Improving the Level and Consistency of Glyphosate-Resistant Canada Fleabane (*Erigeron canadensis* L.) Control With Bromoxynil-Based Tankmixes Applied Preplant to Soybean

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Abstract

Glyphosate-resistant (GR) Canada fleabane (*Erigeron canadensis* L.) is a problematic weed in soybean. Bromoxynil-based tankmixes provide control of GR Canada fleabane in monocot crops; however, there is limited research on preplant (PP) applications in soybean. The objective of this study was to determine the best third tankmix partner with glyphosate plus bromoxynil applied PP to improve the level and consistency of GR Canada fleabane control in soybean. Four field trials were conducted over a two-year (2020, 2021) period. Glyphosate plus bromoxynil controlled GR Canada fleabane 52, 58, and 69% at 2, 4, and 8 weeks after application (WAA) respectively. The addition of saflufenacil to glyphosate plus bromoxynil improved GR Canada fleabane control 39% at 2 WAA and the addition of dicamba to glyphosate plus bromoxynil improved GR Canada fleabane control 39 and 29% at 4 and 8 WAA, respectively. The addition of bromoxynil to glyphosate plus 2,4-D ester or dicamba improved GR Canada fleabane control by 36 and 35%, respectively, at 2 WAA. The addition of bromoxynil to glyphosate plus tiafenacil or pyraflufen-ethyl/2,4-D improved GR Canada fleabane control by 19% at 8 WAA. The addition of bromoxynil to glyphosate plus metribuzin improved GR Canada fleabane control 31, 32, and 36% at 2, 4, and 8 WAA, respectively. Antagonism was observed between bromoxynil plus glyphosate plus tiafenacil or halauxifen-methyl 8 WAA, while the interaction between bromoxynil plus glyphosate plus pyraflufen-ethyl/2,4-D was considered synergistic. The addition of metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba to glyphosate plus bromoxynil improved the consistency of GR Canada fleabane at 2, 4, and 8 WAA. This study concludes that the addition of saflufenacil or dicamba to glyphosate plus bromoxynil can improve the level and consistency of GR Canada fleabane control applied PP to soybean.

Keywords: additive, antagonistic, biomass, density, horseweed, synergy, weed control, grain yield

1. Introduction

Glyphosate-resistant (GR) Canada fleabane was the first documented dicot weed to evolve resistance to glyphosate (Heap, 2020; VanGessel, 2001). GR Canada fleabane can produce thousands of seeds (Bhowmik & Bekech, 1993; Davis et al., 2009) which can move up to 500 km in one dispersal event (Shields et al., 2006). In Ontario, GR Canada fleabane primarily emerges in the spring and in the fall; though, if temperature and soil moisture conditions are favourable, it has been reported to emerge every month of the year except January (Cici & Van Acker, 2009; Main et al., 2006). Seeds within the top 0.3 cm of the soil surface can be stimulated by light and have the greatest likelihood of germinating and emerging (Nandula et al., 2006); though, its ruderal nature allows it to germinate in a variety of landscapes (Weaver, 2001). GR Canada fleabane has been confirmed in 14 countries as of 2020 (Heap, 2020).

GR Canada fleabane interference can reduce soybean yield. GR Canada fleabane interference can result in up to 93% yield loss if no weed management strategies are used (Byker et al., 2013a). In most soybean systems, GR Canada fleabane should be managed before soybean emergence to prevent yield loss (Bruce & Kells, 1990). In conventional-tillage production systems, tillage can be used to manage GR Canada fleabane, but in no-tillage systems, herbicides must be used to manage GR Canada fleabane (Bruce & Kells, 1990). Postemergence (POST)

herbicides provide minimal control of GR Canada fleabane in GR and non-GMO soybean (Byker et al., 2013b); therefore, preplant (PP) and preemergence (PRE) herbicides must be used for acceptable control.

Bromoxynil is a photosystem II (PSII)-inhibiting herbicide (Fedtke & Duke, 2005; Shaner, 2014). Bromoxynil is a contact herbicide (Bayer CropScience Inc., 2019) that can outcompete plastiquinone at the Q_B-binding site on the D1 protein, which inhibits electron transport, uncouples oxidative phosphorylation, and results in the reactive oxygen species formation and lipid peroxidation (Shaner, 2014; United States Environmental Protection Agency, 1998). Bromoxynil applied POST is not registered for use in soybean since it can cause leaf necrosis and stunting, though the plant may recover with little effect on yield (Andersen et al., 1973; Wax et al., 1974). Bromoxynil is registered to control select annual broadleaf weeds in corn, small grains, alfalfa, carrot, garlic, and onion (Boydston & Al-Khatib, 1993; Bayer CropScience Inc., 2019). Bromoxynil is a contact herbicide that is generally used to control annual broadleaf weeds in monocot crops.

