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Response of Dicamba-Resistant Soybean to Glyphosate/Dicamba Application Rate and Timing

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Abstract

Three field experiments were conducted during 2016 and 2017 in southwestern Ontario, Canada to evaluate the effect of glyphosate/dicamba (2:1 ratio) applied at the V2/V3 or V4/V5 growth stage on dicamba-resistant (DR) soybean injury, growth response and yield. At 1 DAA, glyphosate/dicamba at 450, 900, 1350, 1800 and 3600 g·ae·ha^{-1} caused 0, 0.3, 1.8, 3.3 and 5.8% growth response (leaf droop) when applied at the V2/V3 growth stage and 0.3%, 1.3%, 1.5%, 2.3% and 4.5% growth response when applied at V4/V5 growth stage in DR soybean, respectively. The growth response was similar at 3 DAA. This response was transient with no growth response observed at 7 DAA, 2 WAB, 4 WAB, 8 WAB, and late Sept. (R8 growth stage). Glyphosate/dicamba at 450, 900, 1350, 1800 and 3600 g·ae·ha^{-1} caused 0.0, 1.4%, 3.6%, 5.7% and 10.7% injury (speckled chlorosis and necrosis and leaf distortion) when applied at the V2/V3 growth stage and 0.5%, 0.7%, 2.0%, 3.7% and 6.9% injury when applied at the V4/V5 growth stage in DR soybean, respectively. The injury observed was transient with no injury observed at 4 and 8 WAB and late Sept. (R8). There was no impact of glyphosate/dicamba at various rates evaluated on maturity and seed yield except at 1350 g·ae·ha^{-1} (V2/V3) and 3600 g·ae·ha^{-1} (V4/V5) which caused a 6% reduction in yield of DR soybean yield compared to the weed-free control. Based on these results, glyphosate/dicamba at the labelled rates can be safely applied at the V2/V3 and V4/V5 growth stage in DR soybean. However, care is needed to avoid spray overlaps as the 2× rate can result in significant crop injury and yield reduction in DR soybean.

Keywords

Chlorosis, Leaf Distortion, Leaf Droop, Necrosis, Seed Yield, *Glycine max*

1. Introduction

Soybean [*Glycine max* (L.) Merr.] is the most popular grain legume grown in Ontario and contributes significantly to the total value of agriculture sector in the province [1]. In 2018, approximately 4.2 million tonnes of soybean were produced from 1.22 million ha in Ontario with a farm-gate value of nearly $1.7 billion [1]. Controlling weeds, especially herbicide-resistant biotypes, is one of the most important concerns for soybean growers in Ontario. Currently, the problematic glyphosate-resistant (GR) weeds in the province include *Conyza canadensis* (L.) Cronq. (Canada fleabane), *Amaranthus rudis* (waterhemp), *Ambrosia trifida* L. (giant ragweed) and *Ambrosia artemisiifolia* L. (common ragweed). Concerns about herbicide-resistant weeds, especially GR biotypes, have prompted agricultural companies to develop dicamba-resistant (DR) soybean with genetic traits that confer resistance to dicamba and glyphosate [2].

Dicamba is a benzoic acid herbicide that controls nearly 100 broad-leaved annual weeds [3]. Additionally, dicamba has been reported to suppress more than 100 perennial broadleaved weed species including woody species [4]. Globally, there are only six dicamba-resistant weed species [5]. Research has shown that dicamba applied postemergence at the appropriate application timing, alone or in combination with other herbicides, can provide excellent control of troublesome broadleaved weeds including GR biotypes in soybean [2] [6] [7] [8] [9] [10]. Glyphosate/dicamba mixture used with DR soybean will allow glyphosate to be used in combination with another active ingredient with a different site-of-action to control troublesome weed species including GR biotypes and can be one component of a diversified integrated weed management program [11].

Glyphosate/dicamba use along with DR soybean has become popular in recent years among soybean producers in North America. Growers in the United States seeded nearly 10 million hectares of DR soybeans in 2017 [12]. Similarly, soybean growers in eastern Canada seeded 13 and 31% of their total production with DR soybean cultivars in 2017 and 2018, respectively.

