Chapter IV

The Effect of Johnsongrass (*Sorghum halepense*) Control Method on the Incidence and Severity of Virus Diseases in Glyphosate-Tolerant Corn (*Zea Mays*)[1]

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Abstract: Field experiments were conducted in 2000 and 2001 in Virginia to evaluate the incidence and severity of maize chlorotic dwarf virus (MCDV) and maize dwarf mosaic virus (MDMV) in response to postemergence (POST) johnsongrass control in two corn hybrids. Previous research demonstrated increased disease severity in virus susceptible corn hybrids as an indirect effect of POST johnsongrass control. The increased disease severity resulted from greater transmission by insect vectors, which moved from dying johnsongrass to the crop. Recent observations have indicated a lack of virus tolerance in glyphosate-tolerant corn hybrids commercially available in Virginia.

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[3] Letters following this symbol are a WSSA-approved computer code from *Composite List of Weeds*, Revised 1989. Available only on computer disk from WSSA, 810 East 10th Street, Lawrence, KS 66044-8897.

A transgenic glyphosate-tolerant hybrid and a non-transgenic virus-tolerant hybrid, similar in growth characteristics and maturity, were subjected to POST treatments of nicosulfuron, while the glyphosate-tolerant hybrid was also treated with glyphosate. Both nicosulfuron and glyphosate, broadcast or directed, provided essentially complete johnsongrass control, although initial johnsongrass control was greater with glyphosate treatments. Little or no disease incidence occurred in the virus-tolerant hybrid. With the virus-susceptible hybrid, significant increases in disease incidence were observed in response to any herbicide treatment applied to johnsongrass-containing plots relative to the same treatment applied to weed free plots. Johnsongrass control with nicosulfuron or glyphosate caused similar disease incidence and severity in this hybrid, regardless of application method. Results of these experiments indicated that growers' choice of hybrid genetics should focus primarily on disease resistance rather than herbicide resistance in fields that are infested with johnsongrass.

Nomenclature: Glyphosate; nicosulfuron; johnsongrass, *Sorghum halepense* (L.) Pers. #[3] SORHA; corn, *Zea mays* (L). ZEAMX; maize chlorotic dwarf virus; maize dwarf mosaic virus.

Abbreviations: DAT, days after treatment, LSD, least significant difference; MAT, months after treatment; MCD, maize chlorotic dwarf; MDM, maize dwarf mosaic; PRE, preemergence; POST, postemergence; RR, roundup-ready; SS, southern states; WAT, weeks after treatment; WF, weed free.

INTRODUCTION

Johnsongrass [*Sorghum halepense* (L.) Pers.] was first introduced to the United States as a forage crop in the early 1800's (McWhorter 1989). As observed with many exotic species, johnsongrass spread quickly and has become one of the most common and difficult to control weeds in corn [*Zea mays* (L.)] in the southeastern United States (Dowler 1994). Johnsongrass can reduce corn yields by as much as 100% through competition for light and other resources (Bendixon 1986). Extensive production of rhizomes and prolific seed production contribute to the competitive nature of johnsongrass and to difficulty in control. Horowitz (1973) estimated that a single plant can produce up to 28,000 seeds, and Lolas and Coble (1980) determined a single plant could produce 40 to 90 m of rhizomes in a single season.

Selective herbicidal control of johnsongrass in corn first became possible with the introduction of safened EPTC (*S*-ethyl dipropylcarbamothioate) and butylate (*S*-ethyl bis(2-methylpropyl)carbamothiomate). These herbicides, however, are only partially effective for control of rhizome johnsongrass, and fair for control of seedling johnsongrass (Foy and Witt 1990). Glyphosate is effective for the control of johnsongrass prior to crop emergence or after crop harvest (Ghosheh and Chandler 1998; Brown et al. 1988). However, due to the nonselective nature of glyphosate activity, broadcast POST treatments in corn could not be applied. Selective POST herbicides for the control of johnsongrass in corn first became available with the introduction of primisulfuron and nicosulfuron. These herbicides were efficacious in the control of both seedling and rhizomatous johnsongrass (Foy and Witt 1990; Camacho et al. 1991;

