TOLERANCE OF ROUNDUP READY COTTON TO TOPICAL AND POST-DIRECTED GLYPHOSATE

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Abstract

Roundup Ready cotton tolerance to topical and post-directed glyphosate applications was evaluated in a series of experiments conducted in 1997 to 2000 at three University of Arizona Experiment Stations. In the transgenic comparison experiments, glyphosate treated split-plots received a 0.75 lb ae/A topical application at the 2 to 4 true-leaf growth stage, followed by two 0.75 lb ae/A post-directed applications at the base of the cotton at the 10 to 12 node and 18 to 20 node growth stages. In side by side paired plots, there were no significant yield differences between unsprayed and sprayed varieties containing the Roundup Ready gene except for DP458BRR in 1999. On average, varieties containing the Roundup Ready gene yielded about 1% more than the conventional parent variety in these trials. Topical applications of glyphosate at 0.75 lb ae/A made before the fifth true leaf growth stage had no effect on cotton yield. Two sequential topical glyphosate applications at 0.75 lb ae/A with the second application made after the fifth true leaf growth stage had variable effects on cotton yield depending on the year and location. At Marana in 1999, a second 0.75 lb ae/A topical glyphosate application at the 7, 11, and 12 node growth stage did not significantly reduce yield, however, at MAC in 1997, 0.75 lb ae/A topical glyphosate applications made at the 6, 11, and 14 node growth stages reduced yield 5.9 (not statistically significant), 60, and 20.6%, respectively. In Marana in 1999 and 2000 and in Safford in 2000, post-directed glyphosate applications at labeled (0.75 lb ae/A) and "off-label" (1.12 and 1.5 lb ae/A) rates did not result in yield reductions. Similarly, in Marana and Safford in 2000, sloppy post-directed glyphosate applications at labeled (0.75 lb ae/A) and "off-label" (1.5 lb ae/A) rates did not result in yield reductions even when they followed an "off-labeled topical application (1.12 lb ae/A). In contrast, all treatments at Maricopa in 2000 with two or more glyphosate applications yielded significantly less than the unsprayed control (Table 4). These data indicate that Roundup Ready cotton varieties possess significant tolerance to "off-label" or above label rate glyphosate applications but that yield losses can result from such applications when environmental conditions limit mid-season and late-season boll production and retention (i.e., compensation).

Introduction

Roundup Ready cotton varieties exhibit excellent vegetative tolerance to postemergence glyphosate applications but glyphosate can cause abnormal anther development, inhibit pollen shed and self-pollination, and cause boll abortion. Traditional variety testing programs have compared conventional cotton varieties and transgenic varieties expressing Bt and Roundup tolerance traits with respect to agronomic traits such as yield and fiber quality. However, varieties containing the Roundup Ready gene are typically not sprayed in these experiments. Arizona growers have expressed concern about the agronomic performance of Roundup Ready varieties when they are sprayed with glyphosate and question whether their performance is as good as their conventional parent varieties.

The Roundup Ready cotton label specifies a maximum glyphosate rate of 0.75 lb ae/A, that topical applications can be made through the 4 true-leaf growth stage, that glyphosate applications made after the 4 true-leaf growth stage must be directed at the base of cotton plants, and that glyphosate can be applied several times during a season. Growers commonly ask several questions regarding the Roundup Ready cotton label such as: what are the consequences of application errors or "rescue" treatments that result in application rates greater than 0.75 lb ae/A? What are the consequences of making topical glyphosate applications after the 4 true-leaf growth stage? What are the consequences if post-directed sprays contact a substantial portion of the cotton foliage (i.e., a "sloppy" post-directed application is made)? And lastly, what are the cumulative effects of several glyphosate applications on the performance of Roundup Ready cotton varieties? To answer these questions, experiments were conducted in 1997 to 2000 to characterize the tolerance of Roundup Ready cotton varieties grown in Arizona to topical, post-directed, and "sloppy" post-directed glyphosate applications.

Materials and Methods

Roundup Ready cotton tolerance to topical and post-directed glyphosate applications was evaluated in a series of experiments conducted in 1997 to 2000 at three University of Arizona Experiment Stations; the Maricopa Agricultural Center (MAC) at an

elevation of 1,000 ft, the Marana Agricultural Center (MAR) at an elevation of 2000 ft, and the Safford Agricultural Center (SAC) at an elevation of 3,000 ft. Planting densities were high in the experiments and the fields were thinned to a uniform population of about 45,000 plants/A. Plots were typically 4 rows wide by 35 to 40 feet long depending on the location. The glyphosate formulation used in all experiments was Roundup Ultra with ammonium sulfate added at 8.5 to 17 lb/100 gallons of spray volume depending on location and water quality. Standard cotton production practices were used with respect to preplant preemergence herbicides, irrigation, cultivation, fertilization, insect pest management, defoliation and harvest. The experiments were hand weeded when necessary to maintain weed-free plots. The experiments were planted in April and the center two rows of each plot were plant mapped and then machine harvested using a spindle picker modified for harvesting plots in late October or November in each year. The data were analyzed using SAS.

