

Research Article

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Nomenclature:

Atrazine; bentazon; bromoxynil; glufosinate-ammonium; tolpyralate; kochia, *Kochia scoparia* L.; redroot pigweed, *Amaranthus retroflexus* L.; barnyardgrass, *Echinochloa crus-galli* (L.) P. Beauv.; horseweed, *Erigeron canadensis* (L.) Cronq.; common ragweed, *Ambrosia artemisiifolia* L.; waterhemp, *Amaranthus tuberculatus* var. *rudis*; foxtails, *Setaria* spp.; corn, *Zea mays* L.


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Sensitivity of two corn hybrids to tolpyralate plus reactive oxygen species-generating herbicides

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Abstract

Herbicides that inhibit 4-hydroxyphenylpyruvate dioxygenase (HPPD) can be mixed with herbicides that generate reactive oxygen species (ROS) to enhance the spectrum, level, speed, and consistency of weed control efficacy; however, mixtures of these herbicides can increase corn injury. A total of five field trials were conducted from 2021 to 2023 in Ridgetown, Ontario, to determine the sensitivity of two corn hybrids ('DKC39-97' and 'B79N56PWE') to tolpyralate plus ROS-generating herbicides (atrazine, bromoxynil, bentazon, or glufosinate) applied postemergence at the recommended rate (1×) and sequentially to represent a spray overlap (2×) in the field. Tolpyralate plus atrazine, bromoxynil, bentazon, or glufosinate (2× rates) caused greater corn injury to DKC39-97 than B79N56PWE corn at 1, 2, and 4 wk after treatment (WAT). Tolpyralate plus atrazine, bromoxynil, bentazon, or glufosinate (2× rates) caused 38%, 36%, 29%, and 18% injury to DKC39-97 corn, but only 5%, 20%, 9%, and 2% injury to B79N56PWE corn, respectively at 1 WAT. Corn injury to both hybrids decreased over time with ≤2% injury at 8 WAT. Tolpyralate + atrazine, bromoxynil, or bentazon (2× rates) caused a 17%, 16%, and 13% height reduction, respectively, of DKC39-97 corn at 2 WAT; however, tolpyralate + glufosinate did not reduce DKC39-97 corn height. Tolpyralate + bromoxynil or bentazon (2× rates) caused a 12% and 10% height reduction of B79N56PWE corn, respectively, at 2 WAT; however, tolpyralate + atrazine or glufosinate did not reduce B79N56PWE corn height. Tolpyralate + atrazine or glufosinate (2× rates) caused a greater corn height reduction of DKC39-97 corn than B79N56PWE corn at 2 WAT. Grain yield was on average 2% lower from DKC39-97 than B79N56PWE corn. Tolpyralate + bromoxynil or bentazon (2× rates) caused 7% and 6% corn grain yield reduction compared to tolpyralate plus glufosinate (2× rate). Results indicate that tolpyralate + ROS-generating herbicides can cause corn injury, which is influenced by corn hybrid and ROS-generating herbicide. Corn producers need to consider the differential sensitivity of corn hybrids and ROS-generating herbicides when using an HPPD-inhibiting herbicide for weed management.

Introduction

Tolpyralate is a relatively new pyrazole herbicide (categorized by the Weed Science Society of America as a Group 27 herbicide) that is used by corn growers in North America to control problematic weeds, especially herbicide-resistant biotypes (Governa et al. 2022; Soltani et al. 2022; Tonks et al. 2015). Tolpyralate inhibits the 4-hydroxyphenylpyruvate dioxygenase (HPPD) enzyme, which is crucial for carotenoid synthesis in plants. The increase in reactive oxygen species (ROS) following the application of an HPPD-inhibiting herbicide leads to cell organelle membrane destruction, white bleaching, chlorosis, and eventually, death of susceptible plants (Ahrens et al. 2013; Governa et al. 2022; Hawkes 2012; Kikugawa et al. 2015).

