# Preplant Herbicide Programs for the Control of Multiple-Herbicide-Resistant Waterhemp in No-Till Corn

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Received: December 21, 2023	Accepted: February 7, 2024	Online Published: March 15, 2024
doi:10.5539/jas.v16n4p1	URL: https://doi.org/10.5539/jas.v16	5n4p1

# Abstract

Multiple-herbicide-resistant (MHR) waterhemp has been confirmed in 18 Ontario counties. A total of four experiments were conducted in commercial fields with MHR waterhemp in 2020 and 2021 to evaluate preplant (PP) herbicide tank mixtures for control of MHR waterhemp in no-till corn. There was minimal visible corn injury from the herbicide treatments evaluated. At 2 WAA, all herbicide tank mixtures provided greater than 90% control of MHR waterhemp except for pyroxasulfone/carfentrazone + atrazine which controlled MHR waterhemp 85%. WAA, S-metolachlor/mesotrione/atrazine, S-metolachlor/mesotrione/bicyclopyrone/atrazine, At 12 and diflufenican + atrazine + flufenacet controlled MHR waterhemp 86, 91, and 98%, respectively; all other herbicide tank mixtures provided 65 to 83% control. At 8 WAA, S-metolachlor/mesotrione/bicyclopyrone/atrazine and diflufenican + atrazine + flufenacet reduced MHR waterhemp density 97 and 100%, respectively similar to the weed-free control; all other herbicide tank mixtures reduced MHR waterhemp density 64 to 96%. At 8 WAA, S-saflufenacil/dimethenamid-P + mesotrione, S-metolachlor/mesotrione/atrazine, tembotrione + dicamba + flufenacet, metolachlor/mesotrione/bicyclopyrone/atrazine, and diflufenican + atrazine + flufenacet reduced MHR waterhemp biomass 96, 97, 98, 98, and 100%, respectively; all other herbicide tank mixtures reduced MHR waterhemp biomass 72 to 93%. MHR waterhemp interference reduced corn yield 80% in this study. All herbicide tank mixtures resulted in corn yield that was similar to the weed-free control. Among the herbicide tank mixtures evaluated S-metolachlor/mesotrione/bicyclopyrone/atrazine and diflufenican + atrazine + flufenacet provided the greatest control of MHR waterhemp in no-till corn.

Keywords: glyphosate-resistant, multiple-herbicide-resistant, waterhemp, waterhemp biomass, waterhemp control, waterhemp density, corn yield

# 1. Introduction

Waterhemp [*Amaranthus tuberculatus* (Moq.) J.D. Sauer] is a small-seeded, summer annual, broadleaf weed with an extended emergence pattern, has high genetic diversity, is a prolific seed producer, is very competitive, and has spread rapidly throughout the primary corn and soybean production areas of North America (Hartzler et al., 2004; Johnson et al., 2012; Schryver et al., 2017; Vyn et al., 2007). Waterhemp has the potential to become one of the most problematic weeds in Ontario, as it already is in the midwestern USA states. Vyn et al. (2006) and Steckel and Sprague (2004) reported as much as 74% yield reduction in corn from waterhemp interference.

Glyphosate-resistant (GR) waterhemp was first confirmed in Ontario in 2014 (Schryver et al., 2017). Multiple-herbicide-resistant (MHR) waterhemp has now been confirmed in 18 Ontario counties. In Ontario, waterhemp has evolved resistance to Group 2 (imazethapyr), Group 5 (atrazine), Group 9 (glyphosate), Group 14 (lactofen) and Group 27 (mesotrione) herbicides (Benoit et al., 2019a; Symington et al., 2022). Growers need herbicides/herbicide mixtures that have multiple modes of action for the control of MHR waterhemp to avoid the economic losses associated with this problematic weed in Ontario. A recent study has estimated that uncontrolled MHR waterhemp can reduce the yield of grain corn, fodder corn, soybean, winter wheat, spring barley, spring oats, spring wheat, spring mixed grain, white bean, colored dry bean, and canola 19, 19, 42, 3, 12, 12, 12, 12, 50, 50, and 15% valued at approximately \$K 3,064, 362, 7,103, 130, 34, 24, 39, 20, 213, 269 and 30, respectively for total potential loss of \$11.3M per year in Ontario (Soltani et al., 2023).