The transgenic comparison experiments used an unbalance split-plot design where the main plots were "families" which consisted of all conventional, Roundup Ready (RR), Bollgard (Bt) and stacked gene (BRR) transgenic varieties derived from the same varietal background. The split-plots were the different cultivars within a family. For the Roundup ready (RR) varieties, both an unsprayed and sprayed split-plot were included in the main plots to allow paired plot comparisons. For example, the main plot containing the 'DP5415' family included split-plots of 'DP5415', 'NuCOTN 33B', 'NuCOTN 32B', 'DP448B', 'DP5415RR' sprayed, 'DP5415RR' unsprayed 'DP458BRR'and sprayed 'DP458BRR'. The glyphosate treated RR split-plots received a 0.75 lb ae glyphosate/A topical application at the 2 to 4 true-leaf growth stage, followed by two 0.75 lb ae/A post-directed applications at the 10 to 12 node and 18 to 20 node growth stages. The post-directed applications were made with two nozzles per crop row with the spray patterns contacting the cotton stem between the cotyledonary and first node (i.e., a "standard" post-directed application to the base of the plant according to the Roundup Ready cotton label).

The tolerance experiments evaluated the tolerance of selected Roundup Ready varieties to "off-label" applications where either the glyphosate rate or the amount of leaf surface contacted by the spray pattern or both were greater than specified on the Roundup Ready cotton label. The tolerance experiments used a randomized complete block design with 6 to 8 replications depending on location. Glyphosate was applied topically at 0.75 lb, 1.125 or 1.5 lb ae/A at the 2 to 4 leaf growth stage followed by two or more post-directed applications of glyphosate at 0.75, 1.125 or 1.5 lb ae/A depending on the location and treatment (see data tables for rates and timing of applications). In some treatments, a topical 0.75 lb ae/A glyphosate application at the 2 to 4 leaf growth stage was followed by a second topical 0.75 lb ae/A application at later growth stage (see data tables). The post-directed applications were either directed at the base of cotton plants with the spray patterns of nozzles on either side of the seed line intersecting at the cotyledonary to first node (i.e., a "standard post-direct application with minimal contact of foliage by glyphosate) or the spray patterns intersected about 6 inches above the bed-top (i.e., a "sloppy" post-direct application) leading to substantial glyphosate contact with cotton foliage.

Results and Discussion

The transgenic comparison treatments were summarized by pooling data across the three experimental sites by variety for each year (Table 1). In side by side paired plots, there were no significant yield differences between unsprayed and sprayed varieties containing the Roundup Ready gene except for DP458BRR in 1999. Eight of the 10 varieties containing the Roundup Ready gene produced lint yields that were very similar to the lint yields of the conventional parent varieties. One variety containing the Roundup Ready gene, DP420RR, was higher yielding ($p=0.10$) than its conventional parent and another variety with the tolerance gene, DP5690RR, was lower yielding ($p=0.05$) than its conventional parent. On average, varieties containing the Roundup Ready gene yielded about 1% more than the conventional parent in these trials.

In general, topical applications of glyphosate at 0.75 lb ae/A made before the fifth true leaf growth stage had no effect on cotton yield (Table 1). Two sequential topical glyphosate applications at 0.75 lb ae/A with the second application made after the fifth true leaf growth stage (equivalent to five node growth stage) had variable effects on cotton yield depending on year and location (Table 2). At MAR in 1999, a second 0.75 lb ae/A topical glyphosate application at the 7, 10.9, or 12.4 node growth stages did not significantly reduce yield. Similarly at MAC in 2000, a second topical glyphosate application at the 11 or 19 node growth stages did not significantly reduce yield. In contrast, at MAC in 1997, 0.75 lb ae/A topical glyphosate applications made at the 11, and 14 node growth stages significantly reduced yield 60, and 20.6%, respectively (Table 2). At MAR in 2000, yield was significantly reduced by a second 0.75 lb ae/A topical application at the 15 node growth stage. Plant mapping data from the latter two locations showed a change in boll distribution with topical applications made after the five node growth stage causing boll distribution to shift up the plant (i.e., bolls occurred on fruiting branches higher in the plant) and at fruiting positions further out on individual fruiting branches. The differences in yield results between the MAC in 1997 and the MAR in 1999 may have been due to the hotter weather at the MAC in 1997 that resulted in a lack of boll retention above about node 16 combined with early termination and harvest that year at MAC. In contrast, at MAR in 1999, late season boll retention was good so that late season lint production compensated for bolls aborted earlier in the season by topical glyphosate applications.