Glyphosate/dicamba is currently registered at 900 (600 g·ae·ha^{-1} of glyphosate + 300 g·ae·ha^{-1} of dicamba premixed) to 1800 (1200 g·ae·ha^{-1} of glyphosate + 600 g·ae·ha^{-1} of dicamba premixed) g·ae·ha^{-1} for use in DR soybean in Ontario. DR soybean has been commercially available since 2016, but there has been little published information on the sensitivity of DR soybean cultivars to glyphosate/dicamba, especially at the 0.5× and 2× rates when applied at different application timings under Ontario environmental conditions. In non-DR crops, dicamba like other auxinic herbicides generally causes leaf cupping and crinkling, stem and petiole curling and twisting, and interference with phloem sucrose and nutrient transport which can result in chlorosis, wilting, stunting and total necrosis of the crop [6] [13] [14].

The purpose of this study was to determine the effect of glyphosate/dicamba (450, 900, 1350, 1800 and 3600 g·ae·ha^{-1}) applied postemergence at the V2/V3

(POST 1) or V4/V5 (POST 2) growth stages on DR soybean growth response, injury, maturity and yield.

2. Material and Methods

2.1. Study Establishment

There were three field experiments conducted during 2016 and 2017 at the University of Guelph Ridgetown Campus (GPS: 42.454011, −81.879970) near Ridgetown, ON, Canada. The soil was a loam with 47% sand, 37% silt, 16% clay, 5.1% organic matter and pH of 6.3 in 2016 at site A; loam with 40% sand, 34% silt, 26% clay, 3.9% organic matter and pH of 7.6 in 2016 at site B; and loam with 43% sand, 42% silt, 15% clay, 4.2% organic matter and pH of 6.5 in 2017. Seedbed preparation consisted of fall moldboard plowing followed by two passes with a field cultivator with rolling basket harrows in the spring.

The experiments were arranged in a randomized block design with four replications. Treatments included a weed-free control, and glyphosate/dicamba at 450, 900, 1350, 1800 and 3600 g·ae·ha^{-1} applied postemergence (POST) at the V2/V3 and V4/V5 soybean growth stages. Plots were 3 m wide (4 soybean rows spaced 75 cm apart) and 8 m long. DR soybean (DKB 14-41) was planted at the rate of 370,000 seed ha^{-1} in mid to late-May of each year. Herbicides were applied postemergence at the V1/V2 (POST 1) and V4/V5 (POST 2) growth stages when soybean was up to 17 cm and 35 tall, respectively. Herbicides were applied with a CO_2-pressurized backpack sprayer calibrated to deliver 200 L ha^{-1} aqueous solution at 207 kPa using 4 ULD 120-02 nozzles (TeeJet® Ultra low-drift 12002 nozzles, Spraying Systems Company, P.O. Box 7900 Wheaton, IL 60189-7900) spaced 50 cm apart. The experimental area was maintained weed-free during the entire growing season.

2.2. Data Collection

Percent growth response (leaf droop) was evaluated based on a scale of 0% to 100% (0 was no leaf droop and 100 was total leaf droop) at 1, 3 and 7 days after application (DAA); 2, 4 and 8 weeks after POST 2 application (WAB); and late Sept. (R8 growth stage). Percent DR soybean injury (speckled chlorosis and necrosis and leaf distortion) was also evaluated using the same scale (0 was no visible injury and 100 was total soybean necrosis) at 1, 3 and 7 DAA; 2, 4 and 8 WAB; and late Sept. (R8). When soybean reached maturity in October of each year, soybean seed yield was determined by harvesting the middle 2 rows of each plot with an ALMACO™ small plot combine (ALMACO™, 99 M Avenue, Nevada, Iowa 50201-1558 USA). Seed yield was adjusted to 13.0% seed moisture content.

2.3. Statistical Analysis

Data were analyzed using the GLIMMIX procedure in SAS (Ver. 9.4, SAS Institute Inc., Cary, NC). The treatments were arranged in a randomized complete block design replicated 4 times. The fixed effect was herbicide treatment and random effects were year-location combinations (environment), replicate within the environment and the environment by treatment interaction. The best distribution and associated link function for each parameter was chosen by comparing fit statistics, residual plots and the Shapiro-Wilk statistic among the potential distributions. Least square means (LSMEANS) were calculated on the data scale by using the inverse link function, and pairwise comparisons were subjected to Tukey's adjustment before determining treatment differences at $P < 0.05$. The weed-free control was assigned a value of 0 for percent growth response and percent injury due to zero variance. Comparisons were still possible between the other treatments and the value zero using the LSMEANS output and differences were identified. The Gaussian distribution and identity link were used for percent growth response, percent injury 1, 3 and 7 DAA; 4 and 8 WAB; and late Sept. (R8), moisture and yield. The normal distribution (identity link) with an arcsine square root transformation was used for percent injury 2 WAB and the data was back-transformed for the presentation of results.

