Fruiting Sites in Cotton: Seed Quality

Edith J. Conkerton,* Dorselyn C. Chapital, Bryan T. Vinyard, G. Russell Schneider, and Johnie N. Jenkins

Southern Regional Research Center, Agricultural Research Service, U.S. Department of Agriculture, P.O. Box 19687, New Orleans, Louisiana 70179-0687, and Crop Science Research Laboratory, P.O. Box 5367, Mississippi State, Mississippi 39762

Cottonseed, *Gossypium hirsutum* L., production and quality are affected by agronomic practices, cultivar differences, and environmental conditions. Experiments were conducted to compare the quality of seeds produced from six cultivars. Cotton was grown in experimental plots at Mississippi State, MS. At harvest, bolls were collected from selected positions on fruiting branches of the plants. After ginning, seed was collected for evaluation. Quality of the seed was determined by seed weight, kernel weight, size and color, microbial content, oil content, and free fatty acid (FFA) content of the oil. For all cultivars, yield and quality were highest in bolls adjacent to the main stem on branches in the center of the plant. Statistical analysis indicated that seed yield, seed and kernel weight, and oil content exhibited a significant ($p < 0.05$) cultivar by fruiting site interaction. FFA and microbial content were significantly ($p < 0.05$) different among cultivars and fruiting sites.

INTRODUCTION

Historical references indicate the use of the cotton plant as a source of fiber as early as 3000 B.C., with an authenticated reference to cotton fabric in 800 B.C. Although some seed may have been crushed to provide oil for medicinal purposes, for many centuries cottonseed was considered a valueless byproduct. In the United States, cottonseed (*Gossypium hirsutum* L.) did not become an important agricultural commodity until the early 1900s (Cooper, 1948). Since that time, there have been intermittent periods of research on seed composition and effects of genetics, agronomics, and environment on the quality of cottonseed (Cherry et al., 1981; Kohel and Cherry, 1983). Even today, however, changes in cultivars and planting practices are designed to improve the yield and quality of the cotton fiber. There is minimal consideration of how these changes affect the seed.

The most recent changes in cotton production occurred during the mid to late 1980s and resulted from the ability of breeders to reduce the number of days from planting to final harvest, i.e., the development of early maturing cultivars. In 1978, early maturing cultivars constituted 8–22% of the crop in the Mid-South cotton producing area. By 1986, these cultivars made up 76–99% of the crop (Bridges and McDonald, 1987). Early maturity was accompanied, usually, by increased yields of seed cotton. Researchers at Mississippi State noted characteristics of these early maturing cultivars which differed from cultivars more common to that area such as cv. Stoneville 213 (ST 213) (Jenkins et al., 1986; McCarty et al., 1986). Experiments were conducted, therefore, to compare Stoneville cultivars with early maturing cultivars. These studies provided information on yield, boll size, and boll set percentage at various fruiting sites on the plant in the selected cultivars (Jenkins et al., 1990a,b). Simultaneously, seed from these cultivars were collected at Mississippi State and then sent to Southern Regional Research Center (SRRC). The objectives of the research at SRRC were to determine the quality of the seed at selected fruiting sites and to compare the quality of seed from the different cultivars. In this paper the following seed quality parameters will be reported: seed weight, kernel (embryo) weight and condition, microbial content, oil content, and free fatty acid content of the oil. Variations and similarities of the seed from positions on the plant and among the cultivars will be discussed.

* Author to whom correspondence should be addressed at the Southern Regional Research Center.

MATERIALS AND METHODS

Field Studies. The six cultivars used were Stoneville 213 (ST 213), Stoneville 506 (ST 506), Stoneville 825 (ST 825), Deltapine 20 (DPL 20), Deltapine 50 (DPL 50), and Delta Experiment Station 119 (DES 119). The latter three are early maturing cultivars. The production practices and data collection procedures were similar to those used by Jenkins et al. (1990a). Cultivars were planted in two-row plots, 14 m long spaced 1 m apart, in a two-row-planted to one-row-not-planted pattern. Seeds were planted on April 28, 1988, in a Marietta sandy clay loam (fine-loamy, siliceous, thermic Fluvaquentic Eutrochrept) soil and emergence was on May 5. Plants were thinned to approximately 95 000 plants ha^{-1} (10.16 cm between plants). Plots were fertilized with N, with 56 kg ha^{-1} applied at preplant and 84 kg ha^{-1} as a sidedress at first bloom. The fungicide Terrachlor super-x (5-ethoxy-3-(trichloromethyl)-1,2,4-thiadiazole) at the rate of 11.2 kg ha^{-1} and the insecticide aldicarb [2-methyl-2-(methylthio)propionaldehyde *O*-(methylcarbamoyl) oxime] at 0.34 kg of ai ha^{-1} were applied in furrow at planting. A full-season insecticide program was used to control insects and mites. Irrigation water (10 cm) was applied by drip over a 48-h period beginning June 30. Plots were in a randomized complete-block experiment with six replications.