At all locations and in all years where the treatments were included in the experiments, a single topical 0.75 lb ae/A glyphosate application followed by two or three 0.75 lb ae/A post-directed glyphosate applications did not affect yield compared to a single topical application made prior to the 5 true-leaf stage except in the Maricopa 2000 experiment (Table 3 and 4). In Marana in 1999 and 2000 and in Safford in 2000, post-directed glyphosate applications at labeled (0.75 lb ae/A) and "off-label" (1.12 and 1.5 lb ae/A) rates did not result in yield reductions. Similarly, in Marana and Safford in 2000, sloppy post-directed glyphosate applications at labeled (0.75 lb ae/A) and "off-label" (1.5 lb ae/A) rates did not result in yield reductions even when they followed an "off-labeled topical application (1.12 lb ae/A). In contrast, all treatments at Maricopa in 2000 with two or more glyphosate applications yielded significantly less than the unsprayed control (Table 4). Similarly, in Marana in 2000 there was a numerical trend of reduced yield with increasing glyphosate exposure although the trend was not statistically significant (Table 4). These data indicate that Roundup Ready cotton varieties possess significant tolerance to "off-label" or above label rate glyphosate applications but that yield losses can result from such applications when environmental conditions limit mid-season and late-season boll production and retention (i.e., compensation).

Summary

In the transgenic comparison experiments, glyphosate treated split-plots received a 0.75 lb ae glyphosate/A topical application at the 2 to 4 true-leaf growth stage, followed by two 0.75 lb ae/A post-directed applications at the base of the cotton at the 10 to 12 node and 18 to 20 node growth stages. The transgenic comparison treatments were summarized by pooling data across the three experimental sites by variety for each year (Table 1). In side by side paired plots, there were no significant yield differences between unsprayed and sprayed varieties containing the Roundup Ready gene except for DP458BRR in 1999. On average, varieties containing the Roundup Ready gene yielded about 1% more than the conventional parent variety in these trials. Topical applications of glyphosate at 0.75 lb ae/A made before the fifth true leaf growth stage had no effect on cotton yield. Two sequential topical glyphosate applications at 0.75 lb ae/A with the second application made after the fifth true leaf growth stage (equivalent to five node growth stage) had variable effects on cotton yield depending on year and location. At Marana in 1999, a second 0.75 lb ae/A topical glyphosate application at the 7, 11, and 12 node growth stage did not significantly reduce yield, however, at MAC in 1997, 0.75 lb ae/A topical glyphosate applications made at the 6, 11, and 14 node growth stages reduced yield 5.9 (not statistically significant), 60, and 20.6%, respectively. Plant mapping data from the latter two locations showed a change in boll distribution with topical applications made after the five node growth stage causing boll distribution to shift up the plant (i.e., bolls occurred on fruiting branches higher in the plant) and at fruiting positions further out on individual fruiting branches.

In Marana in 1999 and 2000 and in Safford in 2000, post-directed glyphosate applications at labeled (0.75 lb ae/A) and "off-label" (1.12 and 1.5 lb ae/A) rates did not result in yield reductions (Table 3 and 4). Similarly, in Marana and Safford in 2000, sloppy post-directed glyphosate applications at labeled (0.75 lb ae/A) and "off-label" (1.5 lb ae/A) rates did not result in yield reductions even when they followed an "off-labeled topical application rate (1.12 lb ae/A). In contrast, all treatments at Maricopa in 2000 with two or more glyphosate applications yielded significantly less than the unsprayed control (Table 4). These data indicate that Roundup Ready cotton varieties possess significant tolerance to "off-label" or above label rate glyphosate applications but that yield losses can result from such applications when environmental conditions limit mid-season and late-season boll production and retention (i.e., compensation).

Table 1. Effect of a topical 0.75 lb ae/A glyphosate application followed by two post-directed 0.75 lb ae/A glyphosate applications on the yield of unsprayed and sprayed RR and BRR varieties pooled across Arizona locations by variety and year. There were no statistically significant yield differences between unsprayed and sprayed split-plots.

		Lint Yield (lb per acre)	
Year	**Variety**	**Unsprayed**	**Sprayed**
1999	DP5415RR	1260	1264
	DP458BRR	1624	1529*
	DP5690RR	1330	1310
	DP655BRR	1551	1557
	DP429RR	1608	1604
	DP409BRR	1764	1738
	DP422BRR	1465	1534
	PM1560BG	1658	1633
	SG125BRR	1623	1560
	SG501BRR	1554	1568
	STX9903	1780	1788
	STX9902	1739	1788
	1990 Average	1580	1569
2000	DP5415RR	1590	1479
	DP420RR	1630	1573
	DP655BRR	1363	1374
	DP451BRR	1347	1395
	SG521RR	1305	1351
	SG215BRR	1403	1422
	SG501BRR	1607	1583
	2000 Average	1464	1454

*Unsprayed and sprayed were significantly different at P=0.05.

