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WEED SCIENCE

Weed Response and Tolerance of Enhanced Glyphosate-Resistant Cotton to Glyphosate

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ABSTRACT

The commercial release of herbicide-resistant cotton (*Gossypium hirsutum* L.) has dramatically changed weed management practices. Roundup Ready cotton is tolerant of glyphosate applied topically through the 4-leaf stage. Cotton with enhanced glyphosate-resistance is available with Roundup Ready Flex cotton, which is tolerant of glyphosate from emergence until harvest. Studies were conducted with Roundup Ready Flex cotton to determine the response of cotton and weeds to single and multiple glyphosate applications, the effect of early season weed competition on cotton yield, and the best timing for glyphosate applications. No cotton injury or yield loss was observed with glyphosate applied past the 4-leaf stage. Any treatment combination with multiple postemergence (POST) glyphosate applications provided essentially complete control of Palmer amaranth (*Amaranthus palmeri* L.), sicklepod (*Senna obtusifolia* L.), pitted morningglory (*Ipomoea lacunose* L.), and annual grasses. Single glyphosate applications from the 4- to 12-leaf cotton stage provided inconsistent weed control, allowed for competitive losses, and provided variable cotton lint yield. Optimum weed control and lint yield was achieved with multiple glyphosate applications. Roundup Ready Flex cotton can provide producers with acceptable weed control without compromising cotton yield. Early season weed interference had inconsistent effects on cotton yield. Multiple postemergence glyphosate applications can preserve cotton yield potential with acceptable weed control.

The introduction of herbicide-resistant crops has dramatically changed weed management in crop production systems (Owen, 2000). Following the registration of glyphosate-resistant soybean (1996) and cotton (1997), this technology has been readily adopted by producers. Greater than 87% of soybean, 61% of cotton, and 17% of corn hectares were planted to herbicide-resistant cultivars during 2004 (USDA-ERS, 2005). The rapid adoption of herbicide-resistant crops by producers is linked to many factors, including economics, convenience, superior weed control, production flexibility, and promotion of conservation tillage practices (Dill, 2005). In addition to these producer-oriented factors, registration of glyphosate-resistant crops is practical because of the favorable environmental impact of the glyphosate molecule (Franz et al., 1997).

Glyphosate-resistant cotton (Roundup Ready, transformation event Coker 312-1445, henceforth referred to as RR) is only resistant to glyphosate during vegetative growth. Glyphosate is registered for topical application to this cotton from emergence through the 4-leaf stage. Thereafter, glyphosate must be applied as a directed spray to avoid contact with cotton foliage (Anonymous, 2006b). Previous research demonstrated that glyphosate applied topically or inaccurately directed after the 4-leaf stage could result in fruit abortion and yield reduction, which was later elucidated as resulting from morphological changes in reproductive structures and production of non-viable pollen (Jones and Snipes, 1999; Pline et al., 2001; 2002a; 2002b; Viator et al., 2003; 2004). A new transgenic glyphosate-resistant cotton with extended resistance during reproductive growth (Roundup Ready Flex Cotton, transformation event #MON88913, henceforth referred to as RRF) allows for topical application of glyphosate from crop emergence until 7 d prior to harvest (Anonymous, 2006a; 2006b). Research conducted at nine locations across the U.S. Cotton Belt found that RRF produced higher yields than RR cotton when treated topically with glyphosate four times at the 3-, 6-, 10-, and 14-leaf stages, and that fruit distribution and weight were similar regardless of glyphosate treatment (May et al., 2004).

Commercial release of RRF cotton cultivars provides producers with unmatched flexibility for weed management and production options. The maximum

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registered amount of glyphosate that can be applied to RR or RRF cotton per season is limited to 6.67 kg a.e. ha^{-1} (Anonymous, 2006b). The advantage of planting RRF cotton cultivars is the total amount of glyphosate that can be applied topically to the crop in season and number of timings of topical applications (May et al., 2004). Topical applications of glyphosate over RR cotton were limited to 3.15 kg ha^{-1} from ground cracking through the fourth true-leaf, while up to 5.04 kg ha^{-1} may be applied from ground cracking until 7 d before harvest on RRF cotton (Anonymous, 2006a; 2006b). Increased dosages and an extended application time are beneficial since glyphosate provides broad-spectrum control of many annual and perennial grasses, sedges, and broadleaf weeds (Askew and Wilcut, 1999; Burke et al., 2005; Corbett et al., 2004; Culpepper and York, 1998; 1999; Faircloth et al., 2001; Jordan et al., 1997; Scott et al., 2002; Shaw et al., 2001; Wilcut and Askew, 1999). Furthermore, RRF cotton provides growers the flexibility to mix other pesticides with glyphosate for increased management flexibility. Postemergence cotton herbicides, such as pyrithiobac and trifloxysulfuron, can be mixed with glyphosate to increase control of troublesome weeds (Branson et al., 2005). Scroggs et al. (2005) reported that glyphosate mixed with insecticides offers producers the ability to combine pest and crop management strategies and reduce application costs without sacrificing weed control.

