TWO YEAR SUMMARY OF THE AGRONOMIC PERFORMANCE OF ONE BOLLGARD II™ DONOR VARIETY J. S. Mahaffey, K. D. Howard, T. A. Kerby, J. C. Burgess, M. Casavechia and A. Coskrey Delta and Pine Land Company Scott, MS

Abstract

An experimental cotton variety, DPLX 985 EB was planted in a series of 19 trials across the cotton belt during the cropping seasons of 1999 and 2000. Trials were monitored via plant mapping at intervals through the season, machine picked yields, and fiber quality analyzed using HVI. The responses of DPLX 985 EB were well within the acceptable range in all measured categories, indicating that this line could be successfully used in varietal development programs for the future.

Introduction

Beginning with the 1996 cropping season, transgenic cotton plants containing the BollgardTM gene by Monsanto have been utilized by cotton growers across the U.S. cotton belt. These plants express a gene coding for the Cry IA(c) protein from the soil-borne bacterium *Bacillus thuringiensis* (Berliner) subsp. Kurstaki. This protein exhibits insecticidal properties against certain lepidopterous species including *Helicoverpa zea* (cotton bollworm) and *Heliothis virescens* (tobacco budworm) (Beegle and Yamamoto 1992). The adoption of this new class of transgenic pest control tools has enabled many growers to virtually eliminate chemical sprays associated with control of insecticide resistant pests such as *H. virescens* and to reduce costs associated with *H. zea* control (Mullins and Mills 1999).

One of the major concerns surrounding the utilization Bollgard containing cotton varieties in cotton cropping systems concerns the management of resistance to the Cry toxins. In fact, approval of the transgenic varieties by certain governmental agencies hinged on the development of a resistance management plan for these toxins. Plans were developed by Monsanto in conjunction with USDA and University scientists. These multi-faceted plans include restrictions on the ratio of Bollgard/non-Bollgard cotton which may be planted by a given grower, stipulations on management of the crops throughout the season, and monitoring of plan compliance (USDA 1999). The primary objective of this plan is to generate moths which are susceptible to the Cry toxin and allow those moths to mate with any resistant individuals which may emerge from the Bollgard fields.

One resistance management tactic which has been discussed by many authors is the utilization of multiple toxins with dissimilar modes and/or sites of action (i.e. insertion of multiple of traits in the same plant or "stacking") (USDA 1999). This use of two dissimilar toxins should greatly decrease the likelihood of resistance manifestation.

In trials planted during 1999 and 2000, D&PL endeavored to evaluate the agronomic performance of two cotton lines containing both the BollgardTM and the Bollgard IITM genes which could be used as donor parents in breeding programs to develop "stacked" cotton varieties.

Materials and Methods

The Bollgard II[™] gene was inserted into the commercially available cotton variety DP 50B via particle gun bombardment ("gene gun" insertion). Plants were regenerated from these transformations. All transformation and regeneration work was done by Monsanto. These plants were evaluated for gene purity and moved into self pollination and seed increase programs.

Upon availability of sufficient amounts of seed, trials were undertaken to compare the agronomic acceptability of DPLX 985 EB to currently available BG and conventional varieties. The trials described in this paper were performed for that purpose.

Trials were set up as randomized complete blocks with four replications at 19 locations across the cotton belt. Plot size ranged from small, research plot size (four rows by 30 feet long) to larger length-of-the-field sized (4 rows X 600 feet long) plots. All agronomic practices were performed as typical for the area in which the plot was planted. Non-lepidopteran insects were controlled in the plots. No treatments were made to lepidopteran pests.

Plant mapping was initiated on 5 plants from each plot at intervals throughout the season. The primary purpose of the plant mapping was to monitor varieties for aberrant growth characteristics and to measure varietal response to the testing environments of the 1999 and 2000 growing seasons.

Final data collection included machine picking, ginning in a commercialstyle gin, and HVI testing of fiber samples.

Results

Plant growth monitoring results are presented in tables 1 and 2 with appropriate statistics (means, probability levels, and LSD's). Significant differences existed among varieties in all growth parameters measured. However, none of the tested varieties deviated outside the normal range which could be expected among commercially available cotton varieties.

Fiber quality results are presented in tables 3 and 4 with appropriate statistics (means, probability levels, and LSD's). Significant differences existed among varieties when comparing micronaire, strength, and length. Probability levels for the variety by location interaction also indicate that location had a significant effect on fiber properties during the testing seasons and that not all varieties responded in a similar manner to a given environment.