Table 2. Effect of two sequential 0.75 lb ae/A topical glyphosate applications on the yield Roundup Ready cotton compared to the yield of the treatments receiving only the first topical application. The first topical glyphosate applications were made at the 2 to 4 true leaf growth stage. The Roundup Ready variety was DP458BRR with the exception of the 1997 experiment which used DP5690RR.

Location	**Year**	**Treatment**	**Percent of T4 yield**
Marana	1999	T4[1]	(1668 lb seed cotton/A)[2]
		T4 + T7.3	100
		T4 + T10.9	99
		T4 + T12.4	96
Marana	2000	T2	(1237 lb lint/A)
		T2 + T15	65*
Maricopa	1997	T4	(2017 lb seed cotton/A)
		T4 + T6	94
		T4 + T10	40*
		T4 + T14	80*
Maricopa	2000	T4	(1642 lb lint/A)
		T4 + T11	91
		T4 + T19	94

[1]T = topical; number indicates cotton growth stage in nodes.
[2]Numbers in parentheses are the yield of the treatments receiving a 0.75 lb ae/A topical glyphosate application.
*Indicates the percent of T4 yield is significantly reduced according to ANOVA and mean separation using Tukey's HSD.

Table 3. Effect of a topical glyphosate application at the 4 true-leaf growth stage followed by post-directed or sloppy post-directed glyphosate applications at various rates on the yield Roundup Ready cotton in 1999. The Roundup Ready variety was DP458BRR; yield values are lb of seed cotton/A.

Location/Year	Treatment	Yield (mean ± SD)
Marana 1999	unsprayed	1668 ± 215 a[1]
	T4(0.75)[2]	1670 ± 148 a
	T4(1.12) + PD7.3(0.75) + PD10.9(1.12)	1641 ± 201 a
	T4(1.12) + PD7.3(0.75) + PD10.9(1.12) + PD12.4(1.12)	1593 ± 154 a

[1] Values followed by the same letter within a location/year are not significantly different according to ANOVA and mean separation using Tukey's HSD.
[2]T indicates a topical application, PD indicates a post-direct application at the base of the plants, and SPD indicates a sloppy post-direct application. The number immediately following T, PD or SPD indicate the cotton growth stage in nodes. The values in parentheses indicate the glyphosate rate in lb ae/A that was applied.

Table 4. Effect of a topical glyphosate application at the 2 to 4 true-leaf growth stage followed by post-directed or sloppy post-directed glyphosate applications at various rates on the yield Roundup Ready cotton in 2000. The Roundup Ready variety used in all experiments was DP458BRR; yield values are lb of lint/A.

Location/Year	Treatment	Yield (mean ± SD)
Safford 2000	unsprayed	1414 ± 154 a
	T4(0.75)	1543 ± 163 a
	T4(0.75) + PD11(0.75) + PD15(0.75)	1517 ± 216 a
	T4(0.75) + SPD11(0.75) + SPD15(0.75)	1549 ± 136 a
	T4(1.12) + PD11(1.5) + PD15(1.5)	1486 ± 85 a
	T4(1.12) +S PD11(1.5) + SPD15(1.5)	1510 ± 150 a
Marana 2000	unsprayed	1202 ± 111 a
	T2(0.75)	1237 ± 58 a
	T2(0.75) + PD15(0.75) + PD20(0.75)	1223 ± 173 a
	T2(0.75) + SPD15(0.75) + SPD20(0.75)	1164 ± 132 a
	T2(1.12) + PD15(1.5) + PD20(1.5)	1184 ± 135 a
	T2(1.12) +S PD15(1.5) + SPD20(1.5)	1097 ± 91 a
Maricopa 2000	unsprayed	1741 ± 111 a
	T4(0.75)	1642 ± 173 ab
	T4(0.75) + PD11(0.75) + PD19(0.75)	1522 ± 88 bcd
	T4(0.75) + SPD11(0.75) + SPD19(0.75)	1373 ± 121 cd
	T4(1.12) + PD11(1.5) + PD19(1.5)	1366 ± 178 cd
	T4(1.12) + SPD11(1.5) + SPD19(1.5)	1344 ± 91 d

[1] Values followed by the same letter within a location/year are not significantly different according to ANOVA and mean separation using Tukey's HSD.
[2]T indicates a topical application, PD indicates a post-direct application at the base of the plants, and SPD indicates a sloppy post-direct application. The number immediately following T, PD or SPD indicate the cotton growth stage in nodes. The values in parentheses indicate the glyphosate rate in lb ae/A that was applied.