The objectives of this research were as follows: 1) evaluate cotton and weed response to multiple topical applications of glyphosate in RRF cotton; 2) determine the effect of early season weed interference in RRF cotton on yield; and 3) determine what combinations, both number and timing, of glyphosate applications provide the best weed control and preserve cotton yield.

MATERIAL AND METHODS

Field studies were conducted in 2004 and 2005 at the Pee Dee Research and Education Center near Florence, SC. The experimental design was a randomized complete block with four replications. Soil at this site was a Norfolk loamy sand (fine-loamy, kaolinitic, thermic Typic Kandiudult) with <1% organic matter and a pH range of 5.8 to 6.0. Soil preparation included disking and smoothing of the soil followed by bedding prior to planting. Planting dates were 1 June 2004 and 19 May for the two studies in 2005. In 2004, and experimental Roundup Ready cultivar (event MON88913) with Bollgard was planted. In 2005, the cultivar for both tests was Deltapine 117 B2RF (Delta Pine and Land Co.; Scott, MS). The cotton was seeded at 13 seeds per meter of row. Plots were 9.1 m long and four 96-cm rows wide. Seed cotton was harvested from the two center rows. Seed cotton samples (1.12 kg) were hand ginned to determine lint percentage for each treatment. Weed populations averaged 20, 6, 8, and 23 plants per meter square for Palmer amaranth, sicklepod, pitted morningglory, and annual grasses, respectively. Clemson University Cooperative Extension Service recommendations were followed for management of fertility and insect pests (Jones et al., 2005).

Treatments were applied using a tractor-mounted compressed air sprayer with either flat-fan nozzles (TeeJet 8004 nozzles; Spraying Systems Co.; Wheaton, IL) or air induction nozzles (TeeJet 11002AI nozzles; Spraying Systems Co.) calibrated to apply 140 L of spray solution per hectare. Lay-by applications of locally used herbicides were made with a hooded sprayer calibrated to deliver 140 L/ha. Single applications of the potassium salt of glyphosate (Roundup WeatherMax; Monsanto Co.; St. Louis, MO) (0.84 kg ha^{-1}) were applied to 4-, 6-, 8-, 10-, and 12-leaf cotton. Sequential glyphosate applications were made to cotton at 4- and 8-leaf; 4- and 10-leaf; 4- and 12-leaf; 6-and 10-leaf; 6- and 12-leaf; 8- and 12-leaf; 4-, 8-, and12-leaf; 4-, 6-, 8-, and 10-leaf; 4-, 6-, 8-, and 12-leaf; 4-, 8-, 10-, and 12-leaf; 4-, 6-, 8-, 10-, and 12-leaf stages. A local standard treatment of pendimethalin (Prowl 3.3 EC; BASF Corp.; Research Triangle Park, NC) at 1.12 kg a.i. ha^{-1} plus fluometuron (Cotoran 4L; Griffin L.L.C.; Valdosta, GA) at 1.12 kg a.i. ha^{-1} applied premergence (PRE) followed by pyrithiobac (Staple 85WP; Valent Agricultural Products; Walnut Creek, CA) at 32 g a.i. ha^{-1} applied postemergence (POST) followed by prometryn (Caparol 4L; Syngenta Crop Protection; Greensboro, NC) at 0.56 kg a.i. ha^{-1} plus MSMA (MSMA; Helena Chemical Co.; Collierville, TN) at 2.24 kg a.i. ha^{-1} applied at lay-by and an non-treated check were included for comparison. Lay-by applications were made when cotton reached 10 nodes. Cotton was harvested at maturity with a spindle picker modified for small plot research.

Visual estimates of crop response and weed control were recorded throughout the season and prior to harvest. Foliar chlorosis, necrosis, and plant stunting were considered when making the visual evaluations of cotton response. Weed control was estimated using a scale of 0 to 100% with 0 = no weed control and

100 = complete weed control (Frans et al., 1986). Non-treated plots were excluded from the analysis of variance for crop response and weed control. Data were normally distributed with equal variance and arcsine square root transformation did not affect conclusions; therefore, non-transformed means are presented. The mixed procedure of SAS (ver. 8.1; SAS Institute; Cary, NC) was used to analyze data, and data were combined where appropriate. Year by treatment interactions for cotton yield were detected, so data is presented by year. Means were separated using Fisher's Protected LSD at $P \leq 0.05$.