Ghosheh and Chandler 1998; Gubbiga et al. 1995). Extensive field studies demonstrated that nicosulfuron was superior to primisulfuron for the control of rhizomatous johnsongrass (Foy and Witt 1990; Camacho et al. 1991). Limited phytotoxicity has been observed due to POST applications of nicosulfuron to corn, where a 6 to 9% yield reduction was observed when compared to a hand weeded control (Gubbiga et al. 1995). Yield, however, was increased 60 to 250% with nicosulfuron treatments compared to treatments where johnsongrass was not controlled (Gubbiga et al. 1995).

Recently, transgenic corn hybrids that tolerate POST applications of glyphosate have become commercially available. These hybrids were created through the incorporation of a glyphosate-tolerant 5-enolpyruvoyl-shikimate-3-phosphate synthase (EPSP synthase) gene into the corn genome (Pline 1999). These developments allowed glyphosate application for broad-spectrum weed control subsequent to crop emergence. Johnsongrass control was shown to be excellent (> 90%) with glyphosate applied at five weeks after corn emergence with minimal crop injury (Summerlin et al. 1999). Glyphosate and nicosulfuron provided essentially complete johnsongrass control, although initial johnsongrass control was greater with glyphosate treatments (King et al. 2000).

Johnsongrass harbors many crop pathogens, insects and viruses. A list compiled by McWhorter (1989) indicates that 4 bacterial species, 19 fungal species, 36 insect species, one mite species, 11 nematode species, and 11 viruses use johnsongrass as host. Control of johnsongrass has resulted in migration of some of these pests to crops. The movement and subsequent feeding of the blackfaced leaf-hopper [*Graminella nigrifrons* (Forbes.)] on corn has been determined to facilitate the transfer of maize chlorotic dwarf (MCD) virus from MCD infected johnsongrass plants (Bendixen 1988; Nault and Madden 1988; Madden 1990; Eberwine and Hagood 1995; Eberwine et al. 1998). Maize dwarf mosaic (MDM) virus, which can be transmitted mechanically or by insect vectors, has also been shown to increase in corn when johnsongrass is controlled (Eberwine 1995). MDM is transmitted by at least 25 species of aphids (Knoke et al. 1983).

MCD and MDM are two of the most devastating diseases of corn in the United States (Gordon et al. 1981). Symptoms of MCD include stunting, reddening and yellowing of leaves, leaf tearing and a diagnostic chlorosis of the tertiary leaf veins (Gingery and Nault 1990). MDM symptoms include chlorosis, mottling, and slight stunting (Gordon et al. 1981; Eberwine et al. 1998). Double infections of MDM and MCD are responsible for the most severe deleterious effects of disease development and yield reduction (Shurtleff 1980; Eberwine 1995). Plant breeders have recognized the significance of MCD and MDM and have actively pursued the development of corn hybrids that are resistant to these two pathogens (Louie 1990). This resistance is critical because viruses cannot be controlled chemically, and up to 20% of crop losses can be attributed to viral diseases as single or mixed infections (Marry 1994).

Previous research by Eberwine and Hagood (1995) and Eberwine et al. (1998) demonstrated a significant increase in MCD and MDM virus co-infections in corn as a direct result of johnsongrass control with nicosulfuron. In a virus susceptible hybrid, this increased co-infection was manifested in greater expression of disease symptoms and in reduced corn yield. The objective of these experiments was to determine if johnsongrass control with glyphosate would elicit a similar response in a glyphosate-tolerant virus-susceptible corn hybrid. Further, the objective was to compare the level and rapidity of control between nicosulfuron and glyphosate, and to determine if these factors influenced disease incidence and severity.