Tolpyralate provides control of some broadleaf and grass weeds including kochia, horseweed, redroot and green pigweeds, waterhemp, foxtails, and barnyardgrass, including herbicide-resistant biotypes (Governa et al. 2022). Tolpyralate can be co-applied with other herbicides such as atrazine, acetochlor, and dimethenamid-p to increase the spectrum of weeds controlled, especially annual grass weed species (Osipitan et al. 2018; Soltani et al. 2023; Tonks et al. 2015). Tolpyralate is most effective when applied postemergence; especially when applied early postemergence, for weed control in corn and other crops (Kikugawa et al., 2015; Tonks et al. 2015). Tolpyralate is active at relatively low doses, does not persist in the soil, has limited volatility, and has little effect on nontarget organisms, which gives it desirable environmental attributes (Anonymous 2019; Tonks et al. 2015).

Normally, corn can rapidly metabolize HPPD-inhibitor herbicides through expression of the *Nsf1* gene (Osipitan et al. 2018; Tonks et al. 2015; Williams et al. 2024). However, recent studies have highlighted genetic vulnerability in certain corn hybrids leading to unexpected sensitivity

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to tolpyralate (Williams *et al.* 2024). Williams *et al.* (2024) studied six field corn inbreds and 43 sweet corn inbreds and found moderate to severe injury symptoms to some corn inbreds shortly after treatment with tolpyralate + atrazine (Williams *et al.* 2024). The underlying genetic factors responsible for this vulnerability remain an area of active research.

In Ontario, tolpyralate, due to its efficacious activity against some herbicide-resistant biotypes, especially multiple herbicide-resistant (MHR) waterhemp and MHR horseweed, has become a popular herbicide with corn producers in recent years. Earlier field trials with tolpyralate applied at rates up to two times greater than the suggested manufacturer rate applied postemergence have generally shown no, or minimal, corn injury (Fluttert *et al.* 2022a; Langdon *et al.* 2020a,b; Willemsse *et al.* 2021b). Metzger *et al.* (2018a) reported less than 10% corn injury with tolpyralate + atrazine applied postemergence at three times the labeled rate. However, other studies have shown as much as 12% corn injury at 1 wk after treatment (WAT) with tolpyralate applied postemergence (Soltani *et al.* 2023).

HPPD-inhibiting herbicides such as tolpyralate are often co-applied with ROS-generating herbicides such as atrazine to increase the spectrum of weeds to be controlled, elevate the level of weed control, accelerate the speed of weed control, and improve the consistency of weed control (Anonymous 2019; Kim *et al.* 1999; Langdon *et al.* 2021; Metzger *et al.* 2018a, 2019). Co-applying other ROS-generating herbicides such as bromoxynil, bentazon, or glufosinate in place of atrazine with tolpyralate can result in a synergistic improvement in weed control but it may also increase corn injury (Fluttert *et al.* 2022a,b; Metzger *et al.* 2018a,b). Little information exists to compare the addition of bromoxynil, bentazon, or glufosinate in place of atrazine with tolpyralate on corn injury.

The objective of this research was to determine the tolerance of two corn hybrids (DeKalb 'DKC39-97' and Brevant 'B79N56PWE') to tolpyralate (40 g ai ha⁻¹) + atrazine (560 g ai ha⁻¹), bromoxynil (336 g ai ha⁻¹), bentazon (1,080 g ai ha⁻¹) or glufosinate-ammonium (500 g ai ha⁻¹) applied once (1×) and sequentially to represent a spray overlap (2×) in the field.

Materials and Methods

A total of five field trials were conducted from 2021 to 2023 (one site in 2021, two sites in 2022, and two sites in 2023) at the University of Guelph Ridgetown campus (42.45°N, 81.88°W), in Ridgetown, Ontario. Seedbed preparation consisted of moldboard plowing in the autumn followed by cultivation with an S-tine cultivator with rolling basket harrows in the spring.