The is little information in the peer-reviewed literature on preplant (PP) herbicide options to effectively control MHR waterhemp in no-till corn. Herbicide tank mixtures such as saflufenacil/dimethenamid-P,

saflufenacil/dimethenamid-P + mesotrione, isoxaflutole + atrazine, isoxaflutole + atrazine + flufenacet, tembotrione + dicamba + flufenacet, tembotrione + atrazine + flufenacet, pyroxasulfone/carfentrazone + atrazine, mesotrione + atrazine, S-metolachlor/mesotrione/atrazine, S-metolachlor/mesotrione/bicyclopyrone, S-metolachlor/mesotrione/bicyclopyrone/atrazine, or S-metolachlor/mesotrione + atrazine applied preplant (PP) have the potential to control MHR waterhemp in no-till corn. Diflufenican is a newly introduced selective broadleaf herbicide from Bayer CropScience from the pyridinecarboxamide chemical family (Group 12) that has the potential to be used alone or in combination with atrazine and flufenacet to control MHR waterhemp in no-till corn (Bayer 2022). To our knowledge, no study has cumulatively compared the efficacy of these herbicide mixtures for the control of MHR waterhemp in no-till corn. Additionally, there is no published information on the efficacy of herbicide tankmixes with diflufenican for the control MHR waterhemp in corn in Ontario.

The objective of this experiment was to evaluate various preplant herbicide tank mixtures for the control of MHR waterhemp in no-till corn.

## 2. Materials and Methods

#### 2.1 Experimental Methods

A total of four experiments were established in commercial fields in southwestern Ontario with a high density of MHR waterhemp in 2020 (2 trials) and 2021 (2 trials). All sites contained natural infestations of confirmed MHR waterhemp. The experimental design was a randomized complete block design (RCBD) with four replications. Replications were separated by a 2 m alleyway. Treatments included a non-treated weedy control and a weed-free control and saflufenacil/dimethenamid-P, isoxaflutole + atrazine, pvroxasulfone/carfentrazone + atrazine. tembotrione +atrazine +flufenacet, S-metolachlor/mesotrione/bicyclopyrone, mesotrione + atrazine, saflufenacil/dimethenamid-P + glyphosate/S-metolachlor/mesotrione + atrazine, mesotrione. tembotrione dicamba +flufenacet, isoxaflutole +atrazine +flufenacet. +S-metolachlor/mesotrione/atrazine, S-metolachlor/mesotrione/bicyclopyrone/atrazine, and diflufenican + atrazine + flufenacet with adjuvants at rates as listed in Tables 2 and 3. Glyphosate (900 g at  $ha^{-1}$ ) was added to all of the above tank mixtures, with the exception of glyphosate/S-metolachlor/mesotrione + atrazine, to control all other weed species present including glyphosate-susceptible waterhemp. Glyphosate-resistant corn hybrids, DKC45-65RIB<sup>®</sup> and DKC42-60RIB<sup>®</sup> (Bayer CropScience Canada Inc., 160 Quarry Boulevard SE, Calgary, Alberta, Canada, T2C 3G3) were planted at a seeding rate of approximately 80,000 seeds ha<sup>-1</sup> in 2020 and 2021. Corn was planted approximately 4 cm deep in rows spaced 75 cm apart. Plots were 8 m in length and 2.25 m wide. Experimental plots were fertilized according to provincial recommendations for corn production in Ontario (OMAFRA, 2017).

Active ingredients	Trade name	Manufacturer		
Atrazine	Aatrex Liquid 480	Syngenta Canada Inc., Guelph, ON		
Dicamba	Xtendimax	Bayer CropScience Canada, Calgary, AB		
Diflufenican	-	Bayer CropScience Canada, Calgary, AB		
Flufenacet	Cadou SC	Bayer CropScience Canada, Calgary, AB		
Glyphosate	Roundup WeatherMAX	Bayer CropScience Canada, Calgary, AB		
Glyphosate/S-metolachlor/mesotrione	Halex GT Herbicide	Syngenta Canada Inc., Guelph, ON		
Isoxaflutole	Converge Flexx Herbicide	Bayer CropScience Canada, Calgary, AB		
Mesotrione	Callisto 480SC Herbicide	Syngenta Canada Inc., Guelph, ON		
Pyroxasulfone/carfentrazone	Focus	FMC of Canada Limited, Mississauga, ON		
Saflufenacil/dimethenamid-P	Integrity	BASF Canada, Mississauga, ON		
S-metolachlor/mesotrione/atrazine	Lumax EZ Herbicide	Syngenta Canada Inc., Guelph, ON		
S-metolachlor/mesotrione/bicyclopyrone	Acuron Flexi XR	Syngenta Canada Inc., Guelph, ON		
S-metolachlor/mesotrione/bicyclopyrone/atrazine	Acuron XR	Syngenta Canada Inc., Guelph, ON		
Tembotrione	Laudis Herbicide	Bayer CropScience Canada, Calgary, AB		

