity of yarn and creating "ends down in spinning" (van der Sluijs and Hunter, 1999). Neps in raw fibers are the main source of yarn neps (Frydrych et al., 2001) that are associated with poor dyeing in finished dyed yarns and fabrics (Altintas et al., 2007; Jacobsen et al., 2001). A seed coat fragment is a portion of a broken seed with or without fibers attached (ASTM, 1985).

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Seed coat fragments remaining in ginned fibers affect yarn regularity and strength, and could also cause dyeing difficulties and ultimately affect the quality of the finished fabrics (Krifa and Gourlot, 2001; Pearson, 1955). Seed coat neps (SCN) are the seed coat fragments attached to lint and are difficult to remove during cleaning and carding (Baldwin et al., 1995; Krifa and Frydrych, 2002). The number of seed coat fragments after opening in the Advanced Fiber Information System (AFIS) is measured as SCN. A mote is defined as "a whole immature cotton seed" (ASTM, 1985) that could be derived from an aborted ovule or an unfertilized seed. According to Davidonis et al. (2000), unfertilized ovules or embryos that ceased to grow shortly after fertilization produce small motes, i.e., 1 to 30 mg, whereas other ovules with embryos terminated in later stages after fertilization produce large motes, i.e., 35 to 60 mg. These motes usually have thin walls and immature fibers and easily form seed coat neps, and thus are difficult to remove during cleaning. Varied correlation coefficients between number of motes per gram lint and number of seed coat neps per gram lint, $r = 0.02$ to 0.61, were detected in interspecific hybrids of *Gossypium hirsutum* L. and *G. barbadense* L. (Saranga et al., 1997). Differences in neps, SCN, and mote frequency were detected among cotton cultivars previously (Bolek et al., 2007; Boykin, 2008; Novick et al., 1991). Significant genotypic variations in neps and SCN were identified in an introgressed cotton population derived from multiple crosses between *G. hirsutum* and *G. barbadense* (Zeng and Meredith, 2010).

Determination of interrelationships among lint yield and fiber properties is essential for simultaneous improvement of agronomic performance and fiber quality. It is known that lint yield is negatively correlated with strength and fiber length (Meredith and Bridge, 1971; Miller and Rawlings, 1967). In an analysis of an exotic germplasm population, John Cotton (JC) germplasm, derived from multiple crosses between *G. hirsutum* and *G. barbadense*, lint yield was negatively correlated with nep count ($r = -0.47$) and SCN count ($r = -0.31$) (Zeng and Meredith, 2010). The negative coefficients in these traits in that study implied a possibility for simultaneous improvement of lint yield and neps in selection. Conversely, the unfavorable correlation between nep count and fineness ($r = -0.75$) (Zeng and Meredith, 2010) implied an antagonistic relationship between these two traits in selection. The objectives of this study were to detect combining ability for neps, SCN, and motes in hybrids and identify good combiners in cotton germplasm for these traits. Determination of combining ability for neppiness traits in germplasm resources can help predict the potential of genetic improvement for these important fiber properties in cotton cultivars. Use of parents having good combiners for lint yield and the neppiness traits and other fiber properties can simultaneously improve neppiness traits with yield traits and other fiber properties and promote utilization of the exotic germplasm resources.

MATERIALS AND METHODS

Crosses were made in the summer 2007 between four elite germplasm lines: Fibermax 832 (FM832, PVP 9800258), TAM 98D-99ne (Thaxton et al., 2005), MD52ne (Meredith, 2005), and MD15 (Meredith, 2006), as female parents and 12 exotic germplasm lines: SP156 (PI 6545087), SP164, SP192, SP205 (PI 654090), SP224, SP235, JC14 (PI 658308), JC32 (PI 658309), JC60 (PI 658310), JC65 (PI 658311), JC84, and JC186, as male parents in a North Carolina Design II. The male parents, SP156, SP164, SP192, SP205, SP224, and SP235, were selected from Species Polycross (SP) derived from multiple crosses among five *Gossypium* tetraploid species as described previously (Zeng et al., 2007). The male parents, JC14, JC32, JC60, JC65, JC84, and JC186, were selected from John Cotton germplasm (JC) derived from multiple crosses between *G. hirsutum* and *G. barbadense* L. (Zeng and Meredith, 2009). The F_1 hybrids were grown and self-pollinated at a winter nursery at Colima, Mexico during 2007 and 2008.

The 48 F_2 hybrids and the 16 parents were evaluated at two locations at the Delta Research Center, Stoneville, MS during 2008 and 2009 with four replicates at each site in 2008 and three replicates at each site in 2009. The experiments were conducted in a randomized complete block design. Plants were grown in single-row plots 12.2 m × 1.0 m. Environments within row spacing at site 1 and site 2 were distinguished by soil types and planting dates. Soil type of field site 1 was Beulah fine sandy loam (a coarse-loamy, mixed active thermic Typic Dystrochrepts) and soil type of field site 2 was Bosket fine sandy loam (a fine-loamy, active, thermic Mollic Hapludalf). Planting dates were 21 April 2008 at site 1, 30 April 2008 at site 2, 22 April

2009 at site 1, and 23 April 2009 at site 2. Standard conventional field practices were applied during all experiments. The factor of environment was considered as a replacement for factors of year and location for purpose of statistical analysis. The first environment, Environment 1, was a replacement for site 1 in 2008; the second environment, Environment 2, was a replacement for site 2 in 2008; the third environment, Environment 3, was a replacement for site 1 in 2009; and the fourth environment, Environment 4, was a replacement for site 2 in 2009.