Trials were established as split plots with herbicide treatment as the whole plot factor and corn hybrid as the split-plot factor in a randomized complete block design with four replicates. Experiment treatments included a nontreated control, tolpyralate + atrazine (40 + 560 g ai ha⁻¹), tolpyralate + atrazine [40 + 560 g ai ha⁻¹ followed by (fb) 40 + 560 g ai ha⁻¹], tolpyralate + bromoxynil (40 + 336 g ai ha⁻¹), tolpyralate + bromoxynil (40 + 336 g ai ha⁻¹ fb 40 + 336 g ai ha⁻¹), tolpyralate + bentazon (40 + 1,080 g ai ha⁻¹), tolpyralate + bentazon (40 + 1,080 g ai ha⁻¹ fb 40 + 1,080 g ai ha⁻¹), tolpyralate + glufosinate-ammonium (40 + 500 g ai ha⁻¹), and tolpyralate + glufosinate-ammonium (40 + 500 g ai ha⁻¹ fb 40 + 500 g ai ha⁻¹). All treatments included methylated seed oil concentrate (MSO Concentrate[®]; Loveland Products Inc., Loveland CO) at 1% v/v. Plots that received the 2× herbicide rate were applied sequentially to represent a spray overlap (2×) in the field.

Plots were 8 m long and 3 m wide and consisted of two rows (0.75 m apart) of glyphosate/glufosinate-resistant DeKalb DKC39-97 (Bayer Crop Science, Calgary, AB) and two rows of glyphosate/glufosinate/2,4-D-resistant Brevant 'B79N56PWE' Enlist corn hybrid (Corteva Agriscience, Calgary, AB). The two hybrids are locally adapted from the largest corn seed companies in Ontario, one from Bayer Crop Science (DeKalb) and one from Corteva Agriscience (Brevant). To remove the confounding effects of weed interference, the entire experimental area was maintained weed-free during the growing season.

Herbicide treatments were applied postemergence to corn (at the V3 to V4 growth stage) with a CO₂-pressurized backpack sprayer calibrated to deliver 200 L ha⁻¹ at 240 kPa. The spray boom was 2.5 m long and had six ULD120-02 nozzles (Hypro, Pentair, New Brighton, MN) spaced 50 cm apart producing a spray width of 3.0 m.

Visible corn injury evaluations were completed at 1, 2, 4, and 8 WAT on a scale of 0 (no injury) to 100% (corn death). At 2 WAT, corn population (number of corn plants per 4 meters of row) and corn height (average of 10 plants per plot in centimeters per hybrid) were determined. At harvest maturity, the two rows of each hybrid were harvested with a small-plot research combine, and corn grain moisture content and mass were recorded. Corn yield was adjusted to 15.5% moisture.

Statistical Analysis

Data analysis was carried out using the GLIMMIX procedure with SAS software (SAS Institute Inc., Cary, NC). Model fixed effects consisted of herbicide treatment, corn hybrid, and their interaction; random effects were environment (location-year combinations), replicate within environment, and the interactions of herbicide treatment by replicate within environment and environment by herbicide treatment by corn hybrid. The assumptions of analysis were checked using the Shapiro-Wilk statistic, normal probability plots, chi-square/df ratio, and studentized residual plots. To be able to compare the two corn hybrids, corn population, height, and yield were expressed as a percent of the nontreated control, and all parameters were analyzed using a Gaussian distribution, with corn injury being arcsine square root transformed before analysis. The Tukey-Kramer adjustment was applied to pairwise treatment comparisons. Treatments with assigned values and zero variance were excluded from the analysis. However, the P-value generated in the LSMEANS output enabled comparisons of each least square mean to the value zero. Where needed, means were back-transformed for presentation.

Results and Discussion

Table 1 presents the main effects and interaction between corn hybrid and herbicide treatment for visible corn injury at 1, 2, 4, and 8 WAT, and relative corn population, height, and yield. There was an interaction for visible corn injury and height, so the simple effects are presented in Tables 2 and 3.