3. Results and Discussion

Growth response at 7 DAA, 2 WAB, 4 WAB, 8 WAB, and late Sept. (R8) was zero across all locations and years and therefore the date is not presented. Similarly, percent soybean injury at 1 DAA, 4 WAB, 8 WAB, and late Sept. (R8) was zero across all locations and years and therefore is not presented.

3.1. Growth Response (Leaf Droop)

At 1 DAA, glyphosate/dicamba applied at 450, 900, 1350, 1800 and 3600 $g \cdot ae \cdot ha^{-1}$ to V2/V3 DR soybean caused 0.0%, 0.3%, 1.8%, 3.3% and 5.8% growth response, respectively (Table 1). At 3 DAA glyphosate/dicamba applied at 450,

Table 1. Percent growth response, injury, moisture at maturity and yield of dicamba-resistant soybean (Ridgetown 2016-2017) treated with glyphosate/dicamba at various rates at two POST timings. Means followed by a different letter within a column are significantly different according to a Tukey-Kramer multiple range test at $P < 0.05$[a].

Treatment	Rate ($g \cdot ai \cdot ha^{-1}$)	Application Timing	Growth Response (%)		Injury (%)			Moisture (%)	Yield ($T \cdot ha^{-1}$)
			1 DAA	3 DAA	3 DAA	7 DAA	14 DAB		
Weed-free control			0.0 a	0.0 a	0.0 a	0.0 a	0.0 a	14.9 a	4.64 a
Glyphosate/dicamba	450	POST 1	0.0 a	0.0 a	0.0 a	0.0 a	0.0 a	15.2 a	4.47 ab
Glyphosate/dicamba	900	POST 1	0.3 ab	0.3 ab	1.4 abc	0.0 a	0.0 a	14.9 a	4.55 ab
Glyphosate/dicamba	1350	POST 1	1.8 c	1.6 ab	3.6 bc	0.4 ab	0.0 a	14.8 a	4.36 b
Glyphosate/dicamba	1800	POST1	3.3 d	2.7 b	5.7 bcd	1.5 bc	0.0 a	15.1 a	4.52 ab
Glyphosate/dicamba	3600	POST1	5.8 f	3.0 b	10.7 d	4.6 d	0.2 b	14.8 a	4.47 ab
Glyphosate/dicamba	450	POST 2	0.3 ab	0.0 a	0.5 ab	0.0 a	0.0 a	15.2 a	4.42 ab
Glyphosate/dicamba	900	POST 2	1.3 bc	0.4 ab	0.7 ab	0.3 ab	0.0 a	15.1 a	4.46 ab
Glyphosate/dicamba	1350	POST 2	1.5 c	1.2 ab	2.0 abc	1.0 bc	0.0 a	14.9 a	4.44 ab
Glyphosate/dicamba	1800	POST 2	2.3 cd	2.2 b	3.7 bc	2.2 c	0.0 a	14.7 a	4.50 ab
Glyphosate/dicamba	3600	POST 2	4.5 e	3.4 b	6.9 cd	5.2 d	1.2 b	15.0 a	4.36 b

[a]Abbreviations: DAA, days after application; DAB, days after B application; POST, postemergence.

900, 1350, 1800 and 3600 g·ae·ha^{-1} to V2/V3 DR soybean caused 0%, 0.3%, 1.6%, 2.7% and 3.0% growth response, respectively (Table 1). These responses were transient with no growth response observed at 7 DAA, 2 WAB, 4 WAB, 8 WAB, and late Sept. (R8) in DR soybean (data are not shown).

There was generally no difference in growth response between the two application timings (V2/V3 vs V4/V5). At 1 DAA, glyphosate/dicamba applied at 450, 900, 1350, 1800 and 3600 g·ae·ha^{-1} to DR soybean at V4/V5 growth stage caused 0.3, 1.3, 1.5, 2.3 and 4.5% growth response, respectively (Table 1). At 3 DAA, glyphosate/dicamba applied at 450, 900, 1350, 1800 and 3600 g·ae·ha^{-1} to DR soybean at V4/V5 growth stage caused 0.0, 0.4, 1.2, 2.2 and 3.4% growth response, respectively. The growth response was transient with no symptoms observed at 7 DAA, 2 WAB, 4 WAB, 8 WAB, and late Sept. (R8) in DR soybean (data are not shown).