Fifty bolls from each plot were picked randomly by hand. The boll sample from each plot was ginned using a laboratory saw gin to measure lint percentage, which was calculated as lint weight from the boll sample divided by the lint weight plus seed weight of the boll sample. The remaining bolls from each plot were harvested by a mechanical picker for yield measurements. Total seed cotton weight of each plot was the sum of seed cotton weight of the sampled bolls and the remaining bolls in that plot. Lint yield of each plot was calculated from seed cotton weight per plot and lint percentage of that plot.

Fiber properties of nep size (μm), nep count (cnt g^{-1}, counts per gram of ginned fibers), seed coat nep (SCN) size (μm), and SCN count (cnt g^{-1}, counts per gram of ginned fibers) were measured using Uster AFIS (Uster AFIS, 1997). Other simultaneously measured fiber properties in AFIS included upper-quartile length of fibers by weight (UQL), immature fiber content (IFC), short fiber content by number (SFCn), and fineness. Among these properties, UQL was defined as "the length which was exceeded by 25% of the fibers by weight" as described in the Uster AFIS manual; IFC was defined as the percentage of fibers with less than 0.25 circularity; SFCn was defined as the percentage by number of the fibers that were less than 12.7 mm; and fineness was defined as the weight per unit of length (mg km^{-1}). Two samples of lint of 0.5 g each were collected from each ginned boll sample for the AFIS measurements. Mote weight (mg g^{-1}; weight per gram of ginned lint), and mote count (cnt g^{-1}; counts per gram of ginned lint), were determined using two of the four replicates planted at site 1 in 2008, and all three replicates planted at site 1 in 2009. Two samples of ginned lint, 5 g each, from each plot were manually measured for mote traits as described in Method D2496 (ASTM, 1985).

The General Linear Model procedure of the Statistical Analysis System (SAS Institute, 2004) was used for analysis of variance in all traits. A mixed model was used with genotype as fixed effect and environment, genotype × environment, and replicate within environments as random effects. Sum of squares for genotype was partitioned into variation due to parent, parents vs. hybrids, and hybrids. Sum of squares of hybrids was further partitioned into variation due to females, males, and female × male. General combining ability (GCA) effects of the female and male parents were calculated as main effects of the females and males, respectively, and specific combining ability (SCA) effects were calculated as the female × male interactions (Hallauer and Miranda, 1981). For GCA effects of each parent, standard errors for GCA effects of females and males were calculated followed the method described by Cox and Frey (1984). The calculated standard errors were used in two-tailed t tests to determine significance for each GCA effect (Cox and Frey, 1984).

Means among F_2 hybrids were compared using Fisher's least significance difference (LSD) test (5%) from the 48 hybrids. The means of the F_2 hybrids were compared with the means of the parents in t tests using error mean squares in the F_2 hybrids (df = 470) and parents (df = 150) as variances in the tests. Phenotypic correlation coefficients between nep count and SCN count and other traits including lint yield and the AFIS-measured properties were analyzed using means across environments.

RESULTS AND DISCUSSION

Forty-eight F_2 hybrids from crosses between four elite germplasm lines and 12 exotic germplasm lines were evaluated and analyzed for variances in the AFIS-measured properties such as neps, SCN, and manually measured mote traits (Table 1). Data in Table 1 showed that effects of GCA were more important than specific combining ability (SCA) effects for neppiness traits. Effects of GCA, estimated from either female or male parents, were observed for all traits except nep size from male parent and SCN size and mote weight from female parent. In contrast, effects of SCA were not significant for all properties except UQL. Even for UQL, mean square of GCA from female and male parents was 15 and 1.8 times of that for SCA, respectively. According to Sprague and Tatum (1942), GCA and SCA effects were equivalent to additive and nonadditive effects of dominance and epistasis, respectively. Therefore, in this study, the neppiness traits neps, SCN, and motes were controlled mainly by additive genes in F_2 hybrids.

Table 1. Mean squares of AFIS-measured properties and mote traits in F_2 hybrids and the parental lines.