Materials and Methods

Field experiments were established in 2000 and 2001 in Virginia. In 2000, the experiment was conducted in Montgomery County on a Ross silt loam soil (fine-loamy, mixed, mesic *cumulic Hapludolls*) with 2.6% organic matter and pH 6.1. In 2001, the experiment was located in Roanoke County on a Speedwell sandy loam soil (fine-loamy, mixed, mesic *fluventic Hapludolls*) with 2.1% organic matter and pH 6.2. These locations were chosen because of heavy natural infestations of rhizomatous johnsongrass, which had shown symptoms consistent with MDM and MCD double infection (Eberwine and Hagood 1995). A transgenic glyphosate-tolerant hybrid (Southern States[3] 720) and a non-transgenic hybrid (Southern States 943), similar in growth characteristics and maturity, were subjected to POST treatments of nicosulfuron, while the glyphosate-tolerant hybrid was also treated with glyphosate. The Southern States 943 hybrid was chosen for its virus tolerance. The experiments consisted of 13 treatments and included POST nicosulfuron and glyphosate applied both broadcast, post directed, and broadcast to a weed free plot. All nicosulfuron and glyphosate treatments were applied at 35 g ai/ha and 1120 g ai/ha, respectively. Directed treatments were applied to determine if any crop response resulted directly from herbicides, rather than from virus disease. Additional treatments included a weed free control maintained by hand removal of all weeds throughout the growing season, and a weedy control. At planting, the entire experimental area was treated with 1.1 kg ai/ha of *s*-metolachlor and 1.6 kg ai/ha of atrazine for Italian weed control. Experiments were conducted in a split-plot, randomized complete block design with corn hybrid as the main plot and herbicide treatment as the subplot. In each experiment, dependent variables included johnsongrass control, virus incidence, virus severity, and corn yield.

Individual plots were 3.1 m wide by 9.1 m long containing four corn rows. The center 1.8 m of each plot containing the two center corn rows were treated, leaving a 1.2 m non-sprayed area containing two corn rows between each treated plot. Herbicide treatments were applied using a CO_2 backpack sprayer at a volume of 210 L/ha with flat-fan spray tips[4]. All nicosulfuron treatments contained 0.5% V/V of a nonionic surfactant[5]. Corn was planted following conventional tillage in mid-May with a population of 69,200 seeds per hectare in 76 cm rows using a two-row no-till planter[6] in both years, and fertilized with 154 kg/ha of nitrogen when the corn was 10 to 15 cm tall.

POST herbicide treatments were applied 1.5 months after planting when johnsongrass plants were 10 to 20 cm tall and corn plants were 10 to 15 cm tall. Herbicide efficacy ratings for johnsongrass control were made throughout the growing season. A 0-100 rating scale was used, where 0 was equal to the johnsongrass population observed in the weedy control plots and 100 was equal to compete johnsongrass control. The incidence

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[3] Southern States, Crops Division, P.O. box 26234, Richmond, VA. 23260

[4] Teejet 8003 flat fan spray nozzles, Spraying Systems Co., North Ave. Wheaton, IL. 60188

[5] X-77, non-ionic surfactant, Valent USA Corp. 1333 N. California Blvd., Walnut Creek, CA. 94596

[6] Almaco planter, 99 M Avenue, Nevada, Iowa 50201

and severity of virus diseases were also evaluated as symptoms became evident. Virus incidence indicated the percentage of corn plants in a plot showing any disease symptoms, and virus severity indicated the extent of symptom development within the infected plants relative to healthy plants in the same plot. The two center corn rows of each plot were harvested by hand at maturity, and grain moisture and yields were determined for each plot. To determine significance of main effects and interactions for treatment and year, corn hybrid and year, disease incidence or severity and hybrid, and incidence or severity and year, factorial analyses were performed. Johnsongrass control ratings, virus incidence, virus severity, and corn yields were subjected to analysis of variance and the means were separated using the least significant difference (LSD) test at the 5% level of probability.

