

Preplant followed by postemergence herbicide programs and biologically effective rate of metribuzin for control of glyphosate-resistant common ragweed (*Ambrosia artemisiifolia*) in soybean

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Abstract: With no new herbicides with a novel mode of action in the marketplace in corn–soybean cropping systems, control of glyphosate-resistant (GR) weeds requires the re-evaluation of existing herbicides. This necessitates a renewed focus on using herbicide tank-mixes and sequential herbicide programs while also striving to minimize the environmental impact of weed management. Preliminary research identified four preplant (PP) herbicides (2,4-D, saflufenacil/dimethenamid-P, linuron, and metribuzin) and one postemergence (POST) herbicide (fomesafen) that provided good but inconsistent control of GR common ragweed when applied alone in soybean. The objectives of this study were to determine the biologically effective rate of metribuzin and evaluate PP followed by POST herbicide programs for control of GR common ragweed in soybean. The aforementioned PP herbicides reduced GR common ragweed density and aboveground biomass by 82%–94% and 55%–89%, respectively. In contrast, a PP herbicide followed by fomesafen applied POST decreased common ragweed densities and aboveground biomass by 97%–99% and 93%–98%, respectively. Metribuzin applied at 824 and 1015 g a.i. ha⁻¹ controlled GR common ragweed 90% at 4 and 8 wk after application, respectively. This study concludes that GR common ragweed can be controlled with a PP followed by POST herbicide program and metribuzin has potential for control of GR common ragweed in soybean.

Key words: glyphosate-resistant, common ragweed, metribuzin, biologically effective rate.

Résumé : Faute d'herbicide au nouveau mode d'action pour les systèmes culturaux maïs-soja sur le marché, la lutte contre les adventices résistant au glyphosate (RG) nécessite une réévaluation des désherbants existants. Pour cela, on doit examiner avec un œil neuf les mélanges et les applications séquentielles, tout en s'efforçant d'en minimiser l'impact pour l'environnement. Des recherches préliminaires ont permis d'identifier quatre herbicides de présemis (PRÉ) (2,4-D, saflufénacil/diméthénamide-P, linuron et métribuzine) et un herbicide de post-levée (POST) (fomesafen) qui luttent bien, quoique de façon peu uniforme, contre la petite herbe à poux RG quand on les applique seuls aux cultures de soja. La présente étude devait établir la dose biologiquement efficace de métribuzine et évaluer l'application successive d'herbicides PRÉ et POST pour venir à bout de la petite herbe à poux RG dans le soja. Les herbicides PRÉ mentionnés ci-dessus réduisent respectivement la densité de l'adventice et la biomasse de ses organes aériens de 82 à 94 % et de 55 à 89 %. En revanche, l'application d'un herbicide PRÉ puis de fomesafen après la levée diminue respectivement la densité de la petite herbe à poux RG et la biomasse de ses organes aériens de 97 à 99 % et de 93 à 98 %. L'application de 824 g ou de 1015 g de métribuzine par hectare détruit 90 % de la petite herbe à poux RG, quatre et huit semaines après l'application, respectivement. Les auteurs en concluent qu'on peut lutter contre l'adventice avec un herbicide PRÉ, puis l'application d'un herbicide POST, et que la métribuzine pourrait combattre la petite herbe à poux RG dans les cultures de soja. [Traduit par la Rédaction]

Mots-clés : résistance au glyphosate, petite herbe à poux, métribuzine, taux biologiquement efficace.

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Introduction

Since the commercialization of glyphosate-resistant (GR) soybean and canola in 1996, other GR crops such as corn, alfalfa, cotton, and sugar beet have been commercialized. Producers have adopted this technology rapidly due to its excellent weed control, wide margin of crop safety, low cost, and wide window of glyphosate application. Because glyphosate is an efficacious, non-selective, broad-spectrum herbicide, its widespread popularity has resulted in a decline in the use of other herbicides. However, with the 37 GR weeds documented worldwide, alternative and older chemistries are being brought back to the forefront (Heap 2016). Cultivation, cover crops, new crop technologies, and herbicide programs with multiple effective modes of action are all being investigated as components of an integrated weed management program.