Source	df.	Nep size $\times 10^{-3}$	Nep count $\times 10^{-3}$	SCN[x] size $\times 10^{-4}$	SCN count	UQL	SFCn	IFC	Mote weight[y]	Mote count
Environments (E)	3	16.30***[z]	15.50***	15.0	91.0***	156.0***	737.0***	50.0***	4549***	0.94
Replicate (E)	10	4.47***	1.59***	54.0***	25.0***	28.6***	90.0***	4.05***	18	0.32
Genotype (G)	63	1.70*	1.76***	13.9	17.6***	18.0***	14.3***	2.08***	104***	0.72***
Parents (P)	15	2.27*	3.34***	11.0	36.5***	22.8***	18.9***	3.26***	178***	1.00***
Female (F)	3	1.34	0.52	12.6	8.8	10.1***	45.9***	3.13***	51	0.39
Male (M)	11	2.17*	2.59***	6.4	30.5***	26.1***	13.2*	2.90***	205***	0.85
F vs. M	1	6.16*	20.10***	57.0*	185.0***	24.8***	1.1	7.59***	265*	4.79**
Hybrids (H)	47	1.55	1.15***	15.2	10.9**	15.9***	13.1***	1.66***	74*	0.60***
GCA(female)	3	3.64*	3.73***	27.1	35.0**	117.0***	54.8***	6.30***	87	1.86***
GCA(male)	11	1.94	2.43***	26.7*	20.9***	13.3**	26.6***	4.23***	145**	0.82**
SCA	33	1.23	0.50	10.2	5.4	7.6*	4.8	0.37	50	0.41
P vs. H	1	0.00	6.30***	0.5	46.1*	42.2**	0.01	4.53***	410**	2.15**
G × E	189	1.26	0.65***	16.5	7.2	6.4*	6.6*	0.40	72*	0.43*
P × E	45	0.98	0.81**	14.4	9.4	0.9	8.8**	0.44	56	0.59*
(F vs. M) × E	3	0.32	0.70	3.2	0.8	2.6***	14.9	1.24*	3	0.06
F × E	9	1.19	0.27	9.8	2.5	0.6	4.5	0.17	49	0.51
M × E	33	0.97	0.97**	16.7	12.0*	0.9**	9.5*	0.44	62	0.66
(P vs. H) × E	3	1.78	0.41	5.3	7.6	9.2	21.2**	1.04*	47	0.45
H × E	141	1.34	0.61**	17.4	6.5	8.1***	5.6	0.37	78*	0.38
GCA(female) × E	9	1.12	0.92*	9.1	6.2	18.8***	10.0*	0.60	56	0.18
GCA(male) × E	33	1.40	1.07***	18.7	5.9	6.6	5.2	0.36	85	0.54
SCA × E	99	1.34	0.43	17.7	6.7	7.6**	5.3	0.35	77*	0.35
Error	630	1.19	0.45	14.5	7.0	5.1	5.3	0.33	49	0.30

[x] SCN, seed coat nep; UQL, upper quartile length; SFCn, short fiber content by number; IFC, immature fiber content.

[y] Mote weight was measured as mg per gram of ginned lint; mote count was measured as count per gram of ginned lint; these two traits were measured using two of the four replicates planted at site 1 in 2008 (Environment 1) and all three replicates at site 1 in 2009 (Environment 3).

[z] Values followed by *, **, *** are significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively.

Data in Table 1 also showed similar stability across environments for the neppiness traits between hybrids and parents because mean squares of hybrid × environment were nearly equal to those of parent × environment for fiber nep, SCN, and mote traits. When mean squares of hybrid × environment were further partitioned into sources of GCA and SCA, significant GCA$_{female}$ × environment interactions were observed for nep count, UQL, and SFCn and significant GCA$_{male}$ × environment interaction was observed for nep count (Table 1). These significant interactions suggest breeding problem across environments for these specific traits, although significant GCA × environment was not observed for most traits analyzed.

Performance of Parents and Hybrids. Overall genotypic effects among parents were observed for all traits except SCN size (Table 1). When parental effects were further partitioned into female and male effects, the majority of genotypic variations in nep size, nep count, SCN count, mote weight, and mote count were attributed to male parents based on mean squares (Table 1). Among the four female parents, the elite germplasm lines, TAM 98D-99ne performed best in terms of neppiness traits with the lowest nep size, nep count, mote weight, and mote count. MD15 had the lowest SCN size and SCN count (Table 2). The general good performance among female parents for the neppiness traits was expected because these lines were included in the

crossing scheme due to their reputation for good fiber quality in their original regions. As expected, a wider range of these traits was observed among the male parents, the exotic germplasm lines, than those among the female parents. Among the 12 male parents, SP156 performed best with desirable combination between nep count and SCN count and JC84 performed best for mote weight and mote count (Table 2).

Although the means of all AFIS-measured properties and mote traits were not significantly different between hybrids and parents (Table 2), these traits were generally diverse among the 48 F_2 hybrids (Table 3). For example, nep count, SCN count, and mote count ranged from 60 to 103 cnt g^{-1}, 2.1 to 6.6 cnt g^{-1}, and 0.88 to 2.5 cnt g^{-1}, respectively, among the hybrids. Among the 48 F_2 hybrids, MD15 × SP156 was a good performer for a desirable combination between nep count and

SCN count (Table 3) compared to the best female parent, MD15, for the same properties. MD15 × SP235 was another good performer for a desirable combination between mote weight and mote count (Table 3) compared to the best female parent, TAM 98D-99ne for the same traits.

Nep size and SCN size are important fiber properties because a critical size might exist above which fiber neps are registered as yarn faults (Frydrych and Matusiak, 2002). Although the assumed critical values depend on yarn linear density and therefore vary in different studies, genetically improved germplasm lines with reduced nep size and SCN size can limit the number of nep and SCN registered as yarn imperfections. FM832 × SP235 had the lowest SCN size, 950 μm, and MD52 × SP164 and MD15 × SP156 had the lowest nep size, 641 μm, among the 48 F_2 hybrids (Table 3).