GR Canada fleabane is currently not listed as one of the weeds controlled on the brmoxynil label, but prior research with bromoxynil-based tankmixes in corn and winter wheat has demonstrated GR Canada fleabane control. Mahoney et al. (2017) and Metzger et al. (2019) observed 93 and 94% control, respectively of GR and multiple-herbicide-resistant (MHR) Canada fleabane with bromoxynil (280 g ai ha^{-1}) plus atrazine (1500 g ai ha^{-1}) applied POST in corn 8 WAA. Quinn et al. (2021) reported 98% GR Canada fleabane control with a premix of pyrasulfotole/bromoxynil (205 g ai ha^{-1}) plus AMS (1 L ha^{-1}) applied POST in winter wheat 8 WAA. Mahoney et al. (2016) observed bromoxynil (174 g ai ha^{-1}) plus pyrasulfotole (31 g ai ha^{-1}) applied POST controlled 97% of GR Canada fleabane in winter wheat at 8 WAA. Bromoxynil tankmixes control GR Canada fleabane in monocot crops, which suggests potential for preplant (PP) applications in soybean.

Based on the aforementioned research, Westerveld et al. (2021a) evaluated the GR Canada fleabane control with bromoxynil, metribuzin, and bromoxynil plus metribuzin applied PP in soybean. Bromoxynil (35, 70, 140, 280, 560, 1120 g ai ha^{-1}) provided 31 to 96% control, metribuzin (400 g ai ha^{-1}) provided 80% control and the combination of bromoxynil plus metribuzin increased GR Canada fleabane control to 89 to 100% at 8 WAA; similar to current industry standards of glyphosate plus saflufenacil (25 g ai ha^{-1}) plus metribuzin (400 g ai ha^{-1}) (Budd et al., 2016) and glyphosate/dicamba (1800 g ae ha^{-1}) plus saflufenacil (25 g ai ha^{-1}) (Hedges et al., 2018). Therefore, more research is required to investigate possible tankmix partners with bromoxynil for GR Canada fleabane control applied PP in soybean. It is hypothesized the addition of metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba to glyphosate plus bromoxynil applied PP will improve the level and consistency of GR Canada fleabane control in soybean.

2. Materials and Methods

2.1 Experimental Methods

Four field experiments were conducted over a two-year period (2020, 2021) at sites with confirmed GR Canada fleabane in southwestern Ontario. A 2 × 8 factorial was organized in a randomized complete block design (RCBD) with 4 replications. Factor One was a control and bromoxynil, and Factor Two was a control, metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil, and dicamba. The plots were 2.25 m [3 glyphosate/dicamba-resistant (GDR) soybean rows at 0.75 m apart] by 8 m. The replicates were divided by a 2.0 m alley.

Herbicide treatments were made PP when majority of the Canada fleabane was 10 cm in height. Glyphosate (900 g ae ha^{-1}) was added to each herbicide treatment applied PP. A CO_2-pressurized backpack sprayer was calibrated to dispense 200 L ha^{-1} at 240 kPa. The sprayer had four, ULD 120-02 nozzles separated 50 cm apart. The spray width measured 2.0 m. Following PP herbicide applications, a GDR soybean cultivar (DKB12-16) was seeded to about 400,000 seeds ha^{-1} at a 3.75 cm depth. A glyphosate (450 g ae ha^{-1}) cover spray was applied POST to eliminate the effects of other weed species and glyphosate-susceptible Canada fleabane. Site, year, location, soil, and agronomic details are listed in Table 1. Information pertaining to Canada fleabane including height and density at application and the resistance profile of the populations is presented in Table 2. The herbicide treatments that were used in this study are described in Table 3.