Common ragweed was first reported as resistant to glyphosate in 2004 in Missouri and Arkansas (Pollard et al. 2004; Brewer and Oliver 2009). In 2011, common ragweed seed was collected from a suspicious site near Windsor, ON, and confirmed to be resistant to glyphosate in the spring of 2012. A survey by Van Wely et al. (2014) confirmed GR common ragweed at five additional sites from seed collected from 24 sites in 2012 and 2013. There are four herbicide groups with known resistant common ragweed biotypes including chlorimuron-ethyl, cloransulam-ethyl, and imazethapyr (Group 2), atrazine (Group 5), linuron (Group 7), and glyphosate (Group 9). Most GR biotypes in Ontario are also resistant to acetolactate synthase inhibiting herbicides (Heap 2016), which reduces the number of effective herbicide options for common ragweed control.

Common ragweed is an annual, broadleaf weed with a prolonged emergence period in Ontario, and therefore, season-long control is required. Emergence commences in April and May, coinciding with the planting of annual crops such as corn and soybean. Bassett and Crompton (1982) reported that 90% of common ragweed emerges by mid-June. Common ragweed emergence correlates with soil temperature, and so seed close to the surface emerges first, followed by seed at depth as the soil warms (Deen et al. 1998; Shrestha et al. 1999). Though the length of residual activity of herbicides varies, the long emergence period of common ragweed means that more than one herbicide application may be required for season-long control. A preemergence (PRE) followed by postemergence (POST) herbicide program helps in two main ways: first, by controlling weeds during early crop growth, thereby minimizing crop yield loss due to early weed interference, and second, by reducing weed density at the time of the POST application, thereby improving weed control and reducing the selection intensity for herbicide-resistant weeds. Sequential weed control programs, therefore, help to control any weeds not controlled with the first herbicide application,

whether due to resistance or late emergence. Although soybean yield may not be affected by late-emerging weeds, these weeds can contribute to the seed bank, a special concern with GR weeds (Jordan et al. 2010).

Studies conducted by Van Wely et al. (2014) near Windsor evaluated all the preplant (PP) and POST herbicides registered in Ontario for broadleaf weed control in soybean. The results found that the most efficacious herbicides applied PP for the control of GR common ragweed at 4 wk after application (WAA) were 2,4-D ester (up to 83% control), saflufenacil (up to 95% control), linuron (up to 99% control), and metribuzin (up to 98% control). Fomesafen was the most efficacious POST herbicide, providing up to 98% control of GR common ragweed in soybean at 4 WAA (Van Wely et al. 2014). Because none of the herbicides registered in Ontario provided acceptable control of GR common ragweed on their own, a study with PP followed by POST herbicide programs was established to determine whether more consistent control of GR common ragweed could be obtained in soybean (Van Wely et al. 2014). This approach to weed management is also consistent with best management practices, which recommend the use of herbicides with at least two effective modes of action in each field against the same target species, every year. This can be achieved through either sequential herbicide applications or herbicide tank-mixes. This study explores the use of sequential herbicide applications for the control of GR common ragweed in Ontario.

Some of the herbicide alternatives to glyphosate are older and have had minimal use for the last several years. Metribuzin was first marketed in 1971 (Lewis et al. 2016) and is currently used for weed management in soybean applied alone or in a tank-mix. Van Wely et al. (2014) reported that metribuzin (1120 g a.i. ha⁻¹) applied PP provided 86%–98% and 80%–95% control of GR common ragweed at 4 and 8 WAA, respectively, in Ontario. However, this high rate of metribuzin is cost-prohibitive for soybean growers and can cause soybean injury depending on the soil type and cultivar sensitivity. In addition, for environmental reasons, there is a push to reduce the amount of herbicide applied by growers, and therefore, data are needed to determine the lowest effective rate of metribuzin for the control of GR common ragweed.

The objectives of this research were to determine (i) whether season-long control of GR common ragweed can be achieved using a PP followed by POST herbicide program and (ii) the lowest effective rate of metribuzin for the control of GR common ragweed in soybean.