Table 1. Active ingredients, trade names, and manufacturers applied preplant <sup>a</sup>

*Note.* <sup>a</sup> Specimen labels for each product and manufacturer contact information can be found at https://pr-rp.hc-sc.gc.ca/ls-re/index-eng.php.

Herbicide treatments were applied preplant (not incorporated) when the MHR waterhemp in the nontreated control plots was up to 10 cm in height/diameter. All herbicide treatments were sprayed with a CO<sub>2</sub>-pressurized

backpack plot sprayer with 4 ULD120-02 spray nozzles (Pentair, 375 5<sup>th</sup> Avenue NW, New Brighton, Minnesota, USA, 55112) at 50 cm spacing calibrated to deliver 200 L ha<sup>-1</sup> spray volume at 240 kPa pressure producing a spray width of 2 m.

Visible corn injury at 1, 2, 4, and 8 weeks after emergence (WAE) was assessed on a scale of 0 to 100% (0 = no injury and 100 = corn death). Visible MHR waterhemp control at 2, 4, 8, and 12 weeks after application (WAA) was evaluated on a scale of 0 to 100% (0 = no control and 100 = complete control) as a visual assessment of MHR waterhemp biomass reduction compared to the MHR waterhemp aboveground biomass in the nontreated control within the corresponding replicate. Density and biomass of MHR waterhemp were collected at 8 WAA by counting and cutting the MHR waterhemp plants at the soil surface within two 0.25 m<sup>2</sup> randomly placed quadrats in each plot. The cut MHR waterhemp plants from each plot were placed in separate paper bags and dried at 60 °C until the biomass reached constant moisture at which time the waterhemp dry biomass was weighed and recorded. Corn was harvested at maturity with a small-plot research combine from the middle two rows of each plot. Grain corn yields were corrected to 15.5% moisture before data analysis.

## 2.2 Statistical Analysis

The statistical analysis was conducted using SAS Studio v9.4, OnDemand for Academics (SAS Institute, Cary, NC). Data were analyzed using generalized linear mixed models via the GLIMMIX procedure. The variance was partitioned into the fixed effect of herbicide treatment and the random effects of environment (location-year combinations), block nested within environment, and the environment-by-treatment interaction, for all parameters. Waterhemp control at 2, 4, 8, and 12 WAA were arcsine square-root transformed prior to analysis using a normal distribution with identity link. Waterhemp density and biomass were analyzed using the lognormal distribution with identity link while corn yield was analyzed using a normal distribution with identity link. The Pearson chi-square/degrees of freedom ratio and Shapiro-wilk statistic were examined for each parameter to determine model fitness and avoid potential overdispersion. Studentized residual plots and normal probability plots were used to confirm homogeneity of variance and the assumptions of normality, respectively. Arcsine square root transformed data were back-transformed post-analysis. Data analyzed using a lognormal distribution were back-transformed using the omega method (M Edwards, Ontario Agricultural College Statistician, University of Guelph, personal communication). Mean estimates were separated using Tukey-Kramer Least Significant Difference (LSD). Density and biomass treatment means were compared to the weed-free control using P-values produced via the Least Squares Means output. An alpha level of 0.05 was chosen for this analysis.

## 3. Results and Discussion

# 3.1 Corn Injury

There was no visible corn injury from the herbicide treatments evaluated at 1, 2, and 4 WAE except diflufenican + atrazine + flufenacet that caused  $\leq$  5% corn injury at 1, 2, and 4 WAE (data not shown). Results are similar to Willemse et al. (2022) who observed no crop injury with isoxaflutole + atrazine, saflufenacil/dimethenamid-P, or *S*-metolachlor/mesotrione/bicyclopyrone/atrazine applied preemergence (PRE) in corn. In other studies, mixtures of isoxaflutole + flufenacet, *S*-metolachlor + isoxaflutole + atrazine, isoxaflutole + atrazine, isoxaflutole + flufenacet + atrazine, and flufenacet + isoxaflutole + flumetsulam + clopyralid applied preplant incorporated (PPI)/preemergence (PRE) caused as much as 11, 9, 7, 14, and 14% visible injury in corn, respectively (Johnson et al., 2012). However, *S*-metolachlor + atrazine, *S*-metolachlor + atrazine + flumetsulam + clopyralid, and *S*-metolachlor + flumetsulam + clopyralid applied PPI/PRE caused no/minimal injury in corn (Johnson et al., 2012).