Results and Discussion

Homogeneity of variance evaluation indicated significant interactions of year with respect to virus incidence and virus severity at 8 and 10 WAT, and for corn yield. Therefore, results will be presented individually by years. Significance of main effects and interactions were determined for each dependant variable. Where no significant interaction was observed, means will be presented.

Johnsongrass control. In both years, johnsongrass control with glyphosate, either broadcast or directed, was greater at 10 days after treatment (DAT) than with nicosulfuron (Table 1). Johnsongrass control at 1 month after treatment (MAT), however, was excellent with both nicosulfuron and glyphosate. Regardless of application method, 100% and 92% or greater johnsongrass control was observed with these herbicides in 2000 and 2001, respectively, and no significant differences were observed between herbicides. No significant differences between glyphosate and nicosulfuron, or within an individual herbicide between application methods occurred with respect to johnsongrass control in either year at 8 WAT (Table 2).

Virus disease incidence and severity in the virus-susceptible corn hybrid. Eberwine et al. (1998) indicated that virus diseases were initially detected 14 DAT in treatments where johnsongrass was controlled. Therefore, as a function of the rapidity of johnsongrass control between the two herbicides, disease development in virus-susceptible corn was expected to be more rapid in treatments which contained glyphosate compared to those which contained nicosulfuron. Earlier johnsongrass control in plots treated with glyphosate would cause the insects which vector these diseases to migrate to corn sooner than they would in plots treated with nicosulfuron, and result in earlier expression of virus symptoms in corn. In previous research, earlier disease infection resulted in greater yield reductions (Genter et al. 1973; Rosenkranz and Scott 1978; Scheifele 1969). Yield reduction in corn was also expected to be greater in plots treated with glyphosate due to earlier infection; this did not occur, however. The symptoms of disease infection first became evident at approximately 3 WAT and levels of disease incidence and severity were not significantly different between herbicides in either year at 3 WAT (Data not shown).

Disease incidence in both years did not differ between nicosulfuron and glyphosate applied either broadcast or directed (Table 2). Disease incidence in 2001 was not different between herbicides applied to weed-free plots. In 2000, however, nicosulfuron applied as a broadcast treatment to weed-free plots caused higher disease incidence than glyphosate applied to weed-free plots (Table 2). This result was not expected and the cause of the higher disease incidence in 2000 cannot be explained. Typically, no significant difference in disease incidence occurred when either herbicide was applied to a weed free plot compared to either the weed free or weedy control. The absence of johnsongrass in the weed-free plots theoretically lowers vector populations and reduces the potential for vectors to migrate to the corn crop. In general, virus-susceptible corn plants that became diseased reacted similarly with respect to virus severity when treated with either herbicide regardless of application method, and reacted similarly if either herbicide was applied to a weed-free plot at 8 WAT (Table 2). Higher levels of disease severity were generally observed in response to any control method where johnsongrass was controlled relative to the weedy control plots. Overall, these results indicate that there was not a significant difference between glyphosate and nicosulfuron with respect to johnsongrass control, disease incidence and disease severity.

Hybrid effects. Johnsongrass control with nicosulfuron and glyphosate was similar, and the resultant development of virus disease was the same for either herbicide in the virus-susceptible hybrid. For this reason, the effects of the corn hybrid variable on disease incidence and severity and on corn yield will be discussed only with reference to johnsongrass control with nicosulfuron.