Table 2. Means of the neppiness traits and other AFIS-measured properties in parents.

Pedigree	Nep size μm	Nep count cnt g^{-1}	SCN[x] size μm	SCN count cnt g^{-1}	UQL mm	SFCn %	IFC %	Mote weight[y] mg g^{-1}	Mote count cnt g^{-1}
FM832	659	72.5	1200	4.43	32.9	14.7	3.89	9.3	1.54
TAM98D-99ne	641	57.9	1006	3.43	31.1	11.4	3.09	8.9	1.12
MD52ne	663	64.0	1117	3.64	32.0	11.0	2.79	14.7	1.80
MD15	649	63.0	1004	2.50	32.8	11.0	3.43	14.5	1.46
SP156	655	63.4	1272	2.93	31.9	11.1	2.90	32.8	2.90
SP164	650	117.0	1040	4.14	34.1	12.5	3.94	14.9	1.66
SP192	644	76.6	1201	4.29	28.4	11.6	3.23	18.8	2.18
SP205	665	78.9	1179	5.79	30.8	11.1	3.37	13.7	2.10
SP224	668	91.0	1170	6.64	30.4	13.5	4.22	14.8	2.26
SP235	665	93.9	1132	5.07	32.2	10.6	3.26	14.6	1.86
JC14	666	86.8	1218	7.29	32.2	13.0	3.89	23.2	2.68
JC32	672	93.4	1209	5.93	31.0	12.8	4.29	11.1	1.80
JC60	662	81.1	1177	5.00	31.2	12.4	4.03	14.7	2.02
JC65	691	85.6	1287	8.29	31.0	11.6	3.58	14.1	1.64
JC84	662	72.6	1265	5.29	32.3	12.8	3.77	10.2	1.56
JC186	677	93.6	1226	6.57	31.5	13.4	4.22	9.9	1.88
LSD	25	17.0	268	2.10	0.5	1.8	0.48	9.2	0.94
Mean parents[z]	662	81.0	1169	5.08	31.6	12.1	3.62	15.0	1.90
Mean F_2	662	75.0	1174	4.55	32.1	12.1	3.46	12.4	1.72

[x] SCN, seed coat nep; UQL, upper quartile length; SFCn, short fiber content by number; IFC, immature fiber content.

[y] Mote weight was measured as mg per gram of ginned lint; mote count was measured as count per gram of ginned lint; these two traits were measured using two of the four replicates planted at site 1 in 2008 (Environment 1) and all three replicates at site 1 in 2009 (Environment 3).

[z] t tests were made between the means of the 16 parents and the means of the 48 F_2 hybrids.

Table 3. Means of the neppiness traits and other AFIS-measured properties in the 48 F2 hybrids.