Visible Injury

Visible injury symptoms included temporary white bleaching and blotched chlorosis in the youngest corn leaves that had unfurled at the time of application, and stunted growth. At 1 WAT, tolpyralate + atrazine, bromoxynil, bentazon, or glufosinate at the 1× rate, caused 8%, 17%, 5%, and 3% injury, respectively, to DKC39-97 corn; and only 1%, 9%, 1%, and 0% injury, respectively, to B79N56PWE corn (Table 2). Tolpyralate + atrazine, bromoxynil, bentazon, and

Table 1. Response of two corn hybrids to tolpyralate plus reactive oxygen species-generating herbicides applied postemergence from five trials conducted in Ridgeway, Ontario, between 2021 and 2023.^{a-e}

Main effects	Rate	Visible Corn Injury				Relative Population	Relative Height	Relative Yield
		1 WAT	2 WAT	4 WAT	8 WAT			
	g ai ha ⁻¹	%						
Corn hybrid								
DKC39-97		17	13	6	1	99	92	98 b
B79N56PWE		4	3	1	0	98	96	100 a
Hybrid P-value		<0.0001	<0.0001	<0.0001	0.0001	0.1637	<0.0001	0.0436
Herbicide treatment								
Nontreated control		0	0	0	0	100	100	100 ab
Tolpyralate + atrazine	40 + 560	3	2	1	0	99	98	100 ab
Tolpyralate + atrazine fb tolpyralate + atrazine	40 + 560 fb 40 + 560	19	16	9	1	97	89	97 ab
Tolpyralate + bromoxynil	40 + 336	13	8	2	0	99	94	101 ab
Tolpyralate + bromoxynil fb tolpyralate + bromoxynil	40 + 336 fb 40 + 336	27	21	11	1	98	86	96 b
Tolpyralate + bentazon	40 + 1,080	3	2	1	0	99	96	101 ab
Tolpyralate + bentazon fb tolpyralate + bentazon	40 + 1,080 fb 40 + 1,080	18	12	5	0	98	89	97 b
Tolpyralate + glufosinate ammonium	40 + 500	1	1	0	0	97	100	101 ab
Tolpyralate + glufosinate ammonium fb tolpyralate + glufosinate	40 + 500 fb 40 + 500	8	6	2	0	99	96	103 a
Herbicide P-value		<0.0001	<0.0001	<0.0001	0.0068	0.5642	<0.0001	0.0043
Interaction								
Hybrid × herbicide P-value		0.0197	0.0013	0.0138	0.0368	0.9206	0.0219	0.3212

^aAbbreviations: fb, followed by; WAT, weeks after herbicide application.

^bParameters evaluated were crop injury and relative measures of population, height, and yield. Means for a main effect were separated only if the interaction involving the main effect was negligible.

^cMeans followed by the same letter within a column are not significantly different according to a Tukey-Kramer multiple range test at $P < 0.05$.

^dThe nontreated control was excluded from analysis due to zero variance; a comparison of herbicide treatments with the value zero was obtained using the LSMEANS table from the GLIMMIX procedure (SAS software; SAS Institute Inc., Cary, NC).

^eAll treatments included methylated seed oil (1 or 2% mL L⁻¹ for the labelled herbicide rate and twice the labelled rate, respectively).

glufosinate at the 2× rate caused 38%, 36%, 29%, and 18% injury, respectively, to DKC39-97 corn; and only 5%, 20%, 9%, and 2% injury, respectively, to the B79N56PWE hybrid (Table 2). Injury was greater to DKC39-97 corn than to B79N56PWE corn when tolpyralate + atrazine (1× and 2× rates), bromoxynil (2× rate), bentazon (2× rate), or glufosinate (2× rate) were applied. Tolpyralate + bromoxynil (2× rate) caused ≥20% visible injury to both DKC39-97 and B79N56PWE corn hybrids.

At 2 WAT, tolpyralate + atrazine, bromoxynil, bentazon, or glufosinate at the 1× rate caused 5%, 11%, 3%, and 2% injury, respectively, to DKC39-97 corn; and 0%, 5%, 1%, and 0% injury, respectively, to B79N56PWE corn (Table 2). Tolpyralate + atrazine, bromoxynil, bentazon, or glufosinate at the 2× rate caused 34%, 29%, 20%, and 13% injury to DKC39-97 corn; and 4%, 15%, 6%, and 1% injury to B79N56PWE corn (Table 2). Corn injury was greater in DKC39-97 corn than B79N56PWE corn with tolpyralate + atrazine (1× and 2× rates), bromoxynil (1× and 2× rates), bentazon (2× rate), or glufosinate (1× and 2× rates). Tolpyralate + bromoxynil at the 2× rate caused significant injury (≥15%) to both DKC39-97 and B79N56PWE hybrids.