RESULTS AND DISCUSSION

Cotton injury. Injury to cotton was not observed with any glyphosate application or combination of glyphosate applications past the 4-leaf stage in RRF cotton (data not shown). These results are similar to those reported by May et al. (2004).

Lint yield. Single applications of glyphosate provided variable weed control, which resulted in variable cotton lint yield (Table 1). Variability was due to several factors, such as emergence of weeds after treatment, weed competition, and incomplete control of larger weeds with some application timings. Consistent yield preservation was observed with most multiple glyphosate applications because of the elimination of the factors previously mentioned that contributed to the variability. The standard herbicide treatment was included only in 2005, so yield data is not presented for 2004. Regression analysis of cotton yield from single glyphosate applications to predict competitive yield loss was inconclusive ($R^2 = 0.27$, analysis not shown). Current research is focusing on determination of yield loss parameters and models for RRF cotton.

Data indicates that glyphosate applied topically to RRF cotton after the 4-leaf stage does not reduce lint yield, which is consistent with results from May et al. (2004). Glyphosate applied POST over-the-top to weed-free 12-leaf RR cotton resulted in a 19 and 14% yield loss compared with the weed-free non-treated cotton (Edenfield et al., 2005).

Weed control. Control of Palmer amaranth was nearly complete with any treatment containing multiple glyphosate applications (Table 2). Single applications of glyphosate at 4-, 6-, 8-, 10-, or 12-leaf cotton stages and the standard herbicide program provided incomplete and inconsistent control of Palmer amaranth. Variable control with single glyphosate applications was because of Palmer amaranth that emerged after early applications. Palmer amaranth is normally easy to control with glyphosate, but it is more difficult to control and interferes with glyphosate deposition on weeds lower in the crop canopy when plants become large (>45 cm) (Everitt et al., 2003; Grichar et al., 2004; Keeling et al., 2004; Kendig and Nichols, 2005; Nuti et al., 2003).

Table 1 Cotton lint yield from single and multiple glyphosate applications at different cotton growth stages

Cotton leaf stage[z]	Lint yield (kg/ha)[y]		
	2004	2005a	2005a
4	1190 b	820 cd	1520 ab
6	1560 a	770 d	1360 b
8	1170 b	1040 bcd	1410 ab
10	1150 b	1180 ab	1000 c
12	680 c	1000 bcd	910 c
4, 8	1370 ab	1110 abc	1390 ab
4, 10	1300 ab	1140 ab	1480 ab
4, 12	1300 ab	1230 ab	1580 ab
6, 10	1490 ab	1140 ab	1390 ab
6, 12	1400 ab	1200 ab	1480 ab
8, 12	1360 ab	1200 ab	1380 ab
4, 8, 12	1390 ab	1390 a	1380 ab
4, 6, 8, 10	1370 ab	1080 bc	1600 a
4, 6, 8, 12	1280 ab	1270 ab	1520 ab
4, 8, 10 , 12	1310 ab	1220 ab	1480 ab
4, 6, 8, 10, 12	1270 ab	1140 ab	1510 ab
Standard program[x]	---	1000 bcd	1500 ab
Untreated check	550 c	350 e	850 c

[z] Number of fully expanded leaves on the cotton plant at glyphosate application.

[y] Means within a column followed by the same letter are not significantly different according to Fisher's Protected LSD ($P < 0.05$).

[x] Local standard that includes pendimethalin and fluometuron applied preemergence, pyrithiobac applied postemergence, and prometryn and MSMA applied at lay-by.

Sicklepod response was similar to the response observed with Palmer amaranth. Single glyphosate applications at any cotton growth stage provided less than 82% control of sicklepod (Table 2). Discontinuous germination of sicklepod requires multiple herbicide applications to achieve an acceptable level of control. In this study, any treatment with multiple glyphosate applications gave 96% control or greater and the standard program using pyrithiobac POST and prometryn plus MSMA at lay-by provided similar control. Sicklepod has been reported to be controlled (88-96%) by glyphosate in RR cotton (Koger et al., 2005).