After all bolls had opened, bolls were hand picked from selected fruiting sites as described by Jenkins et al. (1990a). Fruiting sites (FS) are specific position and node combinations. As diagrammed in Figure 1, position (P) indicates the order in which buds (potential bolls) form on a fruiting branch, with "1" being closest to the main stem. Node (N) indicates the place on the main stem where branches arise, with the cotyledonary node designated "1". In this study, bolls were collected from positions 1, 2, and 3 and nodes 6–22. Bolls from each FS were kept separate and weighed to provide the yield of seed cotton, i.e., the weight of lint plus seed. Seed yield was calculated from lint percentage determinations and seed cotton yield data. For each cultivar and FS, bolls from two field replicates were combined to acquire sufficient seed for each of the triplicate seed cotton analyses. Seed cotton was stored at ambient temperature (22 °C) until ginned; thereafter, the seeds were stored in paper bags at 22 °C until shipped to Southern Regional Research Center.

Table I. Analysis of Variance: Mean Squares[a]

source of variation[b]	DF[c]	SY	SI	KI	LQK-W	LQK-N	FFA	oil	MC
C	5	1349.5	1.04* [d]	0.54*	0.69	1.04*	1.90*	3.30*	0.42*
REP	2	1023.4	0.57	0.65*	0.37	0.57*	0.29	0.19*	0.05
C × REP	10	696.5	0.82*	0.27*	0.39	0.82	0.24	0.17*	0.17*
FS	8	17914.8*	47.28*	19.86*	16.91*	47.28*	6.31*	2.09*	0.55*
P1	5	9273.2*	48.74*	21.85*	25.32*	48.74*	9.88*	3.30*	0.28*
P2	1	21609.0*	66.57*	30.32*	3.17*	66.57*	0.19	1.77*	0.58*
P1 - P2	1	3816.3*	4.17*	2.00*	0.12	4.71	0.09	1.84*	0.00
C × FS	40	1486.2*	0.34*	0.18*	0.40	0.34*	0.18	0.08*	0.04
C × P1	25	1373.4*	0.20	0.16	0.45	0.20*	0.15	0.05	0.04
C × P2	5	1788.9*	0.13	0.07	0.20	0.13	0.17	0.05	0.04
C × (P1 - P2)	5	1616.5*	0.46	0.23	0.24	0.13	0.28	0.09	0.03
error	87	660.3[e]	0.21[f]	0.10	0.35	37.30	0.13	0.05	0.03[g]

[a] Mean square for the extra DF for FS and the extra five DF for C × FS not listed due to lack of meaning regarding the experimental objective. [b] C, cultivar; FS, fruiting site; P1, position 1; P2, position 2; P1 - P2, difference between P1 and P2. [c] DF, degrees of freedom. [d] *, significant at $\alpha = 0.05$. [e] Error df = 96. [f] Error df = 88. [g] Error df = 86.

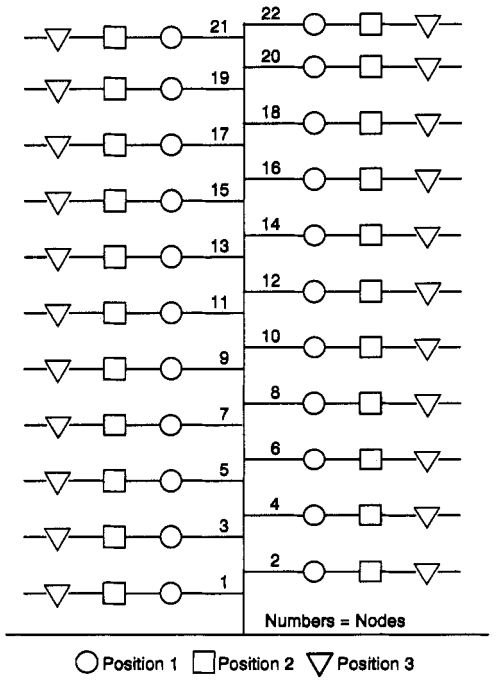

Figure 1. Schematic diagram of a cotton plant indicating expected fruiting sites: position and nodes with respect to main stem.