At 4 WAT, tolpyralate + atrazine, bromoxynil, bentazon, or glufosinate at the 1× rate, caused 2%, 3%, 1%, and 0% injury, respectively, to DKC39-97 corn; and 0%, 1%, 0%, and 0% injury to B79N56PWE corn (Table 2). Tolpyralate + atrazine, bromoxynil, bentazon, and glufosinate at the 2× rate caused 19%, 16%, 10%, and 5% injury, respectively, to the DKC39-97 hybrid; and 2%, 7%, 2%, and 0% injury, respectively, to the B79N56PWE hybrid (Table 2). The injury was consistently greater in DKC39-97 corn than B79N56PWE corn with herbicides evaluated at the 2× rate. Tolpyralate + bromoxynil at the 2× rate caused 16% and 7% visible corn injury to DKC39-97 and B79N56PWE hybrids, respectively.

At 8 WAT, tolpyralate + atrazine, bromoxynil, bentazon, or glufosinate caused no visible corn injury to DKC39-97 corn (at the 1× rate) or to B79N56PWE corn (at the 1× and 2× rates; Table 2). Tolpyralate + atrazine, bromoxynil, bentazon, and glufosinate at the 2× rate caused minimal (0% to 2%) injury to DKC39-97 corn (Table 2).

Recent studies have shown similar differential sensitivity among corn hybrids treated with tolpyralate + ROS-generating herbicides. Williams et al. (2024) evaluated six field corn inbreds treated with tolpyralate and found three corn inbreds ('CIMMYT-CLWQHZN8', 'NPGS-CML 52', and 'MU-Mo25W') were severely injured when tolpyralate was applied postemergence at 39 g ai ha⁻¹, whereas the other three other corn inbreds ('HFS-LH197', 'HFS-LH198', and 'Pioneer-PHBB3') were only moderately injured when the same amount of tolpyralate was applied. Furthermore, Williams et al. (2024) observed similar differential sensitivity between sweet corn inbreds to tolpyralate with 40 sweet corn inbreds exhibiting severe injury symptoms and six inbreds exhibiting moderate injury symptoms when treated with tolpyralate applied postemergence at 39 g ai ha⁻¹. In a greenhouse study, Williams et al. (2024) found as much as 85% injury and 84% biomass reduction of some sweet corn inbreds ('XSEN18') 11 d after treatment with tolpyralate + atrazine + MSO applied postemergence at 39 g ai ha⁻¹; however, the injury was not present in other sweet corn inbreds. Williams et al. (2024) concluded that the sensitivity of XSEN18 corn inbreds to tolpyralate is not due to mutant *Nsf1* alleles that codes for CYP81A9 and is perhaps associated with another genomic variant (single gene at Chr05: 283 240–1 222 909 base pair interval), which may explain why this sensitivity was not identified during the breeding process.

In other studies, Osipitan et al. (2018) found no visible injury to glyphosate-resistant 'Pioneer 35F40' corn treated with various

Table 2. Visible injury of two corn hybrids 1, 2, 4, and 8 wk after treatment following tolpyralate + reactive oxygen species-generating herbicides applied postemergence for five trials conducted in Ridgetown, ON, between 2021 and 2023.^{a-c}