Materials and Methods

Field experiments were conducted near Windsor (2014, 2015), and Adams, NE (2015), in fields with confirmed GR common ragweed. The Windsor site consisted of a sandy clay loam (49% sand, 26% silt, and 25% clay) with

Table 1. Herbicides and herbicide timings for preplant (PP) and postemergence (POST) control of glyphosate-resistant common ragweed.

| Treatment | Preplant ^{ab} | Rate (g a.i. ha ⁻¹) | POST ^{ac} | Rate (g a.i. ha ⁻¹) |
|-----------|-------------------------------------|------------------------------------|--------------------|------------------------------------|
| 1 | Glyphosate (9) ^c | 900 | — | — |
| 2 | 2,4-D ester (4) | 500 | — | — |
| 3 | Saflufenacil/dimethenamid-P (14/15) | 245 | — | — |
| 4 | Linuron (7) | 2250 | — | — |
| 5 | Metribuzin (5) | 1120 | — | — |
| 6 | — | — | Fomesafen (14) | 240 |
| 7 | 2,4-D ester (4) | 500 | Fomesafen (14) | 240 |
| 8 | Saflufenacil/dimethenamid-P (14/15) | 245 | Fomesafen (14) | 240 |
| 9 | Linuron (7) | 2250 | Fomesafen (14) | 240 |
| 10 | Metribuzin (5) | 1120 | Fomesafen (14) | 240 |

Note: All herbicide applications included glyphosate at 900 g a.e. ha⁻¹.

^aPreplant herbicides were applied when common ragweed plants were up to 4 cm tall.

^bPOST herbicides were applied when common ragweed plants were up to 10 cm tall.

^cThe number in parentheses refers to the mode of action of herbicide classified by the Weed Science Society of America.

3% organic matter. At the Adams site, the soil was a clay loam (25% sand, 38% silt, and 37% clay) with 2.7% organic matter. Experiments were arranged as a randomized complete block design with three or four replicates, with each replicate containing a nontreated and a weed-free control. In the PP followed by POST study, herbicide treatments were organized so that PP and POST herbicides were applied alone and in sequence (Table 1). In the metribuzin biologically effective rate study, metribuzin was applied at 35, 70, 140, 280, 560, 840, 1120, 2240, and 4480 g a.i. ha⁻¹. Glyphosate was applied at 900 g a.e. ha⁻¹ to the entire experimental area to remove the confounding effect of other weed species. Plots were 2.25 m wide (three soybean rows spaced 0.75 m apart) and 6 m long. The herbicides were applied with a CO₂-pressurized backpack sprayer at an application rate of 200 L ha⁻¹ at 210 kPa equipped with a 1 m wide spray boom with three ULD 120-20 nozzles (Hypro, New Brighton, MN) spaced 50 cm apart (1.5 m spray width). Herbicides were applied to the centre 1.5 m of each plot.

The hypotheses of this research were to determine if (i) full-season control of GR common ragweed requires a sequential herbicide program and (ii) the full registered rate (1120 g a.i. ha⁻¹) of metribuzin would be required for the control of GR common ragweed. The first hypothesis was tested by spraying either single or sequential applications of herbicides. The second hypothesis required only a single application of herbicide, at the PP timing. The PP applications were made prior to soybean seeding, when common ragweed had emerged and were 2–4 cm in height (Table 1). The POST herbicides were applied when common ragweed had reached up to 10 cm in height (Table 1). Soybean injury was evaluated at 2 and 3 wk after application (WAA) or just prior to the POST application. Control ratings were taken prior to the POST application and then at 4 and 8 wk after POST

(WAP). Common ragweed plants were counted from two 0.25-m² quadrants per plot and the plants were cut at the soil line, placed in paper bags, dried in a kiln, and the aboveground biomass was recorded. Density and aboveground biomass were recorded prior to the POST application for treatments 1–7, 8 WAP for the PP followed by POST experiment, and 4 WAA for the metribuzin biologically effective rate experiment.

Statistical analysis

Data were analyzed using SAS 9.4 (SAS Institute, Inc., Cary, NC). In the PP followed by POST experiment, data were subjected to analysis of variance using PROC MIXED in SAS. Means separation was completed using Tukey's honest significant difference. For the metribuzin biologically effective rate study, data were analyzed using the nonlinear regression procedure.