# 3.2 MHR Waterhemp Control

At 2 WAA, all herbicide tank mixtures evaluated provided greater than 90% control of MHR waterhemp except for pyroxasulfone/carfentrazone + atrazine which provided 85% control of MHR waterhemp in no-till corn (Table 2).

Herbicide treatment	Dete	Visible control			
Herbicide treatment	Rate	2 WAA	4 WAA	8 WAA	12 WAA
	g ae or ai ha <sup>-1</sup>	%			
Saflufenacil/dimethenamid-P	900+75/660	90 ab	83 b	69 c	65 c
Isoxaflutole+atrazine	900+105+1063	91 ab	88 b	68 c	66 c
Pyroxasulfone/carfentrazone+atrazine	900+150/18+1008	85 b	83 b	77 bc	74 bc
Tembotrione+atrazine+flufenacet	900+92+500+500	94 ab	92 ab	77 bc	75 bc
S-metolachlor/mesotrione/bicyclopyrone	900+1484/165/41	91 ab	88 b	79 bc	76 bc
Glyphosate/S-metolachlor/mesotrione+atrazine <sup>b</sup>	1050/1050/105+280	93 ab	88 b	80 bc	77 bc
Mesotrione+atrazine <sup>b</sup>	900+140+280	94 ab	91 b	80 bc	78 bc
Saflufenacil/dimethenamid-P+mesotrione	900+75/660+140	94 ab	93 ab	83 bc	81 bc
Tembotrione+dicamba+flufenacet	900+92+600+500	94 ab	95 ab	84 bc	83 bc
S-metolachlor/mesotrione/atrazine	900+1393/139/524	95 a	94 ab	88 bc	86 abc
S-metolachlor/mesotrione/bicyclopyrone/atrazine	900+1389/154/38/649	96 a	95 ab	92 ab	91 ab
Diflufenican+atrazine+flufenacet	900+605+500+500	97 a	98 a	98 a	98 a

Table 2. Multiple-herbicide-resistant (MHR) waterhemp control 2, 4, 8, and 12 weeks after application from herbicide tank-mixtures applied preplant in no-till corn in field trials across Ontario, Canada in 2020 and 2021 <sup>a,c</sup>

*Note.* <sup>a</sup> Means followed by the same letter within a column are not significantly different according to Tukey-Kramer LSD (P > 0.05).

<sup>b</sup> Treatments included Agral 90 (Syngenta Canada Inc., Guelph, ON) (0.2% v/v).

<sup>c</sup> Abbreviations: WAA; weeks after application.

At 4 WAA, tembotrione + atrazine + flufenacet, saflufenacil/dimethenamid-P + mesotrione, tembotrione + dicamba + flufenacet, *S*-metolachlor/mesotrione/atrazine, and *S*-metolachlor/mesotrione/bicyclopyrone/atrazine, and diflufenican + atrazine + flufenacet controlled MHR waterhemp similarly at 92 to 98% (Table 2). This is similar to another study in which *S*-metolachlor/mesotrione/bicyclopyrone/atrazine applied PRE controlled MHR waterhemp 94% in corn 4 WAA (Willemse et al., 2022). Other herbicide tank mixtures evaluated provided good (83-91%) control of MHR waterhemp in no-till corn (Table 2).

At 8 WAA, *S*-metolachlor/mesotrione/bicyclopyrone/atrazine and diflufenican + atrazine + flufenacet controlled MHR waterhemp 92 and 98%, respectively (Table 2); the other herbicide tank mixtures controlled MHR waterhemp 68 to 88%.

At 12 WAA, *S*-metolachlor/mesotrione/atrazine, *S*-metolachlor/mesotrione/bicyclopyrone/atrazine, and diflufenican + atrazine + flufenacet controlled MHR waterhemp similarly at 86, 91, and 98%, respectively; all other herbicide tank mixtures provided 65 to 83% control (Table 2).