Pedigree	Nep size µm	Nep count cnt g[-1]	SCN[x] size µm	SCN count cnt g[-1]	UQL mm	SFCn %	IFC %	Mote weight[y] mg g^{-1}	Mote count cnt g[-1]
FM832×SP156	675	64.8	1216	3.21	32.4	12.1	2.85	8.7	1.34
FM832×SP164	648	89.9	1138	3.29	34.0	14.7	4.01	9.3	1.76
FM832×SP192	664	66.9	1143	4.57	32.4	12.1	3.19	11.8	1.82
FM832×SP205	650	76.7	1253	4.21	32.4	14.1	3.32	13.9	1.96
FM832×SP224	657	69.8	1035	4.50	31.6	13.7	3.79	14.5	1.60
FM832×SP235	655	75.5	950	4.36	32.9	13.4	3.09	12.3	1.16
FM832×JC14	658	79.5	1120	5.29	32.1	12.6	3.71	12.1	1.82
FM832×JC32	670	72.1	1260	4.50	32.5	12.7	3.84	14.5	1.50
FM832×JC60	672	72.6	1206	4.64	33.4	12.8	3.91	11.8	1.84
FM832×JC65	674	79.9	1111	5.21	32.7	12.6	3.50	7.3	1.52
FM832×JC84	673	79.3	1273	5.36	32.3	12.5	3.40	14.6	1.74
FM832×JC186	671	75.9	1088	5.79	32.9	11.8	3.56	12.5	2.02
TAM98D-99ne×SP156	667	59.5	1241	4.29	31.7	12.2	2.91	10.5	1.56
TAM98D-99ne×SP164	671	71.1	1307	4.07	33.5	12.2	3.10	9.1	1.32
TAM98D-99ne×SP192	678	65.1	1161	5.43	31.3	10.6	2.78	7.4	0.98
TAM98D-99ne×SP205	662	76.1	1381	4.29	31.8	12.5	3.17	8.0	1.08
TAM98D-99ne×SP224	654	63.9	1186	3.86	29.7	12.3	3.34	9.5	1.32
TAM98D-99ne×SP235	675	68.9	1137	5.00	29.8	11.3	2.99	10.3	1.64
TAM98D-99ne×JC14	666	60.5	1045	4.36	30.0	10.9	3.37	12.2	1.86
TAM98D-99ne×JC32	663	65.9	1424	4.36	31.5	11.2	3.31	13.8	1.60
TAM98D-99ne×JC60	653	67.9	1056	4.86	32.2	10.6	3.39	11.2	1.66
TAM98D-99ne×JC65	685	71.9	1269	6.43	31.2	12.4	3.15	12.3	1.86
TAM98D-99ne×JC84	671	68.6	1192	5.29	31.1	10.8	3.05	26.9	1.80
TAM98D-99ne×JC186	662	76.3	1162	6.00	29.2	12.6	3.61	10.6	1.82
MD52×SP156	667	73.0	1237	5.21	32.3	12.6	3.34	13.7	1.70
MD52×SP164	641	92.9	1314	4.07	31.3	12.6	3.53	12.3	2.26
MD52×SP192	645	70.1	1117	4.57	31.3	10.9	2.92	12.9	1.68
MD52×SP205	653	95.0	1093	5.29	32.4	12.6	3.44	10.0	1.68
MD52×SP224	655	76.6	1161	3.93	31.2	13.0	3.83	19.3	2.42
MD52×SP235	660	65.1	1195	4.36	32.8	12.2	2.95	13.4	1.56
MD52×JC14	652	75.5	982	4.07	32.2	11.3	3.46	11.0	2.00
MD52×JC32	664	75.1	1198	5.43	31.9	11.5	3.63	13.4	1.86
MD52×JC60	675	82.1	1231	6.07	31.7	11.9	3.57	12.1	2.16
MD52×JC65	672	92.7	1298	6.57	31.6	12.9	3.82	14.6	2.48
MD52×JC84	669	71.3	1437	4.29	31.7	12.6	3.54	24.4	2.02
MD52×JC186	662	79.6	1180	4.71	31.4	12.6	3.79	13.3	1.70
MD15×SP156	641	64.1	1154	2.14	33.0	10.7	3.12	8.7	1.40
MD15×SP164	651	103.0	1190	3.71	34.3	13.3	4.07	10.7	1.70
MD15×SP192	649	60.1	1028	3.57	31.6	10.2	3.07	16.2	1.90
MD15×SP205	649	75.9	1141	3.57	33.0	13.7	3.71	10.4	1.62
MD15×SP224	656	75.0	1079	2.79	33.1	11.6	3.91	9.1	1.10
MD15×SP235	673	77.1	1187	3.79	32.9	12.6	3.17	6.2	0.88
MD15×JC14	668	69.6	1053	4.21	33.3	10.8	3.78	8.2	1.74
MD15×JC32	656	83.8	1077	4.57	32.8	11.8	3.77	13.1	1.88
MD15×JC60	666	77.0	1199	5.00	32.8	10.9	3.79	11.4	2.22
MD15×JC65	647	75.0	1161	4.36	32.4	12.5	3.82	17.0	2.12
MD15×JC84	669	80.1	1225	4.86	32.5	11.7	3.61	14.4	1.78
MD15×JC186	654	74.6	1081	4.21	33.4	12.0	3.82	14.6	1.86
LSD[z]	26	16.0	283	1.90	1.7	1.7	0.43	8.8	0.59

[x] SCN, seed coat nep; UQL, upper quartile length; SFCn, short fiber content by number; IFC, immature fiber content.

[y] Mote weight was measured as mg per gram of ginned lint; mote count was measured as count per gram of ginned lint; these two traits were measured using two of the four replicates planted at site 1 in 2008 (Environment 1) and all three replicates at site 1 in 2009 (Environment 3).

[z] Multiple comparisons were made among the 48 F_2 hybrids.

Combining Ability for Neppiness Traits. Different female parents were identified with GCA effects for different neppiness traits (Table 4). TAM 98D-99ne and MD15 were good combiners with negative (favorable) GCA effects for different neppiness traits. TAM 98D-99ne had negative GCA effects for nep count and mote count, and MD15 had negative GCA effects for nep size, SCN size, and SCN count. In general, the GCA effects from female parents were consistent across environments except the GCA effect for nep size in MD15, which was significant in two of the four environments (data not shown).

In male parents of the exotic germplasm lines, there were four, three, and two of the 12 parents detected with GCA effects ($P < 0.05$) for reducing nep count, SCN count, and mote count, respectively (Table 4). In general, SP germplasm lines were better combiners for the neppiness traits than JC germplasm lines. SP156 and SP224 were the best combiners with favorable GCA effects for different neppiness traits. SP156 had negative GCA effects for nep count, SCN count, and mote count and SP224 had negative GCA effects for nep size, nep count, SCN size, and SCN count. JC14 was the best combiner among JC germplasm lines with negative GCA effect for nep count and SCN size.

Two to six F_2 hybrids were detected for SCA for different neppiness traits (data not shown). Negative SCA effects ($P < 0.05$) were detected in four F_2 hybrids for nep size, three for nep count, three for SCN size, two for SCN count, and six for mote count. Most of these effects were not consistent across environments with different number of F_2 hybrids detected with SCA effects in different environments (data not shown). These results indicated small heterosis for the neppiness traits in F_2 hybrids.