Table 1. Site, year, location, soil characteristics, soybean seeding and soybean emergence dates for four trials conducted on commercial farms in southwestern Ontario, Canada in 2020 and 2021

Site	Year	Location	Soil traits						Agronomic information		
			Texture	Sand	Silt	Clay	OM[a]	pH	Treatment spray date	Soybean seeding date	Soybean emergence date
				%	%	%	%				
S1	2020	Ridgetown	Sandy loam	75	17	7	1.9	7.1	26 May	5 June	11 June
S2	2020	Zone Centre	Loamy sand	85	9	5	2.9	6.5	17 June	24 June	3 July
S3	2021	Kintyre	Sandy loam	53	29	18	4.4	6.9	19 May	19 May	25 May
S4	2021	Bothwell	Loamy sand	85	11	4	3.3	6.8	24 May	12 June	18 June

Note. [a] Abbreviations: OM, organic matter.

Table 2. Canada fleabane height and density at time of application and resistance profile for trials conducted on commercial farms in southwestern Ontario, Canada in 2020 and 2021

Site	Year	Location	Canada fleabane		Resistance	
			Height	Density	Glyphosate	Cloransulam-methyl
			cm	m^{-2}	%	%
S1	2020	Ridgetown	7	492	100	99
S2	2020	Zone Centre	11	70	79	100
S3	2021	Kintyre	9	34	98	85
S4	2021	Bothwell	8	82	-	-

Table 3. Herbicides used in four field trials in southwestern Ontario, Canada in 2020 and 2021

Active ingredient	Trade name	Manufacturer
Bromoxynil	Pardner®	Bayer CropScience Inc., 160 Quarry Park Blvd S. E., Calgary, AB.
Metribuzin	Sencor® 480SC	Bayer CropScience Inc., 160 Quarry Park Blvd S. E., Calgary, AB.
Tiafenacil[a]	Terrad'or™	ISK Biosciences., 7470 Auburn Rd, Painesville, OH 44077, United States.
Pyraflufen-ethyl/2,4-D	Blackhawk	Nufarm Canada., 5101, 333-96th Ave N.E., Calgary, AB.
2,4-D ester	Ester 700	Nufarm Canada., 5101, 333-96th Ave N.E., Calgary, AB.
Halauxifen-methyl[a]	Elevore	Dow AgroSciences Canada Inc., 2400, 215-2ns Street S. W., Calgary, AB.
Saflufenacil[b]	Eragon® LQ	BASF Canada Inc., 100 Milverton Drive, Mississauga, ON.
Dicamba	Xtendimax™	Bayer CropScience Inc., 160 Quarry Park Blvd S. E., Calgary, AB.

Note. [a] MSO was add to all treatments with halauxifen-methyl (1.0 % v/v) or tiafenacil (0.5 % v/v). [b] Merge (1 L ha^{-1}) was added to treatments with saflufenacil.

Canada fleabane control was assessed as a visual estimate of the reduction in Canada fleabane biomass using a 0 to 100 scale at 2, 4, and 8 weeks after application (WAA). A visual rating of 0 suggested no GR Canada fleabane control and a visual rating of 100 suggested plant death (Canadian Weed Science Society, 2018). Canada fleabane density and biomass were collected 8 WAA. Density was collected by counting Canada fleabane plants in two, 0.25 m^2 quadrats placed randomly in the front and back of each plot. Biomass was collected by cutting the Canada fleabane where the stem meets the soil surface within each quadrat, placing the samples in separate bags for each individual plot, drying the samples to a constant moisture, and then weighing the samples.

A 0 to 100 scale was used to assess visible GDR soybean injury 2 and 4 weeks after soybean emergence (WAE); 0 suggested no soybean injury and 100 suggested soybean death (Canadian Weed Science Society, 2018). At soybean harvest maturity, two GDR soybean rows were harvested per plot using a small plot harvester. Soybean yield and moisture were recorded. Prior to analysis, the grain yield was corrected to a 13.5% moisture content.