3.2. Soybean Injury (Speckled Chlorosis and Necrosis and Leaf Distortion)

Soybean injury followed a similar pattern as growth response. Glyphosate/dicamba applied at 450, 900, 1350, 1800 and 3600 g·ae·ha^{-1} to DR soybean at the V2/V3 growth stage caused 0.0%, 1.4%, 3.6%, 5.7% and 10.7% injury at 3 DAA; 0.0%, 0.0%, 0.4%, 1.5% and 4.6% injury at 7 DAA; and 0.0%, 0.0%, 0.0%, 0.0% and 0.2% injury at 14 DAB, respectively. The injury was transient with no injury observed at 4 WAB, 8 WAB, and late Sept. (R8) in DR soybean (data are not shown).

There was generally no difference in soybean injury between the two application timings (V2/V3 vs V4/V5). Glyphosate/dicamba applied at 450, 900, 1350, 1800 and 3600 g·ae·ha^{-1} to DR soybean at V4/V5 growth stage caused 0.5%, 0.7%, 2.0%, 3.7% and 6.9% injury at 3 DAA; 0.0%, 0.3%, 1.0%, 2.2% and 5.2% injury at 7 DAA; and 0.0%, 0.0%, 0.0%, 0.0% and 1.2% injury at 14 DAB, respectively (Table 1). The injury was transient with no injury observed at 4 WAB, 8 WAB, and late Sept. (R8) in DR soybean (data are not shown).

In contrast, other studies have found 20% to 89% injury when non-DR soybean was exposed to dicamba applied at 4.4 to 280 g·ae·ha^{-1} at the V2/V3 growth stage [15]. Another study reported 8% to 21% injury when non-DR soybean was exposed to dicamba applied at only 3 g·ae·ha^{-1} at the V2/V3 growth stage [7]. Anderson *et al.* [16] reported 40 and 80% injury in non-DR soybean with dicamba applied at 5.6 and 56 g·ae·ha^{-1} at the V2/V3 trifoliate growth stage.

3.3. Soybean Maturity and Yield

Glyphosate/dicamba applied at 450, 900, 1350, 1800 and 3600 g·ae·ha^{-1} to DR soybean at V2/V3 or V4/V5 growth stage had no impact on crop maturity as indicated by seed moisture content at harvest (Table 1). There was no difference in soybean yield among the rates of glyphosate/dicamba (450, 900, 1350, 1800 and 3600 g·ae·ha^{-1}) applied at the V2/V3 and V4/V5 growth stages. Interestingly, DR soybean yield was reduced 6% compared to the weed-free control with glyphosate/dicamba applied at 1350 g·ae·ha^{-1} (V2/V3) and 3600 g·ae·ha^{-1} (V4/V5) (**Table 1**). This observation will have to be confirmed in future studies. There was generally no difference in soybean seed yield between the two application timings (V2/V3 vs V4/V5). In other studies, seed yield loss of 4% and 10% have been reported with dicamba applied at 4.4 and 17.5 g·ae·ha^{-1} at the V2/V3 growth stage in non-DR soybean [15]. Another study reported 8% seed yield reduction when non-DR soybean was exposed to dicamba applied at 11 g·ae·ha^{-1} at the V2/V3 growth stage [17]. Seed yield losses were as much as 34 and 83% when non-DR soybean was exposed to 5.6 and 56 g·ae·ha^{-1} of dicamba applied at the V2/V3 growth stage, respectively [7]. Other studies have also shown soybean seed yield losses of 18% with 1% dicamba tankmix contamination in non-DR soybean [15].

4. Conclusion

Results from this study showed that glyphosate/dicamba at 450, 900, 1350, 1800 and 3600 g·ae·ha^{-1} can cause up to a 6% growth response and 11% injury in DR soybean. This response was transient with no growth response or injury symptoms observed beyond 14 DAB. There was no impact of glyphosate/dicamba at various rates evaluated on maturity, but seed yield was reduced as much as 6% compared to the weed-free control. There were generally no differences in responses observed between the two application timings (V2/V3 vs V4/V5) evaluated. Based on these results, glyphosate/dicamba at the labelled rates can be safely applied at the V2/V3 and V4/V5 growth stage in DR soybean. However, care is needed to avoid spray overlaps as the 2× rate can result in significant DR soybean injury and yield loss.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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