Disease Incidence. In 2000 and 2001, significantly higher levels of virus incidence and virus severity occurred in the virus-susceptible hybrid compared to the virus-tolerant hybrid when any control method containing nicosulfuron was utilized (Tables 3 and 4). Virus incidence decreased from 41 and 31% to 14% when nicosulfuron was applied to a weed free plot compared to nicosulfuron applied either broadcast or directed, respectively (Table 4). In both years, lower levels of virus incidence in the virus-susceptible hybrid were observed in the weed free control, weedy control, and where nicosulfuron was applied to a weed free plot compared to incidence in corn where nicosulfuron was applied either broadcast or directed to plots containing johnsongrass (Tables 3 and 4). No difference in virus incidence between hybrids was observed with either of the controls in either year. No virus was observed in the virus-tolerant hybrid in 2000 (Table 3). In 2001, relatively low levels of disease were observed in the virus-tolerant hybrid, where all control methods resulted in less than 10% virus incidence (Table 4).

Disease severity. Disease severity in the virus-susceptible hybrid was 30-40% and 20-25% higher in plots where johnsongrass was controlled compared to the weedy control in 2000 and 2001, respectively (Tables 3 and 4). Increases in virus severity of approximately 35-55% in 2000 and approximately 30% in 2001 occurred in the virus-susceptible hybrid compared to the virus-tolerant hybrid when an effective johnsongrass control method was implemented.

Corn Yield. The virus-tolerant hybrid in 2000 produced 2576 kg/ha more grain compared to the virus-susceptible hybrid when yields were averaged over control methods (Table

3). Corn yield between hybrids in 2001, however, were not significantly different even though virus incidence and severity were greater in the virus-susceptible hybrid (Table 4). This result indicates that non-infected virus-susceptible corn plants in 2001 produced enough grain to offset the yield loss incurred by plants that were infected. Corn yield was lower when nicosulfuron was broadcast to plots containing johnsongrass compared to nicosulfuron applied to a weed-free plot or the weed-free control (Table 4).

The results of these experiments indicated that there was no difference between nicosulfuron and glyphosate with respect to johnsongrass control, corn yield, or the development and severity of virus diseases when either of these herbicides was applied to plots containing johnsongrass. These results are consistent with those observed by Eberwine and Hagood (1995) in which disease severity was significantly higher in corn hybrids that do not have genetic resistance to MCDV and MDMV. Significant increases in disease incidence occurred in response to any herbicidal treatments applied to johnsongrass containing plots relative to the same herbicidal treatments applied to weed-free plots in the virus-susceptible hybrid. Differences in virus incidence and virus severity between hybrids, however, were generally significant. Levels of disease incidence and severity regardless of application method or herbicide in the virus-susceptible corn hybrid were consistently higher than that observed in the virus-tolerant hybrid. It is concluded that choice of hybrid genetics in fields infested with johnsongrass should be focused primarily on disease resistance rather than herbicide resistance.

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Table 1. The effect of postemergence glyphosate and nicosulfuron applications on johnsongrass control at 10 DAT and at 1 MAT[a].

Treatment	Johnsongrass Control			
	2000		2001	
	10 DAT	1 MAT	10 DAT	1 MAT
	------------------------------ % ------------------------------			
Nicosulfuron[b]				
Broadcast	64 b	100 a	83 b	93 ab
Directed	69 b	100 a	79 bc	93 ab
Glyphosate				
Broadcast	88 a	100 a	99 a	99 a
Directed	93 a	100 a	92 a	92 ab
LSD (0.05)	6	3	9	8

[a] Values within a column followed by the same letter do not significantly differ, LSD, p = 0.05

[b] Nicosulfuron and glyphosate applied at 35 and 1120 g ai/ha, respectively.

Table 2. The effect of postemergence glyphosate and nicosulfuron applications on johnsongrass control, disease incidence, and disease severity within SS 720 RR hybrid in 2000 and 2001 at 8 WAT[a].