Seed Analysis. On receipt at SRRC, the seeds were stored in paper bags in an environmental room at 4 °C. All analyses were done in triplicate. Before any seeds were removed for analysis, each of the samples from the selected FS and cultivar was mixed by hand, quartered, and remixed five times. When necessary, seeds from selected FS were combined to provide a representative and sufficiently large sample for analysis. The seed characteristics measured were as follows: seed index, weight of whole seed, i.e., linters or fuzz, hull plus kernel (embryo); kernel index, weight of kernel; low-quality kernel number and weight; microbial content; oil content of the kernel; and free fatty acid content of the oil.

Three subsamples of 100 seeds were collected from each replicate cultivar and FS sample and weighed to obtain seed index (SI). One subsample was dehulled by hand to determine the weight of 100 kernels (KI). The kernels were separated into high-quality kernel (HQK) and low-quality kernel (LQK) fractions on the basis of visual observation. HQK were creamy-gray and filled the hull cavity. LQK were either discolored (brown to black) and fully formed or creamy and shriveled. The number (LQK-N) and weight (LQK-W) of LQK in each 100 kernels were measured. The HQK and LQP were recombined and then stored at −20 °C until analyzed.

To determine oil content (KO), the 100 kernels were suspended in petroleum ether (2.5 g/100 mL) in a cryovac bag. The oil was extracted in 2.5 min with a Stomacher 400 (Tekmar Co., Cincinnati, OH). Oil was separated from residual meal by filtration through fluted filters (qualitative grade 370, Baxter Health Care Corp., McGraw Park, IL). Comparative KO contents of these samples were determined by weighing the extracted oil. The free fatty acid (FFA) content of the oil was determined according to the official AOCS method (Ca 5a-40; AOCS, 1988) except that *m*-cresol purple was used as the indicator.

Another subsample of 100 seeds was used for determination of microbial contamination (MC) according to the method of Klich et al. (1984). The seeds were delinted with concentrated sulfuric acid and then rinsed three times with deionized water and air-dried. The dry delinted seeds were surface-sterilized by stirring for 2 min in an aqueous solution of sodium hypochlorite (2% w/v) containing 0.001% Triton X-100. Seeds were rinsed three times in sterile, deionized water. Using aseptic techniques, seeds were placed on malt dextrose agar Petri plates (100 × 15 mm), five seeds per plate. After incubation at 25 °C for 7 days, the number of seeds visibly contaminated with mold or bacteria was counted.

Statistical Analyses. A three-way analysis of variance (Milliken and Johnson, 1984) was conducted for all seed characteristics to determine how the fruiting sites behaved among the cultivars (C). The sources of variation composing this ANOVA model are listed in the first column of Table I. The effects of FS and of the C by FS interaction were examined in more detail by contrasts which considered only FS at P1 and P2 and the difference between the collective measurements made at P1 and P2. The C by replicate interaction was included to show that failure to randomize seed processing over time did not introduce a significant postharvest environmental condition effect. To satisfy the necessary assumptions for statistical analysis, data observed for four of the seed characteristics were transformed: LQK-W, $(\text{LQK-W} + 0.05)^{1/2}$; FFA, $(\%\ \text{FFA} + 0.5)^{1/2}$; KO, $(\%\ \text{KO})^{1/2}$; MC, arcsin $(0.01 \times \%\ \text{MC})^{1/2}$. Mean comparisons, appropriate for the significant effects occurring in the analysis of each seed characteristic, were conducted using pairwise comparisons of means or of least-squares means when sample sizes, on which means were based, differed (SAS, 1989).

RESULTS AND DISCUSSION

Total yields for the six cultivars were similar, ranging from 2056 kg ha^{-1} for DPL 50 to 2383 kg ha^{-1} for DES 119. In all cultivars, over 66% of the yield was from bolls located at P1. Samples from P1–N8,13 and P2–N6,11 had higher SI than those from higher positions on the plant (Table II). In all cultivars, the kernel represented more than 45% of the seed weight at all FS except P1–N18,22 and P2–N,12,22, where they averaged 38% and 43%, respectively. The highest LQK, by number and weight, were at P1–N8,9 and –N18,22 in all cultivars, but the actual values differed among the cultivars. For example, at these positions LQK-N values were 46% and 50% in DPL 20 but only 37% and 36% in DES 119. At P1–N8,9 the high amount of LQK probably resulted from exposure to weather in the field before harvest and may be associated