Potential of Simultaneous Genetic Improvement for Neppiness Traits with Other Fiber Traits. FM832 was the only female parent with positive GCA effect for lint yield and negative GCA effect for one neppiness trait, SCN size (Table 4). Although MD15 had favorable GCA effects for three of the five neppiness traits, negative GCA effect for lint yield was also detected in this parent. TAM 98D-99ne had negative GCA effects for two of the five neppiness traits, nep count and mote count, but it had no GCA effect for lint yield. Therefore, female parents were not good general combiners when both lint yield and multiple neppiness traits were considered.

Table 4. General combining ability for neps, seed coat neps, and other AFIS-measured properties of the 16 parents across environments.

	Lint yield[y] kg ha⁻¹	Nep size µm	Nep count cnt g⁻¹	SCN[x] size µm	SCN count cnt g⁻¹	Mote count cnt g⁻¹	UQL mm	SFCn %	IFC %	Fineness mg km⁻¹
FM 832	96.3***[z]	2.03	0.63	-24.9*	0.025	-0.041	0.529**	0.770***	0.060*	-1.70*
TAM 98D-99ne	12.0	5.33***	-6.64***	38.9**	0.299*	-0.173***	-1.030***	-0.519***	-0.273***	8.38***
MD52ne	-35.7	-2.08	4.41***	29.1**	0.329**	0.245***	-0.310*	0.081	0.031	0.35
MD15	-72.5**	-5.31**	1.61*	-43.1***	-0.653***	-0.031	0.810***	-0.333**	0.183***	-7.02***
SP156	95.3***	0.68	-9.28***	37.7	-0.837***	-0.220*	0.261*	-0.258*	-0.400***	5.24***
SP164	1.0	-9.37***	14.30***	62.8*	-0.766***	0.045	1.140***	1.020***	0.224***	-2.54**
SP192	59.0**	-2.75	-9.08***	-62.4*	-0.016	-0.120	-0.452**	-1.180***	-0.465***	8.21***
SP205	225.0***	-8.49**	6.31**	42.4	-0.213	-0.130	0.300*	1.060***	-0.042	0.851
SP224	84.4***	-6.11*	-3.31*	-59.2*	-0.784***	-0.105	-0.704*	0.483**	0.262***	-1.61*
SP235	-142.0***	3.95*	-2.97	-57.1*	-0.177	-0.405**	-0.032	0.231	-0.406***	5.92***
JC14	-157.0***	-0.82	-3.35*	-124.0***	-0.070	0.140	-0.241	-0.735***	0.126**	-5.33***
JC32	5.0	1.47	-0.37	65.3*	0.162	-0.005	0.086	-0.353*	0.183***	-1.60*
JC60	-147.0***	4.84*	0.28	-1.5	0.591**	0.255*	0.402*	-0.592**	0.210***	-5.36***
JC65	44.1*	7.45**	5.28*	35.5	1.090***	0.280*	-0.155	0.468**	0.119**	-0.08
JC84	-41.9*	8.90***	0.19	107.0**	0.394*	0.120	-0.211	0.266*	-0.053	-1.01
JC186	-26.6	0.18	1.96	-46.6*	0.627**	0.135	-0.399*	0.118	0.244***	-2.70***

[x] SCN, seed coat nep; UQL, upper quartile length; SFCn, short fiber content by number; IFC, immature fiber content.

[y] GCA effects in columns of lint yield and fineness were reported previously (Zeng et al., 2011) and listed in this table solely for discussion of their simultaneous improvement with the neppiness traits.

[z] Values followed by *, **, *** are significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively.

Overall, SP156, SP192, and SP224 were better combiners than the female parents with positive GCA effects for lint yield and negative GCA effects for at least two neppiness traits (Table 4). Among them, SP156 was detected with favorable GCA effects for three neppiness traits, nep count, SCN count, and mote count, with positive GCA effect for lint yield. This line also had negative GCA effects for SFCn and IFC, -0.26% and -0.40%, respectively, and positive GCA effect for UQL, 0.26 mm. SP224 had favorable GCA effects for four of the five neppiness traits, nep size, nep count, SCN size, and SCN count with positive GCA effect for lint yield, but this line had unfavorable GCA effects for UQL, SFCn, and IFC. Some of these GCA effects were not consistent across environments, e.g., GCA effect for nep count in SP224 was only significant in two of the four environments and GCA effect for mote count in SP156 was only significant in one of the two environments. Most GCA effects were consistent in the environments with significant effects in at least three of the four environments (data not shown). Results indicated that SP156 can be used as a parent in breeding for simultaneous genetic improvement of multiple neppiness traits with lint yield and other AFIS-measured properties.

Phenotypic Correlations Between Neps and Other Traits. Negative correlation between lint yield and nep count in parents (r = -0.52) (Table 5) was consistent with the genotypic correlation between the two traits (r = -0.25) in 200 JC lines evaluated previously (Zeng and Meredith, 2010). In F_2 hybrids, the correlation coefficient between lint yield and nep count was not significant (Table 5). Generally, correlations between SCN and other traits were either low or inconsistent among parents and F_2 hybrids (Table 5).

Table 5. Phenotypic correlation coefficients of fiber nep and SCN to other traits.