Variances were divided into random (environment, location, and year), replication within environment, and the treatment × environment interaction and fixed effects (herbicide treatment). A Z test was used to test the significance of environment, replication (environment), and environment × treatment interactions. An F test was used to test the significance of fixed effects. Sites were analyzed separately or grouped so that the interaction between environment and treatment was nonsignificant. Residual plots were used to confirm that variance analysis for random, independent, and homogeneous error assumptions were met. The UNIVARIATE procedure in SAS was used to generate the Shapiro–Wilk statistic for tested data normality. The dose-response equation was $Y = c + \frac{d-c}{1+e^{b \cdot \log(\text{dose}) - \log(t)}}$, where c is the response at the highest rate, d is the response at the lowest rate, b is the slope at which there is the greatest rate of change, and t is the I_{50} value at which there is a 50% reduction in control, density, or biomass.

Table 2. Glyphosate-resistant common ragweed control at 4 and 8 wk after POST application (WAP).

| Treatment | Density ^{ab} | Aboveground biomass ^a | Control | Control | Density | Aboveground biomass |
|-----------------------------------------------------------|--------------------------------|----------------------------------|---------------|--------------|------------------------------|----------------------------|
| | At POST (no. m ⁻²) | At POST (g m ⁻²) | 4 WAP (%) | 8 WAP (%) | 8 WAP (no. m ⁻²) | 8 WAP (g m ⁻²) |
| Nontreated control | 1056 (±215.7) b | 72 (±12.6) c | — | — | 534 (±152.7) c | 192 (±29.6) c |
| Weed-free control | — | — | 100 a | 100 a | — | — |
| Glyphosate (9) | 447 (±142.8) a | 32 (±5.8) b | 29 (±6.8) d | 26 (±4.6) f | 248 (±54.2) b | 167 (±33.2) c |
| 2,4-D ester (4) | 128 (±61.2) a | 7 (±2.2) ab | 73 (±4.4) c | 51 (±2.3) e | 95 (±30.8) ab | 86 (±34.8) b |
| Saflufenacil/dimethenamid-P (14/15) | 8 (±4.0) a | 0 a | 84 (±5.1) bc | 64 (±3.2) d | 58 (±19) ab | 87 (±33.2) b |
| Linuron (7) | 7 (±4) a | 0a | 88 (4.9) ab | 70 (±4.6) cd | 32 (±17.5) a | 38 (±19.3) ab |
| Metribuzin (5) | 0 a | 0 a | 92 (±10.5) ab | 78 (±4.2) bc | 34 (±9.3) a | 21 (±7.5) ab |
| Fomesafen POST | — | — | 74 (±3.7) c | 63 (±5.9) de | 107 (±27.4) ab | 61 (±20.0) ab |
| 2,4-D ester (4) followed by fomesafen | — | — | 91 (±3.0) ab | 86 (±3.0) b | 16 (±6.2) a | 14 (±6.2) a |
| Saflufenacil/dimethenamid-P (14/15) followed by fomesafen | — | — | 98 (±0.9) a | 93 (±1.1) a | 5 (±2.0) a | 3 (±1.4) a |
| Linuron (7) followed by fomesafen | — | — | 96 (±1.9) ab | 92 (±2.6) a | 8 (±2.8) a | 5 (±2.4) a |
| Metribuzin (5) followed by fomesafen | — | — | 95 (±2.0) ab | 93 (±2.2) a | 14 (±8.1) a | 9 (±5.1) a |

Note: Density and aboveground biomass of treatments compared with nontreated control with preplant followed by postemergence herbicide applications. Common ragweed density and aboveground biomass were collected prior to postemergence herbicide application and then at 8 wk after POST (WAP).

^aGlyphosate-resistant common ragweed density and aboveground biomass prior to POST herbicide application.

^bMeans within a parameter are not significantly different if followed by the same letter.

These curves were generated using SAS 9.4 using the glyphosate-only treatment as the metribuzin 0 rate, and were then used to compute the dose required for 90% and 95% control and (or) reduction in aboveground biomass at 4 and 8 WAA.

Results and Discussion

PP followed by POST study

Prior to fomesafen applied POST, 2,4-D, saflufenacil/dimethenamid-P, linuron, and metribuzin reduced GR common ragweed density 71%, 98%, 98%, and 100% and aboveground biomass 78%, 100%, 100%, and 100%, respectively, relative to glyphosate-only treatment (Table 2).