In other studies, isoxaflutole + atrazine applied PRE provided 70-97, 77-97, and 78-97% control of MHR waterhemp at 4, 8, and 12 WAA in corn, respectively (Willemse et al., 2022). Also, saflufenacil/dimethenamid-P applied PRE provided 88-99, 94-98, and 95-99% control of MHR waterhemp at 4, 8, and 12 WAA in corn, The respectively (Willemse al., 2022). et control was more consistent with S-metolachlor/mesotrione/bicyclopyrone/atrazine which provided 94-99, 93-99, and 95-99% control of MHR waterhemp in corn, respectively (Willemse et al., 2022). Similarly, Benoit et al. (2019b) observed 84% MHR waterhemp control with isoxaflutole + atrazine and 94% MHR waterhemp control with S-metolachlor/mesotrione/bicyclopyrone/atrazine at 4 WAA in corn. Vyn et al. (2006) observed 97% control of a triazine-resistant waterhemp population 10 WAA with isoxaflutole + atrazine applied PRE in corn. Sarangi and Jhala (2017)observed greater than 95% control of MHR waterhemp with S-metolachlor/mesotrione/bicyclopyrone/atrazine applied PRE in corn. Benoit et al. (2019b), Steckel et al. (2002), and Vyn et al. (2006) also reported 91%, 98%, and 97% to 100% control of MHR waterhemp with saflufenacil/dimethenamid-P, dicamba/atrazine, and S-metolachlor/atrazine at 4 WAA in corn, respectively.

## 3.3 MHR Waterhemp Density

At 8 WAA, saflufenacil/dimethenamid-P, isoxaflutole + atrazine, pyroxasulfone/carfentrazone + atrazine, tembotrione + atrazine + flufenacet, S-metolachlor/mesotrione/bicyclopyrone, glyphosate/S-metolachlor/mesotrione + atrazine, mesotrione + atrazine, saflufenacil/dimethenamid-P +

+dicamba + flufenacet. isoxaflutole +flufenacet. mesotrione. tembotrione  $^{+}$ atrazine S-metolachlor/mesotrione/atrazine. S-metolachlor/mesotrione/bicvclopyrone/atrazine. and diflufenican + atrazine + flufenacet reduced MHR waterhemp density 93, 70, 64, 90, 75, 83, 87, 94, 94, 81, 96, 97, and 100%, In other studies, isoxaflutole + atrazine, saflufenacil/dimethenamid-P, respectively (Table 3). S-metolachlor/mesotrione/bicyclopyrone/atrazine, and mesotrione + atrazine applied PRE reduced MHR waterhemp density 94, 99, 99, and 89% (Willemse et al. 2022). Benoit et al. (2019b) reported that mesotrione + atrazine. isoxaflutole + atrazine. saflufenacil/dimethenamid-P, S-metolachlor/atrazine. S-metolachlor/mesotrione/atrazine and S-metolachlor/mesotrione/bicvclopyrone/atrazine applied PRE reduced MHR waterhemp density 92, 94, 98, 96, 98 and 97%, respectively.

Table 3. Multiple-herbicide resistant waterhemp density and biomass 8 weeks after application and corn yield from herbicide tank mixtures applied preplant in no-till corn in field trials across Ontario, Canada in 2020 and  $2021^{a,c}$ 

Herbicide treatment	Rate	Density	Biomass	Yield
	g ae or ai ha <sup>-1</sup>	Plants m <sup>-2</sup>	g m <sup>-2</sup>	kg ha <sup>-1</sup>
Non-treated control	-	417 e	697 d	1,980 b
Weed-free control	-	0 a	0 a	9,860 a
Saflufenacil/dimethenamid-P	900+75/660	69 cd	176 c	8,660 a
Isoxaflutole+atrazine	900+105+1063	124 de	92 c	8,400 a
Pyroxasulfone/carfentrazone+atrazine	900+150/18+1008	152 cd	198 c	7,560 a
Tembotrione+atrazine+flufenacet	900+92+500+500	40 cd	74 c	9,110 a
S-metolachlor/mesotrione/bicyclopyrone	900+1484/165/41	105 cd	111 c	8,420 a
Glyphosate/S-metolachlor/mesotrione+atrazine <sup>b</sup>	1050/1050/105+280	71 cd	87 c	8,400 a
Mesotrione+atrazine <sup>b</sup>	900+140+280	54 cd	51 c	9,360 a
Saflufenacil/dimethenamid-P+mesotrione	900+75/660+140	26 cd	32 bc	9,610 a
Tembotrione+dicamba+flufenacet	900+92+600+500	27 cd	18 bc	9,170 a
Isoxaflutole+atrazine+flufenacet	900+79+500+500	78 cd	85 c	7,860 a
S-metolachlor/mesotrione/atrazine	900+1393/139/524	18 c	22 bc	9,190 a
S-metolachlor/mesotrione/bicyclopyrone/atrazine	900+1389/154/38/649	13 bc	19 abc	9,650 a
Diflufenican+atrazine+flufenacet	900+605+500+500	0 ab	0 ab	10,460 a