	Lint yield	UQL[y]	SFCn	IFC	Fineness	Mote count
Parents						
Nep count	-0.52*[z]	0.31	0.35	0.63**	-0.48	0.15
SCN count	-0.45	-0.17	0.32	0.54*	-0.45	0.38
F_2						
Nep count	0.00	0.31*	0.52***	0.60***	-0.50***	0.32*
SCN count	-0.05	-0.34*	-0.07	-0.01	0.19	0.36*

[y] UQL, upper quartile length; SFCn, short fiber content by weight; IFC, immature fiber content; SCN, seed coat nep

[z] Values followed by *, **, *** are significant at $P < 0.05$, $P < 0.01$, $P < 0.001$, respectively.

The correlations between mote count and nep count and between mote count and SCN count were either not significant or low in parents and F_2 hybrids (Table 5). These results are not consistent with the argument that motes in ginned fibers easily cause neps and SCN. The unexpected results could be explained by (1) the different samples used for mote counts and AFIS measurements and (2) more importantly, the size of motes identified in this study. According to Davidonis et al. (2000), small motes (1- 30 mg mote[-1]) usually have fibers shorter than half of the length as those on normal seeds. Obviously, all motes detected in the F_2 hybrids and parents belonged to small motes because the derived trait, weight per mote, ranged from 6 to 11 mg mote[-1] among the 64 entries (data not shown). It is possible that most of these motes with short fibers fell off the lint stream during opening of fibers in AFIS measurements.

Relationships among nep count, SFCn, and fineness determine the possibility for simultaneous improvement or antagonism among these properties. Correlation coefficients between nep count and SFCn were positive in F_2 hybrids. In contrast, correlation coefficients between nep count and fineness were negative in both parents and F_2 hybrids (Table 5). Similar results were reported previously in an evaluation of 200 JC germplasm lines with genotypic correlations of 0.74 and -0.75 for nep count vs. SFCn, and nep vs. fineness, respectively (Zeng and Meredith, 2010). The negative correlation between nep count and fineness is understandable because finer fibers are more likely to entangle into fiber neps (van der Sluijs and Hunter, 1999). The positive correlation between nep count and SFCn implies possibility for simultaneous improvement of the two properties, and the negative correlation between nep count and fineness implies an antagonistic relationship between the two properties in selection. However, the low correlation coefficient between nep count and fineness, e.g., -0.50 in F_2 hybrids in this study, usually implies the possibility of recombination between the two properties in hybrids if unfavorable linkage exists. These relationships were reflected by combining ability for these three properties in F_2 hybrids when none of the SP lines and female parents was detected with favorable GCA effects for all three properties. However, among the JC germplasm lines, JC14 had favorable GCA effects for nep count, SFCn, and fineness (Table 4).

In summary, significant GCA effects were detected in F$_2$ hybrids for the neppiness traits. The neppiness traits are controlled mainly by additive genes in F$_2$ hybrids. These results indicate that selection for neppiness traits can be made in early generations in breeding using SP and JC germplasm. The identification of favorable GCA effects for most neppiness traits and other AFIS-measured properties with positive GCA effect for lint yield provides evidence for the possibility of simultaneous improvement of multiple neppiness traits with lint yield and other fiber properties in Upland cotton. Because no parent was identified with favorable GCA effects in all neppiness and other fiber traits, multiple crosses will be necessary to further select for simultaneous genetic improvement of these traits with enhanced recombination among these parents.

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DISCLAIMER

Mention of trade names or commercial products in this article is solely for the purpose of providing specific information and does not imply recommendation by the U.S. Department of Agriculture.

REFERENCES

Altintas, P.Z., J.L. Simonton, and M.G. Beruvides. 2007. Pilot study to examine the relationship between AFIS fiber properties and white speck occurrence. J. Cotton Sci. 11:118–127.

American Society for Testing and Materials (ASTM). 1985. Standard test method for seed coat fragments and funiculi in cotton fiber samples, D2496. *In* Annual Book of American Society of Testing and Materials. Vol. 07.01 Textiles. American Society for Testing and Materials,, West Conshohocken, PA.

American Society for Testing and Materials (ASTM). 1999. Standard terminology relating to textiles. p. 43. *In* Annual Book of American Society of Testing and Materials. Vol. 1 D123-96. American Society for Testing and Materials,, West Conshohocken, PA.

Baldwin, J.C., M. Qaud, and A.C. Schleth. 1995. AFIS seed coat nep measurement. p. 1250–1251. *In* Proc. Beltwide Cotton Conf. San Antonio, TX. 4-7 Jan. 1995. Natl. Cotton Council Am., Memphis, TN.

Bolek, Y, M. Oglakci, and K. Ozdin. 2007. Genetic variation among cotton (*G. hirsutum* L.) cultivars for motes, seed-coat fragments and loading force. Field Crop Res. 101:155–159.

Boykin, J.C. 2008. Seed coat fragments, motes, and neps: Cultivar differences. J. Cotton Sci. 12:109–125.

Cotton Incorporated. 2011. U.S. Cotton Market Monthly Economic Letter – July 13, 2011 [Online]. Available at http://www.cottoninc.com/Marketinformation/MonthlyEconomicLetter/ (verified 1 Dec. 2011).

Cox, D.J., and K.J. Frey. 1984. Combining ability and the selection of parents for interspecific oat matings. Crop Sci. 24:963–967.