The last line of tables 3 and 4 is labeled "Contrast DPLX 985 EB vs. DP 50 XX". This data is the result of the orthogonal contrast of mean fiber properties from DPLX 985 EB to the same properties of the varieties DP 50, DP 50 B, and DP 50 B/RR when grouped collectively, for a given characteristic. Note that some significant differences were indicated by this test among the groups. DPLX 985 EB had significantly higher micronaire and longer fiber that the collective DP 50 XX group. No significant difference was indicated when contrasting the fiber strength of the two groups. However none of the variety means are outside of the normal range of fiber quality found among cotton varieties.

Tables 5 and 6 contain the percent lint (turnout) and lint yield data with appropriate statistics (means, probability levels, and LSD's) from the trials. Note that significant differences were indicated among varieties when comparing both turnouts and lint yields. Also, the location by variety interaction indicates that not all of the tested varieties responded to the testing environments in a similar manner.

The last line of table 3 is labeled "Contrast DPLX 985 EB vs. DP 50 XX". This data is the result of the orthogonal contrast of mean turnouts and lint yields from DPLX 985 EB to the same properties of the varieties DP 50, DP

50 B, and DP 50 B/RR when grouped collectively, for a given characteristic. No significant differences were seen in lint turnouts among the groups. The contrast for lint yields indicates that the DPLX 985 EB variety, in both 1999 and 2000, yielded significantly more lint than the other "DP 50-type" varieties, as a group, which were included in the trials.

Summary

Throughout the course of these trials no unacceptable characteristics in plant growth, yield, or fiber properties were observed in any of the tested varieties. Although a range of response was measured in all of the measured parameters, none of the tested varieties fell outside of the acceptable range for commercial cotton varieties. Also, location by variety interactions were significant across almost every measured parameter. This indicates that the environment which a variety is planted into has a significant effect on the performance of that variety, in the parameters of plant growth, fiber quality, and yield.

DPLX 985 EB performed very well throughout this series of tests. In all of the measured characteristics this variety responded much the same as other non-BGII varieties. However, some significant differences were observed when comparing the DPLX 985 EB to the other "DP50-type" varieties. Even thought these varieties are very similar in their lineage, they were not identical in their response to these testing environments. This indicates that even though new transgenic, or for that matter, non-transgenic varieties, are derived from well known parents, adequate testing must be performed to accurately quantify their characteristics prior to commercial introduction.

The fact that DPLX 985 EB performed in a similar manner to other commercially available varieties in this series of tests indicates that it may successfully be used in future varietal development programs.

References

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Table 1. Plant growth monitoring results from 10 trials conducted by Delta and Pine Land Company for evaluation of BG II lines during 1999. All data are taken from end of the season plant maps.

Variety	Height ¹	#Nodes ²	# FB ³	# N95 ⁴	FR 95%
DPLX 985 EB	32.7	17.5	12.2	15.1	56.1
DP 450 B/RR	35.0	17.5	12.2	15.5	54.3
DP 50	33.8	17.6	12.2	15.7	47.2
DP 50 B	33.5	17.2	12.1	14.9	56.5
NuCOTN 33B	35.1	18.4	12.8	15.9	48.5
Variety p	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001
LSD p< 0.05	0.8	0.3	0.3	0.3	2.2

¹ Total plant height

² Total number of nodes

³ Total number of fruiting branches

⁴ Total number of nodes accounting for 95% of the harvestable yield

⁵ Percent first and second position fruit retention in the fruiting zone containing 95% of the harvestable yield

Table 2. Plant growth monitoring results from 8 trials conducted by Delta and Pine Land Company for evaluation of BG II lines during 2000. All data are taken from end of the season plant maps

Variety	Height ¹	#Nodes ²	# FB ³	# N95 ⁴	FR 95% ⁵
DPLX 985 EB	33.9	18.5	13.0	15.1	61.6
DP 450 B/RR	35.7	18.4	13.0	15.1	56.9
DP 50	35.3	19.1	13.6	15.7	49.8
DP 50 B	34.8	18.4	13.3	14.9	59.4
NuCOTN 33B	35.5	19.3	13.6	15.9	51.5
Variety p	0.0186	< 0.0001	0.0275	0.0001	< 0.0001
LSD p< 0.05	0.8	0.3	0.3	0.3	2.2

¹ Total plant height

² Total number of nodes

³ Total number of fruiting branches

 4 Total number of nodes accounting for 95% of the harvestable yield

⁵ Percent first and second position fruit retention in the fruiting zone containing 95% of the harvestable yield

Table 3. Fiber property results from 9 trials conducted by Delta and Pine Land Company for evaluation of BG II lines during 1999.¹