2.2 Statistical Analysis

SAS 9.4 was the statistical software used in this study. The GLIMMIX procedure was used for the analysis. Herbicide treatment represented the fixed effect and block, block within location, and treatment by location represented the random effects. The Shapiro-Wilk test and the residual plots were utilized when assessing normality. Since no treatment by location interaction was found, the sites were pooled together for the analysis. An arcsine square root back transformation was used to meet normality assumptions for GR Canada fleabane

control at 2, 4, and 8 WAA. Soybean yield data was assessed using the default link identity. A lognormal distribution using the link identity was used to assess GR Canada fleabane density and biomass; the omega procedure was used to back-transform the means from the analysis format (M. Edwards, Ontario Agricultural College Statistician, University of Guelph, personal communication). Mean separations were completed using the Tukey-Kramer's multiple range test ($p < 0.10$). Significance was indicated by different letter codes within Table 4. To calculate the expected GR Canada fleabane control the Colby's equation was used. A: Bromoxynil and B: Tankmix partner.

$$\text{Expected} = (A + B) - \frac{A \times B}{100} \tag{1}$$

The expected GR Canada fleabane biomass and density was calculated using a modified Colby's equation. The 'W' in the equation represents the non-treated control mean.

$$\text{Expected} = \frac{A \times B}{W} \tag{2}$$

A t-test was used to analyze the expected versus the observed values, significance was indicated when $p < 0.10$. The interactions were considered additive when the observed control value was the same as the expected control value. The interaction was considered synergistic or antagonistic when the observed control values were greater than or less than the expected control values, respectively (Colby, 1967). Conversely, if the observed density or biomass values were greater than or less than the expected density or biomass values then the interaction was considered antagonistic or synergistic, respectively (Colby, 1967). The coefficient of variation was calculated to ascertain the consistency of GR Canada fleabane control for each least square mean estimate.

3. Results and Discussion

3.1 Soybean Injury

Soybean injury was ≤ 10% at all sites across both years (data not presented).

3.2 Glyphosate-Resistant Canada Fleabane Control

The simple effects are presented in Table 4 since the interaction between Factor One (control and bromoxynil) and Factor Two (control, metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil, and dicamba) for GR Canada fleabane control 2, 4, and 8 WAA was statistically significant ($p < 0.10$).

Table 4. Glyphosate-resistant Canada fleabane control 2, 4, and 8 weeks after application (WAA), density, and biomass with bromoxynil-based tankmixes from four field trials conducted in southwestern Ontario, Canada in 2020 and 2021

Bromoxynil	Tankmix partners		
	None	Bromoxynil	SE
Weed control 2 WAA	------------------------------- % -------------------------------		
None	0 c Y	52 b Z	7
Metribuzin	43 b Y	62 ab Z (67)	6
Tiafenacil[c]	60 ab Z	60 ab Z (70)*	7
Pyraflufen-ethyl/2,4-D	58 ab Z	69 ab Z (74)**	5
2,4-D ester	47 b Y	74 ab Z (73)	4
Halauxifen-methyl[c]	58 ab Z	71 ab Z (78)*	4
Saflufenacil[b]	84 a Z	85 a Z (81)	5
Dicamba	52 b Y	80 ab Z (76)	4
SE	3	3	
Weed control 4 WAA			
None	0 c Y	58 b Z	7
Metribuzin	45 b Y	66 ab Z (72)	6
Tiafenacil[c]	50 ab Z	62 ab Z (71)	7
Pyraflufen-ethyl/2,4-D	66 ab Z	78 ab Z (82)**	4
2,4-D ester	77 ab Z	82 ab Z (87)*	3
Halauxifen-methyl[c]	87 a Z	86 ab Z (93)*	2
Saflufenacil[b]	87 a Z	92 ab Z (87)	3
Dicamba	87 a Z	95 a Z (94)	2
SE	3	2	
Weed control 8 WAA			
None	0 d Y	69 b Z	7
Metribuzin	46 c Y	72 ab Z (78)	6
Tiafenacil[c]	51 bc Y	70 b Z (76)*	7
Pyraflufen-ethyl/2,4-D	68 abc Y	87 ab Z (83)**	4
2,4-D ester	84 ab Z	85 ab Z (90)	7
Halauxifen-methyl[c]	89 a Z	86 ab Z (93)*	2
Saflufenacil[b]	87 a Z	95 ab Z (89)	3
Dicamba	94 a Z	97 a Z (97)	1
SE	3	2	
Density	----------------------------- plants m^{-2} -----------------------------		
None	164 c Y	30 ab Z	23
Metribuzin	36 ab Y	24 ab Z (46)*	8
Tiafenacil[c]	75 bc Y	39 a Z (50)	15
Pyraflufen-ethyl/2,4-D	53 b Y	21 ab Z (51)**	8
2,4-D ester	29 ab Z	24 ab Z (42)*	6
Halauxifen-methyl[c]	20 ab Z	28 ab Z (43)*	6
Saflufenacil[b]	17 a Z	7 a Z (38)**	5
Dicamba	25 ab Z	9 ab Z (12)**	4
SE	7	4	
Biomass	----------------------------- g m^{-2} -----------------------------		
None	154 b Y	88 b Z	20
Metribuzin	151 b Y	123 b Z (62)	28
Tiafenacil[c]	91 b Z	111 b Y (74)	15
Pyraflufen-ethyl/2,4-D	77 b Z	89 b Y (73)	11
2,4-D ester	24 ab Z	48 b Z (75)	9
Halauxifen-methyl[c]	13 a Z	18 ab Z (82)**	3
Saflufenacil[b]	27 ab Z	19 ab Z (61)**	8
Dicamba	4 a Z	3 a Z (85)**	1
SE	9	8	