Herbicide treatment	Rate	Visible corn injury					
		DKC39-97			B79N56PWE		
	g ai ha ⁻¹	%					
1 WAT							
Nontreated control		0	a		0	a	
Tolpyralate + atrazine	40 + 560	8	bc	Y	1	ab	Z
Tolpyralate + atrazine fb tolpyralate + atrazine	40 + 560 fb 40 + 560	38	e	Y	5	bc	Z
Tolpyralate + bromoxynil	40 + 336	17	cd		9	cd	
Tolpyralate + bromoxynil fb tolpyralate + bromoxynil	40 + 336 fb 40 + 336	36	e	Y	20	d	Z
Tolpyralate + bentazon	40 + 1,080	5	bc		1	ab	
Tolpyralate + bentazon fb tolpyralate + bentazon	40 + 1,080 fb 40 + 1,080	29	de	Y	9	cd	Z
Tolpyralate + glufosinate ammonium	40 + 500	3	b		0	a	
Tolpyralate + glufosinate ammonium fb	40 + 500 fb	18	cd	Y	2	bc	Z
Tolpyralate + glufosinate ammonium	40 + 500						
2 WAT							
Non-treated control		0	a		0	a	
Tolpyralate + atrazine	40 + 560	5	bcd	Y	0	a	Z
Tolpyralate + atrazine fb tolpyralate + atrazine	40 + 560 fb 40 + 560	34	f	Y	4	b	Z
Tolpyralate + bromoxynil	40 + 336	11	cde	Y	5	bc	Z
Tolpyralate + bromoxynil fb	40 + 336 fb	29	f	Y	15	c	Z
Tolpyralate + bromoxynil	40 + 336						
Tolpyralate + bentazon	40 + 1,080	3	bc		1	ab	
Tolpyralate + bentazon fb	40 + 1,080 fb	20	ef	Y	6	bc	Z
Tolpyralate + bentazon	40 + 1,080						
Tolpyralate + glufosinate ammonium	40 + 500	2	b	Y	0	a	Z
Tolpyralate + glufosinate ammonium fb tolpyralate + glufosinate ammonium	40 + 500 fb 40 + 500	13	de	Y	1	ab	Z
4 WAT							
Nontreated control		0	a		0	a	
Tolpyralate + atrazine	40 + 560	2	b		0	a	
Tolpyralate + atrazine fb tolpyralate + atrazine	40 + 560 fb 40 + 560	19	d	Y	2	bc	Z
Tolpyralate + bromoxynil	40 + 336	3	bc		1	ab	
Tolpyralate + bromoxynil fb tolpyralate + bromoxynil	40 + 336 fb 40 + 336	16	d	Y	7	c	Z
Tolpyralate + bentazon	40 + 1,080	1	b		0	a	
Tolpyralate + bentazon fb tolpyralate + bentazon	40 + 1,080 fb 40 + 1,080	10	cd	Y	2	bc	Z
Tolpyralate + glufosinate ammonium	40 + 500	0	a		0	a	
Tolpyralate + glufosinate ammonium fb tolpyralate + glufosinate ammonium	40 + 500 fb 40 + 500	5	bc	Y	0	a	Z
8 WAT							
Nontreated control		0	a		0	a	
Tolpyralate + atrazine	40 + 560	0	a		0	a	
Tolpyralate + atrazine fb tolpyralate + atrazine	40 + 560 fb 40 + 560	2	b	Y	0	a	Z
Tolpyralate + bromoxynil	40 + 336	0	a		0	a	
Tolpyralate + bromoxynil fb tolpyralate + bromoxynil	40 + 336 fb 40 + 336	2	b	Y	0	a	Z
Tolpyralate + bentazon	40 + 1,080	0	a		0	a	
Tolpyralate + bentazon fb tolpyralate + bentazon	40 + 1,080 fb 40 + 1,080	1	ab	Y	0	a	Z
Tolpyralate + glufosinate ammonium	40 + 500	0	a		0	a	
Tolpyralate + glufosinate ammonium fb tolpyralate + glufosinate ammonium	40 + 500 fb 40 + 500	0	a		0	a	

^aAbbreviations: fb, followed by; WAT, weeks after herbicide application.

^bMeans followed by the same letter within a column (a–d) or row (Y–Z) are not significantly different according to a Tukey-Kramer multiple range test at $P < 0.05$. Rows without an uppercase letter have no differences between hybrids.