*Note.* <sup>a</sup> Means followed by the same letter within a column are not significantly different according to Tukey-Kramer LSD (P > 0.05).

<sup>b</sup> Treatments included Agral 90 (Syngenta Canada Inc., Guelph, ON) (0.2% v/v).

<sup>c</sup> Abbreviations: WAA; weeks after application.

## 3.4 MHR Waterhemp Aboveground Biomass

At 8 WAA, saflufenacil/dimethenamid-P, isoxaflutole + atrazine, pyroxasulfone/carfentrazone + atrazine, tembotrione atrazine flufenacet. S-metolachlor/mesotrione/bicvclopvrone. ++glyphosate/S-metolachlor/mesotrione + atrazine, mesotrione + atrazine, saflufenacil/dimethenamid-P + mesotrione. tembotrione + dicamba  $^{+}$ flufenacet. isoxaflutole +atrazine +flufenacet. S-metolachlor/mesotrione/atrazine, S-metolachlor/mesotrione/bicyclopyrone/atrazine, and diflufenican + atrazine + flufenacet reduced MHR waterhemp aboveground biomass 75, 87, 72, 90, 84, 88, 93, 96, 98, 88, 97, 98, and 100%, respectively (Table 3). Similar to this study, Benoit et al. (2019b) reported mesotrione + atrazine, isoxaflutole + atrazine, saflufenacil/dimethenamid-P, S-metolachlor/atrazine, S-metolachlor/mesotrione/atrazine, and S-metolachlor/mesotrione/bicyclopyrone/atrazine applied PRE reduced MHR waterhemp aboveground biomass 93, 94, 94, 96, 98, and 96%, respectively.

## 3.5 Corn Yield

MHR waterhemp interference reduced corn yield 80% in this study (Table 3). All herbicide mixtures reduced waterhemp interference so that corn yield was similar to the weed-free control (Table 3). Results are similar to Willemse et al. (2022) who observed no yield loss in corn from waterhemp interference with isoxaflutole + atrazine, saflufenacil/dimethenamid-P, or *S*-metolachlor/mesotrione/bicyclopyrone/atrazine applied PRE in corn. Benoit et al. (2019b) also reported no yield loss from waterhemp interference with mesotrione + atrazine,

isoxaflutole + atrazine, saflufenacil/dimethenamid-P, S-metolachlor/atrazine, S-metolachlor/mesotrione/atrazine, and S-metolachlor/mesotrione/bicyclopyrone/atrazine applied PRE.

#### 4. Conclusions

Among the herbicide mixtures evaluated, *S*-metolachlor/mesotrione/atrazine, *S*-metolachlor/mesotrione/bicyclopyrone/atrazine, and diflufenican + atrazine + flufenacet provided the highest control of MHR waterhemp in no-till corn. Other herbicide tank mixtures evaluated provided 65 to 83% control of MHR waterhemp by 12 WAA.

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## Acknowledgments

We would like to acknowledge the technical support provided by Chris Kramer and the summer students at the University of Guelph Ridgetown Campus.

## **Authors Contributions**

Drs. Peter Sikkema and Nader Soltani were responsible for the study design and writing of this manuscript. Christian A. Willemse conducted the statistical analysis of the data collected.

## Funding

Funding for this research was provided in part by The Grain Farmers of Ontario (GFO) and the Alliance Funding program of the Ontario Ministry of Agriculture, Food and Rural Affairs.

## **Competing Interests**

No other competing interests have been declared.

#### **Informed Consent**

Obtained.

## **Ethics Approval**

The Publication Ethics Committee of the Canadian Center of Science and Education.

The journal's policies adhere to the Core Practices established by the Committee on Publication Ethics (COPE).

## **Provenance and Peer Review**

Not commissioned; externally double-blind peer reviewed.

## Data Availability Statement

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

#### **Data Sharing Statement**

No additional data are available.

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