Note. Abbreviations: SE, standard error. Means accompanied by different letters in a column (a-c) or row (Z, Y) differ significantly based on Tukey Kramer's LSD (P < 0.10). ** Significant at P < 0.01; * significant at P < 0.10 based on a t-test conducted for observed versus expected values. [a] Values in parentheses represent the expected values from Colby's analysis. [b] Merge, 1 L ha^{-1} was added to all treatments with saflufenacil. [c] MSO, 1 % v/v or 0.5 % v/v was added to all treatments with halauxifen-methyl and tiafenacil, respectively.

At 2 WAA, glyphosate plus bromoxynil controlled GR Canada fleabane 52% (Table 4). When saflufenacil was added to glyphosate plus bromoxynil GR Canada fleabane control improved from 52 to 85%; control was not improved when metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl or dicamba were added to glyphosate plus bromoxynil. Limited research exists on GR Canada fleabane control using glyphosate plus bromoxynil plus saflufenacil though; previous literature has reported improved GR Canada fleabane control when saflufenacil was added to other herbicide tankmix partners. Hedges et al. (2018) reported when saflufenacil (25 g ai ha^{-1}) was added to glyphosate/dicamba (1800 g ae ha^{-1}) GR Canada fleabane control improved from 54 to 97% at 2 WAA. Glyphosate plus metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba controlled GR Canada fleabane 43, 60, 58, 47, 58, 84, and 52%, respectively. When bromoxynil was added to glyphosate plus metribuzin, 2,4-D ester or dicamba, GR Canada fleabane control was improved from 43 to 62%, 47 to 74%, and 52 to 80%, respectively; control was not improved when bromoxynil was added to glyphosate plus tiafenacil, pyraflufen-ethyl/2,4-D, halauxifen-methyl or saflufenacil. Similarly, Westerveld et al. (2021a) reported the addition of bromoxynil (280 g ai ha^{-1}) to glyphosate (900 g ae ha^{-1}) plus metribuzin (400 g ai ha^{-1}) improved GR Canada fleabane control from 78 to 97% at 2 WAA. Based on the Colby's equation, glyphosate plus bromoxynil plus tiafenacil, pyraflufen-ethyl/2,4-D or halauxifen-methyl was antagonistic since the observed control was less than the expected control; all other tankmixtures were additive.

At 4 WAA, glyphosate plus bromoxynil provided 58% GR Canada fleabane control (Table 4). When dicamba was added to glyphosate plus bromoxynil GR Canada fleabane control improved from 58 to 95%; control was not improved from the addition of metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl or saflufenacil to glyphosate plus bromoxynil. To our knowledge, there is currently no existing research on GR Canada fleabane control with glyphosate plus bromoxynil plus dicamba though; previous research has reported improved GR Canada fleabane control when dicamba was added to other herbicide tankmix partners. Zimmer et al. (2018b) reported the addition of dicamba (280 g ae ha^{-1}) to glyphosate (560 g ae ha^{-1}) plus 2,4-D (560 g ae ha^{-1}) applied PP in soybean, improved GR Canada fleabane control from 71 to 95% at 5 WAA. Glyphosate plus metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba controlled GR Canada fleabane 45, 50, 66, 77, 87, 87, and 87%, respectively. When bromoxynil was added to glyphosate plus metribuzin GR Canada fleabane control improved from 45 to 66%; control was not improved when bromoxynil was added to glyphosate plus tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba. Similarly, Westerveld et al. (2021a) reported the addition of bromoxynil (280 g ai ha^{-1}) to glyphosate (900 g ae ha^{-1}) plus metribuzin (400 g ai ha^{-1}) improved GR Canada fleabane control from 80 to 98% at 4 WAA; which is much higher than the 66% GR Canada fleabane control reported in the present study at 4 WAA. Based on the Colby's equation, the observed control values for bromoxynil plus glyphosate plus pyraflufen-ethyl/2,4-D, 2,4-D ester or halauxifen-methyl were less than the expected control values indicating an antagonistic interaction; all other tankmixtures were additive.