Table 2. End of the season weed response to single and multiple glyphosate applications at different cotton growth stages

Cotton leaf stage[z]	Control (%)[y]			
	AMAPA	CASOB	IPOLA	Annual grass
4	79 bcd	65 d	21 e	48 c
6	84 bc	71 d	58 cd	53 c
8	92 ab	81 bcd	79 abc	50 c
10	89 ab	77 cd	71 bcd	85 ab
12	71 d	77 cd	51 d	90 ab
4, 8	99 a	96 ab	94 a	97 a
4, 10	98 a	98 ab	96 a	98 a
4, 12	99 a	98 ab	90 ab	98 a
6, 10	98 a	98 ab	98 a	99 a
6, 12	99 a	98 ab	98 a	99 a
8, 12	98 a	98 ab	95 a	99 a
4, 8, 12	99 a	99 ab	99 a	99 a
4, 6, 8, 10	97 a	96 ab	98 a	99 a
4, 6, 8, 12	99 a	99 ab	99 a	99 a
4, 8, 10 , 12	99 a	99 ab	99 a	99 a
4, 6, 8, 10, 12	99 a	99 ab	99 a	99 a
Standard program[x]	74 cd	93 abc	91 ab	65 bc

[z] Number of fully expanded leaves on the cotton plant at glyphosate application.

[y] AMAPA (Palmer amaranth), CASOB (sicklepod), IPOLA (pitted morningglory), and annual grass (mix of large crabgrass and goosegrass). Means within a column followed by the same letter are not significantly different according to Fisher's Protected LSD ($P < 0.05$).

[x] Local standard that includes pendimethalin and fluometuron applied preemergence, pyrithiobac applied postemergence, and prometryn and MSMA applied at lay-by.

Pitted morningglory control was less than 80% with all single glyphosate applications regardless of treatment timing. Emergence of pitted morningglory after an early single glyphosate application, or the presence of large pitted morningglory at the time of a late single application of glyphosate contributed to reduced control. All treatments with multiple glyphosate application timings and the standard herbicide program provided acceptable control (≥90%). Flint et al. (2005) reported pitted morningglory populations decreased in the third year of a glyphosate study because of good control from glyphosate applications during the first two years of the study.

Annual grass, large crabgrass [*Digitaria sanguinalis* (L.) Scop.] and goosegrass [*Eleusine indica* (L.) Gaertn.], control was similar to the response of the other weeds in this study. Control with single applications of glyphosate was inconsistent. Annual grasses typically emerged after rainfall events, and glyphosate does not have any soil residual activity to control these grasses. Multiple glyphosate applications provided essentially complete annual grass control. Glyphosate mixed with *S*-metolachlor increased control of late season annual grasses 14 to 43 percentage points compared with control by glyphosate alone in one North Carolina study (Clewis et al., 2006). Results of this study are similar to those reported in another North Carolina study in which annual grasses were completely controlled by applications of glyphosate (Parker et al., 2005).

Results from these studies confirm those of May et al. (2004) that RRF cotton has enhanced resistance to glyphosate. The results of this study in relation to the stated objectives were as follows: 1) Roundup Ready Flex cotton can provide producers with acceptable weed control without compromising cotton yield, 2) early season removal of weed interference had inconsistent effects on cotton yield, and 3) multiple postemergence glyphosate applications can preserve cotton yield potential with acceptable (>90%) weed control. In the absence of glyphosate-resistant weeds (Culpepper et al., 2006; Main et al., 2004; Mueller et al., 2003; Scott, 2005; Van Gessel, 2001), multiple glyphosate applications can control Palmer amaranth, sicklepod, pitted morningglory, and annual grasses. While cotton yield potential may be preserved with less than complete weed control, consideration of the replenishment of the weed seed bank should be of some concern, especially with the development of herbicide resistance.

Reliance on glyphosate as a sole means of weed control is strongly discouraged because of the potential development of glyphosate-resistant weeds. Research from North Carolina found that herbicide systems in glyphosate-tolerant cotton that included soil-applied herbicides required one to two treatments of glyphosate POST and post-directed for season-long weed control and high cotton lint yields, whereas the same herbicide systems without soil-applied herbicides required two to three glyphosate treatments (Burke et al., 2005). Implementation of a proactive herbicide program (in terms of resistance management) that includes multiple herbicide modes-of-action, herbicides that have soil residual properties, and timely herbicide applications to small weeds will help preserve RRF cotton technology into the future.

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DISCLAIMER

Mention of a trademark, proprietary product vendor does not constitute a guarantee or warranty of the product by Clemson University and does not imply its approval to the exclusion of other products or vendors that may also be suitable.

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