Table II. Seed Index (Grams per 100 Seeds) of Six Cotton Cultivars: Cultivar × Fruiting Site Interaction[a]

fruiting site							
position	node	ST 213	ST 506	ST 825	DPL 20	DPL 50	DES 119
1	8, 9	w 11.1 A	w 10.7 A	w 11.1 A	w 10.7 A	w 10.5 A	w 10.9 A
	10, 11	w 10.7 AB	w 10.9 A	w 11.2 A	w 11.2 A	w 10.1 A	w 10.8 AB
	12, 13	wx 10.4 ABC	w 10.4 A	w 11.0 A	w 10.7 A	wx 9.6 AB	w 10.7 AB
	14, 15	x 9.9 C	x 9.3 B	x 9.6 B	x 9.9 B	x 8.8 B	x 9.2 CD
	16, 17	y 8.6 D	y 8.4 BC	y 8.3 C	y 8.4 C	y 7.3 C	x 8.6 D
	18, 22	z 6.6 E	z 7.1 D	z 6.1 E	z 6.8 E	z 5.7 D	y 6.9 E
2	6, 11	w 10.1 BC	w 10.6 A	w 10.7 AB	w 11.0 A	w 9.9 A	w 9.9 BC
	12, 22	x 7.1 E	x 7.9 CD	x 7.6 CD	x 8.1 CD	x 6.9 C	x 7.6 E
3	6, 22	6.5 E	7.9 CD	6.6 DE	7.5 DE	7.5 C	7.1 E

[a] w–z: separate means comparison test was conducted for each cultivar among nodes at a fixed position. Node means at a fixed position and given cultivar are not statistically different ($\alpha = 0.05$) when associated with the same letter. A–E: separate means comparison test was conducted for each cultivar across nodes at all positions. Node means associated with the same letter for a given cultivar are not statistically different ($\alpha = 0.05$).

Table III. Oil Content (Percent) of Six Cotton Cultivars: Cultivar × Fruiting Site Interaction[a]

fruiting site							
position	node	ST 213	ST 506	ST 825	DPL 20	DPL 50	DES 119
1	8, 9	x 19.9 AB	xy 20.2 BCD	x 22.1 B	xy 24.2 AB	x 27.0 AB	x 27.1 AB
	10, 11	x 20.9 A	xy 21.7 ABC	x 24.7 AB	x 25.1 AB	xy 25.5 AB	x 27.3 AB
	12, 13	x 21.2 A	x 23.3 A	x 25.6 AB	xy 25.5 AB	xy 24.9 AB	x 28.6 A
	14, 15	x 20.0 AB	xy 21.9 ABC	x 24.4 AB	xy 24.9 AB	xy 25.4 AB	x 28.2 AB
	16, 17	x 19.5 AB	y 20.0 CD	x 22.3 AB	yz 22.6 BC	xy 20.3 ABC	x 25.7 AB
	18, 22	y 13.8 C	z 13.8 E	xy 15.2 C	z 17.3 C	y 15.9 C	y 22.4 C
2	6, 11	x 20.9 A	x 23.1 AB	x 27.9 A	x 26.6 A	x 28.6 AB	x 28.8 A
	12, 22	x 16.6 BC	y 18.6 D	x 23.6 AB	x 22.3 AB	x 20.5 ABC	x 25.3 BC
3	6, 22	16.3 BC	18.1 D	16.6 C	17.9 D	19.2 BC	17.9 D

[a] x–z: means comparison test for each cultivar among nodes at a fixed position. Node means at a fixed position and given cultivar are not statistically different ($\alpha = 0.05$) when associated with the same letter. A–E: means comparison test for each cultivar across nodes at all positions. Node means associated with the same letter for a given cultivar are not statistically different ($\alpha = 0.05$).

with the high free fatty acid content of the oil from these samples. At P1–N18,22, the LQK resulted from immature of incompletely developed seed and could be associated with the lower oil content in these samples.

In all cultivars, MC values of seeds from P1–N8,9, P1–N10,11, and P1–N12,13 were higher than that in seed from other nodes at P1. However, the MC was not consistent at any position from one replicate to another within a cultivar. For example, in ST 825, MC values of the replicates were 68%, 91%, and 81% at P1–N8,9 and 22%, 90%, and 55% at P1–N12,13. MC, therefore, does not appear to be a factor in the variations noted in other seed characteristics.