At 8 WAA, glyphosate plus bromoxynil controlled GR Canada fleabane 69% (Table 4). When dicamba was added to glyphosate plus bromoxynil GR Canada fleabane control improved from 69 to 97%; control was not improved from the addition of metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl or saflufenacil to glyphosate plus bromoxynil. Previous studies reported improved GR Canada fleabane control when dicamba was added to other herbicide tankmixtures. Budd et al. (2016) reported when dicamba (600 g ae ha^{-1}) was added to glyphosate (900 g ae ha^{-1}) plus saflufenacil (25 g ai ha^{-1}) applied PP in soybean GR Canada fleabane control improved from 88 to 98% at 8 WAA. Glyphosate plus metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba controlled GR Canada fleabane 46, 51, 68, 84, 89, 87, and 94%, respectively. When bromoxynil was added to glyphosate plus metribuzin, tiafenacil or pyraflufen-ethyl/2,4-D GR Canada fleabane control improved from 46 to 72%, 51 to 70%, and 68 to 87%, respectively; control was not improved when bromoxynil was added to glyphosate plus 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba. Westerveld et al. (2021a) reported similar results when bromoxynil (280 g ai ha^{-1}) was added to glyphosate (900 g ae ha^{-1}) plus metribuzin (400 g ai ha^{-1}); GR Canada fleabane control improved from 76 to 97% at 8 WAA, which is much higher than the 72% GR Canada fleabane control reported in the present study at 8 WAA. Based on the Colby's equation, the observed control for glyphosate plus bromoxynil plus tiafenacil or halauxifen-methyl were less than the expected control demonstrating an antagonistic interaction; in contrast, the observed control for glyphosate plus bromoxynil plus pyraflufen-ethyl/2,4-D was greater than the expected indicating a synergistic interaction. All other tankmixtures were considered additive. Westerveld et al. (2021b) reported synergism when pyraflufen-ethyl/2,4-D (527 g ai ha^{-1}) was added to glyphosate (900 g ae ha^{-1}) plus metribuzin (400 g ai ha^{-1}) applied PP in soybean 8 WAA.

3.3 Glyphosate-Resistant Canada Fleabane Consistency in Control

The coefficient of variation (CV) is a standardized measure of the standard deviation to the mean and is useful for investigating data variability (Shechtman, 2013). The CV for each least square mean was used as an indicator to demonstrate the consistency of GR Canada fleabane control (Table 5). When comparing CVs, a lower CV would indicate greater consistency in control (Shechtman, 2013).

Table 5. Glyphosate-resistant Canada fleabane consistency of control 2, 4, and 8 weeks after application (WAA) with bromoxynil-based tankmixes from four field trials conducted in southwestern Ontario, Canada in 2020 and 2021

Bromoxynil	Tankmix partners	
	None	Bromoxynil
Consistency of control 2 WAA	-------------- Coefficient of variation -------------	
None	-	99
Metribuzin	111	88
Tiafenacil[c]	90	90
Pyraflufen-ethyl/2,4-D	92	82
2,4-D ester	105	77
Halauxifen-methyl[b]	92	79
Saflufenacil[a]	69	68
Dicamba	99	82
Consistency of control 4 WAA		
None	-	78
Metribuzin	92	71
Tiafenacil[c]	86	74
Pyraflufen-ethyl/2,4-D	71	62
2,4-D ester	63	59
Halauxifen-methyl[b]	56	56
Saflufenacil[a]	56	52
Dicamba	56	50
Consistency of control 8 WAA		
None	-	68
Metribuzin	89	66
Tiafenacil[c]	84	67
Pyraflufen-ethyl/2,4-D	69	56
2,4-D ester	57	57
Halauxifen-methyl[b]	54	56
Saflufenacil[a]	55	50
Dicamba	51	48

Note. [a] Merge, 1 L ha^{-1} was included in all treatments with saflufenacil. [b] MSO, 1 % v/v was included in all treatments with halauxifen-methyl and tiafenacil.