^cAll treatments included methylated seed oil (1% or 2% mL L⁻¹ for the labeled herbicide rate and twice the labeled rate, respectively).

rates of tolpyralate (5, 20, 29, 40, 50 and 100 g ai ha⁻¹) applied alone or in a mixture with atrazine (560 g ai ha⁻¹). Metzger *et al.* (2019) observed up to 10% injury at 4 WAT with tolpyralate + atrazine applied at 1× and 2× rates (40 + 1,000 or 80 + 2,000 g ai ha⁻¹) to the ‘DKC46-82’ corn hybrid. Soltani *et al.* (2023) observed 12% injury at 1 WAT and 5% injury at 4 WAT to DKC39-97 and ‘RIB/DKC42-04’ corn hybrids with tolpyralate + atrazine applied postemergence. Similarly, Langdon *et al.* (2020b) observed slightly higher injury to ‘P9998AM’ and ‘P9840AM’ hybrids compared with ‘DKC42-60’ and ‘DKC43-47’ corn hybrids at 1 WAT when tolpyralate + atrazine was applied postemergence at the 2× rate, but the difference in injury was transient and not present at 2, 4, and 8 WAT and had no adverse effect on corn population or grain yield (Langdon *et al.* 2020b). Other studies have also shown no, or minor, visible corn injury ($\leq 2\%$) to DKC39-97, DKC42-04, and DKC42-60 corn hybrids at

1, 2, and 4 WAT with tolpyralate alone or in combination with atrazine (Flutters *et al.* 2022a; Kohrt and Sprague 2017; Willemse *et al.* 2021a).

Flutters *et al.* (2022a) observed 6% injury to corn hybrids DKC42-04, DKC42-60, and DKC39-97 at 1 WAT, and 3% injury at 2 WAT with mixtures of tolpyralate + bromoxynil, but there was minimal injury (1% or less) at 1 or 2 WAT with mixtures of tolpyralate + atrazine, bentazon, or glufosinate. Another study reported 16% visible corn injury with tolpyralate + bromoxynil and 11% injury with tolpyralate + bentazon 1 WAT, but the visible injury was $< 8\%$ at 2 WAT and 2% at 4 WAT (Willemse *et al.* 2021a).

Corn Height

At 2 WAT, the height of the DKC39-97 hybrid was reduced 17% with tolpyralate + atrazine (2× rate), 8% with tolpyralate + bromoxynil

Table 3. Relative corn height 2 wk after treatment following tolpyralate plus reactive oxygen species-generating herbicides applied postemergence for five trials conducted in Ridgetown, ON, between 2021 and 2023.^{a-c}

Herbicide treatment	Rate	Relative corn height			
		DKC39-97		B79N56PWE	
		%			
	g ai ha ⁻¹				
Non-treated control		100	a	100	a
Tolpyralate + atrazine	40 + 560	96	ab	100	a
Tolpyralate + atrazine fb tolpyralate + atrazine	40 + 560 fb 40 + 560	83	d	95	abc
Tolpyralate + bromoxynil	40 + 336	92	bc	95	abc
Tolpyralate + bromoxynil fb tolpyralate + bromoxynil	40 + 336 fb 40 + 336	84	d	88	c
Tolpyralate + bentazon	40 + 1080	96	ab	96	ab
Tolpyralate + bentazon fb tolpyralate + bentazon	40 + 1,080 fb 40 + 1,080	87	cd	90	bc
Tolpyralate + glufosinate ammonium	40 + 500	99	ab	100	a
Tolpyralate + glufosinate ammonium fb tolpyralate + glufosinate ammonium	40 + 500 fb 40 + 500	93	abc	99	a

^aAbbreviations: fb, followed by; WAT, weeks after herbicide application.

^bMeans followed by the same letter within a column (a–d) or row (Y–Z) are not significantly different according to a Tukey-Kramer multiple range test at $P < 0.05$. Rows without an uppercase letter have no differences between hybrids.

^cAll treatments included methylated seed oil (1% or 2% mL L⁻¹ for the labeled herbicide rate and twice the labeled rate, respectively).