	2000			2001		
Treatment	SORHA	Disease		SORHA	Disease	
	Control	Incidence	Severity	Control	Incidence	Severity
	------------------- % -----------------			------------------- % -----------------		
Nicosulfuron[b]						
Broadcast	100 a	26 a	31 b	94 a	39 ab	48 a
Directed	97 b	21 a	49 ab	95 a	29 bc	41 a
Broadcast WF[c]	100 a	23 a	45 ab	98 a	14 d	39 ab
Glyphosate						
Broadcast	100 a	16 a	38 b	99 a	46 a	51 a
Directed	99 ab	29 a	60 a	92 ab	26 c	43 ab
Broadcast WF	99 ab	6 bc	26 bc	99 a	14 d	40 ab
Controls						
Weed free	99 ab	7 bc	45 ab	99 a	6 d	31 bc
Weedy	0 c	1 c	8 cd	0 c	5 d	21 cd
LSD (0.05)	2	12	20	7	11	14

[a] Values within a column followed by the same letter do not significantly differ, LSD, p = 0.05

[b] Nicosulfuron and glyphosate applied at 35 and 1120 g ai/ha, respectively.

[c] Abbreviation: WF, weed free

Table 3. Influence of corn hybrid and johnsongrass control method on virus incidence, virus severity at 10 WAT, and corn yield-2000[a].

	Incidence		Severity		Yield		Yield
Treatment	SS RR 720	SS 943	SS RR 720	SS 943	SS RR 720	SS 943	Mean
	----------------- %	-----------------			------ kg/ha[b]	------	--- kg/ha ---
Nicosulfuron[c]							
Broadcast	30	0	36	0	4476	7479	5977
Directed	24	0	54	0	4998	7573	6287
Broadcast WF[d]	16	0	53	0	4713	7615	6164
Controls							
Weed free	8	0	46	0	5239	7594	6419
Weedy	1	0	8	0	4412	6461	5439
Mean[e]	-	-	-	-	4768	7344	

[a] LSD (á = 0.05) for incidence for comparison of hybrids within a control method = 9
LSD (á = 0.05) for incidence for comparison of control methods within an individual hybrid = 8
LSD (á = 0.05) for severity for comparison of hybrids within an individual control method = 10

LSD (á = 0.05) for severity for comparison of control methods within an individual hybrid = 15
LSD (á = 0.05) for yield for comparison of hybrids averaged over control methods = 764

[b] Yield calculated at 15 % moisture.

[c] Nicosulfuron and glyphosate applied at 35 and 1120 g ai/ha, respectively.

[d] Abbreviations: CM, control method; NS, not significant; WF, weed free.

[e] Means only calculated when a nonsignificant interaction of hybrid by control method was observed.

Table 4. Influence of corn hybrid and johnsongrass control method on virus incidence, virus severity at 10 WAT, and corn yield-2001[a].

	Incidence		Severity		Severity	Yield		Yield
Treatment	SS RR 720	SS 943	SS RR 720	SS 943	Mean	SS RR 720	SS 943	Mean
	------------------------------ %------------------------------				----- % -----	------------ kg/ha[b] ------------		--- kg/ha ---
Nicosulfuron[c]								
Broadcast	41	9	49	14	31	4603	4760	4679
Directed	31	5	43	13	28	5061	5205	5133
Broadcast WF[d]	14	3	41	10	26	6160	6016	6088
Controls								
Weed free	6	1	33	3	18	6957	5706	6334
Weedy	6	1	20	1	10	6767	4051	5409
Mean[e]	-	-	37	8		-	-	

[a] LSD (á = 0.05) for incidence for comparison of hybrids within a control method = 8
LSD (á = 0.05) for incidence for comparison of control methods within an individual hybrid = 6
LSD (á = 0.05) for severity for comparison of hybrids averaged over control methods = 18
LSD (á = 0.05) for yield for comparison of control methods averaged over hybrids = 1022

[b] Yield calculated at 15 % moisture.

[c] Nicosulfuron and glyphosate applied at 35 and 1120 g ai/ha, respectively.

[d] Abbreviations: CM, control method; NS, not significant; WF, weed free.

[e] Means only calculated when a nonsignificant interaction of hybrid by control method was observed.