At 4 and 8 WAP, fomesafen (POST) controlled GR common ragweed 74% and 63%, respectively. At 4 WAP, 2,4-D and saflufenacil/dimethenamid-P applied PP controlled GR common ragweed 73% and 84%, respectively, and control was improved by 18% and 14%, respectively, when fomesafen POST was applied in a two-pass program (Table 2). At 4 WAP, linuron and metribuzin applied PP controlled GR common ragweed 88% and 92%, respectively, and there was no improvement in control when they were followed by fomesafen POST (Table 2). Linuron and metribuzin both provide residual weed control, and therefore, no benefit of the fomesafen (POST) was observed at 4 WAP. At 8 WAP, 2,4-D, saflufenacil/dimethenamid-P, linuron, and

metribuzin controlled GR common ragweed 51%–78%, which improved to 86%–93% with fomesafen POST in a two-pass program. This was consistent with the weed density and aboveground biomass at the end of the season. The PP application of 2,4-D, saflufenacil/dimethenamid-P, linuron, and metribuzin reduced GR common ragweed density and aboveground biomass by 62%–86% and 48%–87%, respectively, relative to the glyphosate treatment. However, the density and aboveground biomass decreased by 94%–98% and 92%–98% with a two-pass program of a PP herbicide followed by fomesafen POST relative to the glyphosate-only treatment.

These results are consistent with studies by Bassett and Crompton (1982) that recorded 90% of common ragweed emergence by mid-June. Depending on soybean seeding and herbicide application timing, as well as on the length of residual activity, it is reasonable to assume that additional common ragweed emergence will occur following the PP herbicide application, and therefore, these escapes will require additional control measures later in the season. In this study, the PP followed by POST herbicide programs had greater control of common ragweed at 8 WAA (Table 2). Control of GR common ragweed with a PP-only application provided 51%–78% control, whereas PP followed by POST programs provided 86%–93% control at 8 WAA (Table 2).

Table 3. Regression parameters for dose-response equation^a to predict response of glyphosate-resistant common ragweed to varying rates of metribuzin.

| Rating | C | D | B | T | ED90 | ED95 |
|---------------------|----------------|----------------|--------------|----------------|---------|---------|
| Control 4 WAA | 105.7 (±12.97) | 30.7 (±3.88) | 1.8 (±0.67) | 345.8 (±92.28) | 824.4 | 1019.35 |
| Control 8 WAA | 94.7 (±8.14) | 22.3 (±15.68) | 2.47 (±0.86) | 335.5 (±54.8) | 1015.12 | — |
| Density | 8.9 (±49.3) | 363.3 (±45.16) | -3.7 (±3.19) | 232.2 (±62.5) | — | 785.5 |
| Aboveground biomass | 0 | 68.5 (±10.72) | -1.5 (±0.74) | 307.3 (±137.4) | 1108.38 | 1751.93 |

^a $Y = c + \frac{d-c}{1+e^{b \cdot \log(\text{dose}) - \log(t)}}$, where c is the response at the highest rate, d is the response at the lowest rate, b is the slope at which there is the greatest rate of change, and t is the I_{50} value at which there is a 50% reduction in control, density, or biomass.

The impact of these late escapes on soybean yield will depend on the time of emergence, density, and growing conditions for the remainder of the growing season. The second impact that must be considered is weed seed return to the soil, which will impact weed management in future years, especially in cases of herbicide-resistant weed biotypes. However, these late-emerging GR common ragweed plants may contribute little to the seed bank due to their late emergence. This is further influenced by weed seed predation and degradation in the soil (Swanton et al. 2008).

In this study, single herbicide applications did not provide complete control of GR common ragweed. Where a single PP herbicide application is used, more GR common ragweed are likely to reach reproductive maturity and provide a source of weed seed to increase the seed bank in future years, despite having less of an effect on soybean yield. However, where a POST-only herbicide is used, GR common ragweed is present during the critical weed-free period and may have a substantial impact on soybean yield. A two-pass program addresses these issues.