(1× rate), 16% with tolpyralate + bromoxynil (2× rate), and 13% with tolpyralate + bentazon (2× rate) but it was not affected by other tolpyralate mixtures evaluated in this study (Table 3). B79N56PWE corn hybrid height was reduced 12% with tolpyralate + bromoxynil (2× rate) and 10% with tolpyralate + bentazon (2× rate) compared with the nontreated control, but it was not adversely affected by other tolpyralate mixtures evaluated (Table 3). Tolpyralate + atrazine and tolpyralate + glufosinate at the 2× rates caused a 12% and 6% greater reduction in height of DKC39-97 hybrid compared with B79N56PWE hybrid, respectively; all other herbicide treatments resulted in similar heights of both corn hybrids (Table 3).

Corn Population and Yield

There was no effect of corn hybrid or herbicide treatment on the corn population (Table 1). DKC39-97 grain yield was 2% lower than B79N56PWE (Table 1). Tolpyralate + bromoxynil or bentazon (2× rates) caused a 7% and 6% corn grain yield reduction, respectively, compared to tolpyralate + glufosinate (2× rate), respectively; all herbicide treatments resulted in comparable corn grain yields (Table 1). Results are similar to other studies that reported corn grain yield is not affected by the interaction between HPPD-inhibiting herbicides such as tolpyralate with ROS-generating herbicides such as atrazine, bromoxynil, bentazon, or glufosinate (Fluttert et al. 2022a; Willemse et al. 2021a). Other studies have also shown no adverse effect on the yield from various corn hybrids (DKC39-97, DKC42-04, DKC42-60, DKC43-47, DKC46-82, P9998AM, P9840AM) with tolpyralate alone or in combination with atrazine (Fluttert et al. 2022a,b; Kohrt and Sprague 2017; Langdon et al. 2020a,b, 2021; Metzger et al. 2018a,b, 2019; Soltani et al. 2023; Willemse et al. 2021a,b).

This research concludes that tolpyralate + atrazine, bromoxynil, bentazon, or glufosinate applied postemergence can cause corn significant corn injury to some corn hybrids. Injury was generally greater IN DKC39-97 than B79N56PWE corn hybrid. Generally, corn injury was greatest when tolpyralate was co-applied with bromoxynil and the least injury occurred when tolpyralate was applied in a mixture with glufosinate. Generally, corn injury was greater at the 2× rates with the herbicide mixtures evaluated in these studies. The visible corn injury was transient, with minimal injury observed at 8 WAT. The co-application of tolpyralate with bromoxynil in place of atrazine did not accentuate

corn injury to the DKC39-97 hybrid, but injury to the B79N56PWE hybrid was greater at 1 and 2 WAT; however, this increased injury was not present by 4 and 8 WAT. The co-application of tolpyralate with bentazon in place of atrazine resulted in similar corn injury to both hybrids at 1, 2, 4, and 8 WAT. The co-application of tolpyralate with glufosinate in place of atrazine resulted in similar or less corn injury at 1, 2, 4, and 8 WAT. At 2 WAT, tolpyralate + atrazine and tolpyralate + glufosinate applied at the 2× rates caused 12% and 6% greater reduction in the height of the DKC39-97 hybrid compared with the B79N56PWE hybrid, respectively; however, the corn height of both hybrids was similar with all other tolpyralate mixtures. Corn producers need to consider corn hybrid sensitivity and ROS-generating herbicide when using tolpyralate for weed management in corn.

Practical Implications

Tolpyralate is a relatively new pyrazole herbicide (Group 27) registered for weed management in corn. Tolpyralate plus ROS-generating herbicides (atrazine, bromoxynil, bentazon, or glufosinate) can provide broad-spectrum weed control, and it can control problematic weeds, especially MHR waterhemp and horseweed biotypes. Tolpyralate + ROS-generating herbicides can cause significant visible corn injury, especially when used at the 2× rates. Corn injury from tolpyralate + ROS-generating herbicides is influenced by corn hybrid; greater injury occurred to the DKC39-97 hybrid than to the B79N56PWE hybrid. The addition of bromoxynil in place of atrazine mixed with tolpyralate applied postemergence can accentuate corn injury. Visible injury was transient and decreased over time with minimal effect on corn grain yield. The differential sensitivity of corn hybrids and ROS-generating herbicides needs to be considered when using tolpyralate for weed management in corn.

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