At 2, 4, and 8 WAA, the addition of metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba to glyphosate plus bromoxynil improved the consistency of GR Canada fleabane control, as indicated by lower CVs compared to the CV of glyphosate plus bromoxynil. When bromoxynil was added to glyphosate plus metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba the consistency of GR Canada fleabane control was improved as indicated by lower CVs except for glyphosate plus bromoxynil plus halauxifen-methyl at 4 and 8 WAA, which did not improve the consistency of control. It has been suggested that a third herbicide into the tankmix improves the consistency of GR Canada fleabane control (Mellendorf et al., 2013). Soltani et al. (2020b) reported improved consistency of GR Canada fleabane control when a third herbicide, such as metribuzin (400 g ai ha^{-1}), was added to glyphosate (900 g ae ha^{-1}) plus 2,4-D ester (500 g ae ha^{-1}) or saflufenacil (25 g ai ha^{-1}) 8 WAA. Budd et al. (2016) observed an improvement in the consistency of GR Canada fleabane control when metribuzin (400 g ai ha^{-1}) was added to glyphosate (900 g ae ha^{-1}) plus saflufenacil (25 g ai ha^{-1}).

3.4 Glyphosate-Resistant Canada Fleabane Density and Biomass

There was a significant interaction between Factor One (control and bromoxynil) and Factor Two (control, metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil, and dicamba) for GR Canada fleabane density and biomass ($P = 0.10$), so the simple effects are presented (Table 4).

There were relatively high populations of GR Canada fleabane at the study sites, with an average of 164 GR Canada fleabane plants per m^{-2} across the four sites at 8 WAA. At 8 WAA, glyphosate plus bromoxynil reduced GR Canada fleabane density 82%. GR Canada fleabane density was not reduced when metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba were added to glyphosate plus bromoxynil. Glyphosate plus metribuzin, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba reduced GR Canada fleabane density 78, 68, 82, 88, 90, and 85%, respectively; density was not reduced with glyphosate plus tiafenacil. Similar reductions in GR Canada fleabane density have been reported in previous research (Budd et al., 2016; Eubank et al., 2008; Soltani et al., 2020a; Westerveld et al., 2021c; Zimmer et al., 2018b). When bromoxynil was added to glyphosate plus metribuzin, tiafenacil or pyraflufen-ethyl/2,4-D GR Canada fleabane density was reduced 7, 22, and 20%, respectively; density was not reduced when bromoxynil was added to glyphosate plus 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba. In contrast, Westerveld et al. (2021a) reported no reduction in GR Canada fleabane density when bromoxynil (280 g ai ha^{-1}) was added to glyphosate (900 g ae ha^{-1}) plus metribuzin (400 g ai ha^{-1}) 8 WAA. Based on the Colby's equation, the observed density for glyphosate plus bromoxynil plus metribuzin, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba were less than the expected indicating synergistic interactions; the tankmix of bromoxynil plus glyphosate plus tiafenacil was additive.

At 8 WAA, glyphosate plus bromoxynil reduced GR Canada fleabane biomass 43%. When dicamba was added to glyphosate plus bromoxynil GR Canada fleabane biomass was reduced by 97%; there was no reduction in biomass when metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl or saflufenacil was added to glyphosate plus bromoxynil. There is minimal research on the efficacy of glyphosate plus bromoxynil plus dicamba though, the addition of dicamba (600 g ae ha^{-1}) to other herbicides such as glyphosate (900 g ae ha^{-1}) plus saflufenacil (25 g ai ha^{-1}) reduced GR Canada fleabane biomass up to 100% (Byker et al. 2013a) and 86% (Budd et al., 2016), respectively. Glyphosate plus halauxifen-methyl or dicamba reduced GR Canada fleabane biomass 92 and 97%, respectively; biomass was not reduced with glyphosate plus metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester or saflufenacil. Similar reductions in GR Canada fleabane biomass have been reported in past research (Budd et al., 2016; Byker et al., 2013a; Quinn et al., 2021). When bromoxynil was added to glyphosate plus metribuzin GR Canada fleabane biomass was reduced by 18%; interestingly, there was no benefit of adding bromoxynil to glyphosate plus tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba in respect to Canada fleabane biomass. Westerveld et al. (2021a) reported a 30% reduction in GR Canada fleabane biomass when bromoxynil (280 g ai ha^{-1}) was added to glyphosate (900 g ae ha^{-1}) plus metribuzin (400 g ai ha^{-1}) 8 WAA. Based on the Colby's equation, the observed biomass values for bromoxynil plus glyphosate plus halauxifen-methyl, saflufenacil or dicamba were less than the expected indicating a synergistic interaction; all other tankmixtures were considered additive.