Davidonis, G.H., A. Johnson, and J.A. Landivar. 2000. Cotton mote frequency under rainfed and irrigated conditions. J. Cotton Sci. 4:1–9.

Frydrych, M., M. Matusiak, and T. Swiech. 2001. Cotton maturity and its influence on nep formation. Textile Res. J. 71:595–604.

Frydrych, M., and M. Matusiak. 2002. Predicting the nep number in cotton yarn – determining the critical nep size. Textile Res. J. 72:917–923.

Hallauer, A.R., and J.B. Miranda. 1981. Quantitative Genetics in Maize Breeding. Iowa State Univ. Press, Ames, IA.

Hebert, J.J., G. Mangialardi, and H.H. Ramey. 1988. Anatomy of a nep. Textile Res. J. 58:380– 382.

Jacobsen, K.R., Y.L. Grossman, Y.L. Hsieh, R.E. Plant, W.F. Lalor, and J.A. Jernstedt. 2001. Neps, seed coat fragments, and non-seed impurities in processed cotton. J. Cotton Sci. 5:53–67.

Krifa, M., and R. Frydrych. 2002. Seed coat fragments: The consequences of carding and the impact of attached fibers. Textile Res. J. 72:259–265.

Krifa, M., and J.P. Gourlot. 2001. Effect of seed coat fragments on cotton yarn strength: Dependence on fiber quality. Textile Res. J. 71:981–986.

Mangialardi, G.J., Jr. 1986. Evaluation of nep formation in the cotton gin. Textile Res. J. 55:756–761.

Meredith, W.R., Jr. 2005. Registration of MD52ne high fiber quality cotton germplasm and recurrent parent MD 90ne. Crop Sci. 45:807–808.

Meredith, W.R., Jr. 2006. Registration of MD15 upland cotton germplasm. Crop Sci. 46:2722.

Meredith, W.R., and R.R. Bridge. 1971. Breakup of linkage blocks in cotton, *Gossypium hirsutum* L. Crop Sci. 11:695–698.

Miller, P.A., and J.O. Rawlings. 1967. Breakup of initial linkage blocks through intermating in a cotton breeding program. Crop Sci. 7:199–204.

Novick, R.G., J.E. Jones, W.S. Anthony, W. Aguillard, and J.I. Dickson. 1991. Genetic trait effects on nonlint trash of cotton. Crop Sci. 31:1029–1034.

Pearson, N.L. 1955. Seed coat fragments in cotton — an element of yarn quality. USDA Tech. Bull. No.1116. USDA, Washington, DC.

Saranga, Y., N. Sass, Y. Tal, C. Shimony, and R. Yucha. 1997. Effects of motes on lint quality of interspecific cotton hybrids. Crop Sci. 37:1577–1581.

SAS Institute. 2004. SAS User's Guide. Version 9. SAS Inst., Cary, NC.

Smith, R. 2010. Quality remains crucial factor for continued cotton export success. Farm Press, 4 January 2010. Available online at http://southwestfarmpress.com/ cotton/cotton-exports-0104/. (verified 1 Dec. 2011).

Sprague, G.F., and L.A. Tatum. 1942. General vs. specific combining ability in single crosses of corn. Agron. J. 34:923–932.

Thaxton, P.M., C.W. Smith, and R. Cantrell. 2005. Registration of TAM 98D-102 and TAM 98D-99ne upland cotton germplasm lines with high fiber strength. Crop Sci. 45:1668–1669.

USDA Economic Research Service (ERS), 2011. U.S. Upland , ELS, and all cotton estimates [online]. Available at http://www. usda.gov/oce/commodity/wasde/(verified 1 Dec. 2011).

USDA-Foreign Agricultural Service (FAS), 2008. World cotton import demand trends up

[Online]. Available at http://www.fas.usda.gov/cotton/ highlights/2008/IATRoverview04-08-08.pdf. (verified 1 Dec. 2011).

Uster AFIS. 1997. Instruction Manual of the Advanced Fiber Information System: Instrument for

Measuring Neps, Length, Diameter, and Trash for Fibers. Zellweger Uster, Inc., Knoxville, TN.

van der Sluijs, M.H.J., and L. Hunter. 1999. Neps in cotton lint. p. 1–51. *In* J.M. Layton (ed.) Textile Progress. Vol. 28, No. 4. The Textile Institute, Oxford, UK.

Zeng, L. and W.R. Meredith, Jr. 2009. Associations among lint yield, yield components, and fiber properties in an introgressed population of cotton. Crop Sci. 49:1647–1654.

Zeng, L., and W.R. Meredith, Jr. 2010. Neppiness in an introgressed population of cotton: Genotypic variation and genotypic correlation. J. Cotton Sci. 14:17–25.

Zeng, L., W.R. Meredith, Jr., and D.L. Boykin. 2011. Germplasm potential for continuing improvement of fiber quality in upland cotton: Combining ability for lint yield and fiber quality. Crop Sci. 51:60–68.

Zeng, L., W.R. Meredith, Jr., D.L. Boykin, and E. Taliercio. 2007. Evaluation of an exotic germplasm population derived from multiple crosses among *Gossypium* tetraploid species. J. Cotton Sci. 11:118–127.