The most economically important portion of the cottonseed is the oil, and the highest yield of oil for all cultivars was produced at P1–N10,15 and P2–N6,11, but the Stoneville cultivars contained less oil than the early maturing cultivars (Table III). While the amount of oil is important, the quality of the oil is also important. One criteria of quality is the FFA content. To be certified as prime seed, the oil extracted from the seed must have a FFA content of 1.8% or less (*Trading Rules*, 1989). In all cultivars, prime seed was collected only from P1–N12,17 (Table IV). The overall level of FFA content, however, was lower in the Stoneville cultivars than in the early maturing cultivars (Table V). These generalizations based on physical and chemical measurements were corroborated by statistical analysis of the data.

Table II illustrates the behavior of seed index (SI) relative to cultivar and fruiting site. The highest SI occurred at P1–N8,13 for all cultivars and at P2–N6,11 for all cultivars except ST 213 and DES 119. The relative position of SI among nodes at a given position remains statistically constant for all cultivars, as illustrated by the letters w–z. However, when SI from nodes at all positions are compared, using the letters A–E, the SIs relative to one another show significant change ($p < 0.05$) from cultivar to cultivar. This explains why the overall C × FS interaction is significant, while there is no interaction between C and FS at a fixed position, C × P1, C × P2, or C × (P1 − P2) in Table I. Statistical analysis of kernel index showed that this parameter exhibited behavior identical to that of SI relative to FS and C.

Table IV. Average Free Fatty Acid Content of Oil in Seed from Selected Fruiting Sites in All Cultivars

fruiting site			fruiting site		
position	node	FFA[a]	position	node	FFA[a]
1	8, 9	11.62 A	2	6, 11	4.08 B
	10, 11	3.77 B		12, 22	3.82 BC
	12, 13	1.91 D			
	14, 15	1.20 D	3	6, 22	2.67 CD
	16, 17	1.84 D			
	18, 22	3.83 B			

[a] Percent, as oleic acid. FS averages with the same letter in a column are not significantly different ($p > 0.05$). Comparisons were based on standard errors of $(\%\ \text{FFA} + 0.5)^{1/2}$, which ranged from 0.086 to 0.093.

Table V. Average Free Fatty Acid Content of Oil in Seed from All Fruiting Sites in Each Cultivar

cultivar	FFA[a]	cultivar	FFA[a]
ST 213	3.31 B	DPL 20	5.40 A
ST 506	2.62 B	DPL 50	3.80 AB
ST 825	2.64 B	DES 119	5.38 A

[a] Percent, as oleic acid. Averages with the same letter are not significantly different ($p > 0.05$). Comparisons were based on standard errors of $(\%\ \text{FFA} + 0.5)^{1/2}$, which ranged from 0.096 to 0.215.

The kind of cultivar differences occurring in oil content, i.e., C by FS interaction, are illustrated in Table III. The relative amounts of oil content among nodes at a given position remain statistically constant across cultivars as shown by the letters x–z. However, when oil contents from nodes at all positions are compared, the oil contents relative to one another show significant change from cultivar to cultivar, indicated by the letters A–D. Seed from Stoneville cultivars contained significantly less oil at P1–N8,9 than seed from the other three cultivars. At P1–N12,13, ST 213 seed had a significantly lower amount of oil than DES 119 and the DPL cultivars. At P1–N18,22, seed from the Stoneville cultivars and DPL 50 had statistically similar oil contents. However, at this fruiting site all four of these cultivars contained significantly less oil than the DES 119 and DPL 20 cultivars. Cultivar differences were also apparent at P2. Oil contents of seed from P3 in all cultivars were, most often, significantly lower than that from seed at other positions on the plant.

The overall level of free fatty acid content of seed differed significantly among cultivars (Table I). The Stoneville cultivars contained significantly less FFA than the early maturing cultivars ($p < 0.05$) (Table IV). However, as evidenced by the absence of a significant C × FS interaction (Table I), differences in FFA content of seed among nodes at P1 remained constistent for all cultivars (Table V). In all cultivars, FFA content of seed from P1–N8,9 were exceptionally high, ranging from 7% to 17% (statistically higher than any other fruiting site). From the data on oil and FFA content, it is obvious that, although the Stoneville cultivars contained less oil, the FFA content of oil from these seeds were lower than that of the oil from the other three cultivars studied.

The most important quality characteristics for utilization of the cottonseed as a source of vegetable oil are high oil content and low FFA content. Using the data for yield of seed per plant and the yield and quality of oil for each cultivar, it was calculated that 1000 lb of crude oil from the Stoneville cultivars would contain 25 lb of FFA, whereas 1000 lb of crude oil from the early maturing cultivars would contain 50 lb of FFA. Therefore, while an evaluation of the early maturing cultivars based on oil content would make them more desirable than the three Stoneville cultivars, this advantage would be offset by the higher FFA content of the oil in the early maturing cultivars.

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