3.5 Soybean Yield

There was no interaction between Factor One (control, bromoxynil) and Factor Two (control, metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil, and dicamba) for GR Canada fleabane interference on soybean yield, so the main effects are presented (Table 6). All herbicide treatments resulted in similar soybean yields. Zimmer et al. (2018a) reported similar soybean yields compared to this study. In contrast, Budd et al. (2016) reported a 69% soybean yield reduction from GR Canada fleabane interference.

Table 6. Main effects and interaction for glyphosate-resistant Canada fleabane control 2, 4, and 8 weeks after application (WAA), density, biomass, and soybean yield with bromoxynil-based tankmixes from four field trials conducted in southwestern Ontario, Canada in 2020 and 2021

Main effects	Rate (g ai/ae ha^{-1})	Canada fleabane control			Density[a]	Biomass[a]	Soybean yield
		2 WAA	4 WAA	8 WAA			
		---------------- % ----------------			Plants m[-2]	g m^{-2}	t ha^{-1}
Bromoxynil		NS	*	*	*	NS	NS
None	-	0	0	0	47	60	2.05
Bromoxynil	280	67	76	79	20	45	2.03
SE		2	2	2	4	6	0.051
Tankmix partners		**	**	**	**	**	NS
None	-	0	0	0	89	125	2.04
Metribuzin	400	53	55	58	20	151	1.84
Tiafenacil[c]	25	57	57	60	27	107	1.85
Pyraflufen-ethyl/2,4-D	532	61	69	73	31	85	2.09
2,4-D ester	528	58	76	81	12	33	2.14
Halauxifen-methyl[c]	5	63	84	85	11	15	2.28
Saflufenacil[b]	25	79	86	87	6	22	2.03
Dicamba	600	64	89	94	10	3	2.32
SE		2	2	2	4	6	0.051
Bromoxynil × Tankmix partner		**	**	**	**	*	NS

Note. Abbreviations: SE, standard error.

**Significant at $P < 0.01$; *significant at $P < 0.10$; NS, non-significant. [a] Density and biomass were collected 8 WAA. [b] Merge, 1 L ha^{-1} was added to all treatments with saflufenacil. [c] MSO, 1 % v/v or 0.5 % v/v was added to all treatments with halauxifen-methyl and tiafenacil, respectively.

In conclusion, limited research exists on GR Canada fleabane control with bromoxynil-based tankmixtures applied PP to soybean. Glyphosate plus bromoxynil plus metribuzin, tiafenacil, pyraflufen-ethyl/2,4-D, 2,4-D ester, halauxifen-methyl, saflufenacil or dicamba caused ≤ 10% visible soybean injury. At 2 WAA, when saflufenacil was added to glyphosate plus bromoxynil GR Canada fleabane control improved from 52 to 85%. At 4 WAA, when dicamba was added to glyphosate plus bromoxynil GR Canada fleabane control improved from 58 to 95%. At 8 WAA, when dicamba was added to glyphosate plus bromoxynil GR Canada fleabane control improved from 69 to 97%. At 2 WAA, the interaction between glyphosate plus bromoxynil plus tiafenacil, pyraflufen-ethyl/2,4-D or halauxifen-methyl was antagonistic. At 4 WAA, the interaction between glyphosate plus bromoxynil plus pyraflufen-ethyl/2,4-D, 2,4-D ester or halauxifen-methyl was antagonistic. At 8 WAA, the interaction between glyphosate plus bromoxynil plus tiafenacil or halauxifen-methyl was antagonistic; while the interaction between glyphosate plus bromoxynil plus pyraflufen-ethyl/2,4-D was synergistic. The interactions for the other tankmixtures were additive. In conclusion, the addition of saflufenacil to glyphosate plus bromoxynil at 2 WAA and dicamba to glyphosate plus bromoxynil at 4 and 8 WAA improves the level and consistency of GR Canada fleabane control applied PP to soybean.

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