Although herbicide resistance continues to increase, no new herbicide modes of action have been commercialized since the 1990s in corn/soybean. Studies in Nebraska have examined the influence of spring tillage on common ragweed emergence as an alternative strategy to herbicides for control of GR common ragweed. Results from 2014 showed that spring tillage had no effect on total seedling emergence or time to 50% emergence (Barnes et al. 2014). The history of herbicide resistance has shown that reliance on any single herbicide provides selection pressure for the evolution of resistant biotypes within a population. The evolution of GR weeds has forced weed management practitioners to pay attention to the mode of action of the herbicide they are using. Because growers prefer to use less tillage for soil conservation and prefer the ease of herbicide application given their large farm size, promoting the use of multiple effective modes of action on each weed species forms an essential part of an integrated weed management program to delay or minimize the selection of herbicide-resistant biotypes (Owen et al. 2015). Growers should be encouraged to adopt holistic approaches to

weed management that combine cultural, mechanical, biological, and chemical weed control.

Biological effective rate of metribuzin study

At 4 WAA, the rate of metribuzin required for 90% and 95% GR common ragweed control was 824 and 1019 g a.i. ha⁻¹, respectively (Table 3). However, at 8 WAA, the rate of metribuzin required for 90% GR common ragweed control increased to 1015 g a.i. ha⁻¹. Therefore, a higher rate of metribuzin is required for longer residual weed control. At 8 WAA, the rate of metribuzin required for a 95% reduction in GR common ragweed density was 786 g a.i. ha⁻¹ and the rate of metribuzin required for 90% and 95% reduction in GR common ragweed aboveground biomass was 1108 and 1751 g a.i. ha⁻¹, respectively (Table 3). However, as shown in the PP followed by POST study, following an application of metribuzin followed by fomesafen results in more consistent weed control. Therefore, although a two-pass herbicide program is more expensive in terms of herbicides, fuel, and labour, a single application of metribuzin at a higher rate would require additional herbicide costs and create the possibility of crop injury, especially on light textured, high pH soils. Future studies should focus on whether the rate of metribuzin could be further reduced from the 824 g a.i. ha⁻¹ found in this study when used in a two-pass herbicide program.

Metribuzin (1120 g a.i. ha⁻¹) PP controlled GR common ragweed 28%–98% (Van Wely et al. 2014). Although this rate has provided excellent control of GR common ragweed, it is cost-prohibitive for grower use. In addition, previous studies have documented concerns regarding soybean injury, such as one by Askew et al. (1999), who reported tank-mixes with metribuzin resulting in 5%–6% soybean injury, however, this did not result in a yield loss. In this study, no herbicide injury was reported in 2014 at Windsor or in 2015 in Adams with metribuzin applied at 1120 g a.i. ha⁻¹, but in 2015, up to 5% soybean injury was observed at 4 WAA. This may be due to excess water at this location throughout the growing season, making the herbicide more available to the soybean plants. There was no soybean injury observed at the lower rates of metribuzin.

Control with metribuzin can be variable depending on the soil moisture, texture, and pH. Previous studies have shown that tank-mixes of imazethapyr + metribuzin, S-metolachlor + metribuzin, and S-metolachlor + metribuzin + linuron controlled common ragweed 88%–100% in two environments; however, control was reduced to 7%–55% in a third environment that had lower precipitation than the 20-yr average (Stewart et al. 2010). In contrast, common ragweed control with metribuzin can be reduced by excessive rainfall within 7 d of application on sandy soil (Stewart et al. 2010). In summary, season-long common ragweed control with metribuzin was variable.

Conclusion

In conclusion, the results of this study are consistent with previous studies that support the use of multiple effective herbicide modes of action for the control of herbicide-resistant weeds. Recommendations for reducing the selection of herbicide-resistant weed biotypes are to use multiple effective herbicide modes of action on every acre, every year, whether by tank-mixing herbicides or by sequential herbicide applications. This study found that sequential applications of saflufenacil/dimethenamid-P, linuron, or metribuzin PP followed by fomesafen POST provided more consistent control of GR common ragweed than a single application of any of these herbicides in soybean. Future studies should evaluate herbicide tank-mixes to determine whether acceptable control can be achieved compared with each of these herbicides on its own to avoid an over-reliance on a single herbicide. The metribuzin biologically effective rate study concludes that a minimum of 786 g a.i. ha⁻¹ is required for 95% control of GR common ragweed in soybean. However, if tank-mixing and sequential applications are incorporated into a management program for GR common ragweed, there may be opportunities to further reduce metribuzin rates without compromising season-long weed control in soybean.

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