Variety	Micronaire	Strength	Length
DPLX 985 EB	4.51	29.15	1.16
DP 409 B/RR	4.16	28.81	1.14
DP 428 B	4.49	28.47	1.15
DP 450 B/RR	4.41	28.34	1.14
DP 451 B/RR	4.50	28.55	1.15
DP 50	4.27	28.99	1.14
DP 50B	4.25	29.28	1.17
NuCOTN 33B	4.39	30.11	1.14
PM 1218 BG/RR	4.55	28.14	1.10
PM 1560 BG/RR	4.05	29.83	1.15
SG 501 B/R	4.76	30.73	1.13
Variety - p value	< 0.0001	< 0.0001	< 0.0001
LSD (p<0.05)	0.09	0.33	0.007
V X L - p value	< 0.0001	0.0025	0.0001
Contrast DPLX 985			
EB vs. DP 50 XX ²	0.0001	0.1348	0.0180

¹ All fiber properties were derived via standard HVI testing.

 2 This statistic is an orthogonal contrast of the DPLX 985 EB mean value for a given parameter versus the varieties DP 50, DP 50 B, and DP 50 B/RR (collectively grouped as "DP 50 XX") for the same parameter. The value is the probability that the varieties are not different.

Table 4. Fiber property results from 10 trials conducted by Delta and Pine Land Company for evaluation of BG II lines during 2000.¹

Variety	Micronaire	Strength	Length
DPLX 985 EB	4.24	28.20	1.10
DP 428 B	4.15	27.59	1.10
DP 450 B/RR	4.23	27.71	1.09
DP 451 B/RR	4.09	27.98	1.10
DP 50	4.12	27.95	1.10
DP 50B	3.91	28.20	1.09
NuCOTN 33B	3.96	28.70	1.10
PM 1218 BG/RR	4.22	27.45	1.07
Variety - p value	< 0.0001	0.1093	0.2208
LSD (p<0.05)	0.10	NS	NS
V X L - p value	0.0003	0.3424	0.3252
Contrast DPLX 985			
EB vs. DP 50 XX ²	0.0090	0.4924	0.3370

¹ All fiber properties were derived via standard HVI testing.

 2 This statistic is an orthogonal contrast of the DPLX 985 EB mean value for a given parameter versus the varieties DP 50, DP 50 B, and DP 50 B/RR (collectively grouped as "DP 50 XX") for the same parameter. The value is the probability that the varieties are not different.

Table 5. Lint turnout and yield results from 9 trials conducted by Delta and Pine Land Company for evaluation of BG II lines during 1999.

Variety	% Turn Out ¹	Lint Yield
DPLX 985 EB	30.1	819
DP 409 B/RR	32.9	838
DP 428 B	31.2	818
DP 450 B/RR	30.0	793
DP 451 B/RR	30.7	824
DP 50	30.1	689
DP 50B	29.7	777
NuCOTN 33B	32.4	777
PM 1218 BG/RR	34.9	825
PM 1560 BG/RR	33.4	743
SG 501 B/R	33.3	824
Variety - p value	< 0.0001	< 0.0001
LSD (p<0.05)	0.34	32
V X L - p value	<0.0001	<0.0001
Contrast `DPLX 985		
EB vs. DP 50 XX ²	0.5781	0.0012

¹ Turnouts determined through ginning of plot samples.

 2 This statistic is an orthogonal contrast of the DPLX 985 EB mean value for a given parameter versus the varieties DP 50, DP 50 B, and DP 50 B/RR (collectively grouped as "DP 50 XX") for the same parameter. The value is the probability that the varieties are not different.

Table 6 Lint turnout and yield results from 10 trials conducted by Delta and Pine Land Company for evaluation of BG II lines during 2000.

Variety	% Turn Out ¹	Lint Yield
DPLX 985 EB	31.5	852
DP 428 B	33.3	819
DP 450 B/RR	31.84	861
DP 451 B/RR	32.82	835
DP 50	31.83	717
DP 50B	31.98	857
NuCOTN 33B	33.13	806
PM 1218 BG/RR	36.53	891
Variety - p value	< 0.0001	< 0.0001
LSD (p<0.05)	0.38	28
V X L - p value	0.0029	< 0.0001
Contrast `DPLX 985		
EB vs. DP 50 XX ²	0.1472	0.0231

¹ Turnouts determined through ginning of plot samples.

 2 This statistic is an orthogonal contrast of the DPLX 985 EB mean value for a given parameter versus the varieties DP 50, DP 50 B, and DP 50 B/RR (collectively grouped as "DP 50 XX") for the same parameter. The value is the probability that the varieties are not different.