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An integrated approach to control glyphosate-resistant *Ambrosia trifida* with tillage and herbicides in glyphosate-resistant maize

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Summary

Glyphosate-resistant *Ambrosia trifida* is a competitive and difficult-to-control annual broad-leaved weed in several agronomic crops in the Midwestern United States and Ontario, Canada. The objectives of this study were to compare treatments for control of glyphosate-resistant *A. trifida* with tillage followed by pre-emergence (PRE) and/or post-emergence (POST) herbicides in glyphosate-resistant maize and to determine the impact of *A. trifida* escapes on maize yield. Field experiments were conducted in 2013 and 2014 in grower fields infested with glyphosate-resistant *A. trifida*. Tillage prior to maize sowing resulted in 80–85% control compared with no tillage. Tillage followed by PRE application of saflufenacil plus dimethenamid-*P* with or without atrazine resulted in 99% control compared with ≤86 and 96% control with PRE herbicides alone at 7 and 21 days after application respectively. Tillage or POST-only herbicides resulted in 4–14 *A. trifida* plants m^{-2}, whereas a PRE and POST programme had <3 plants m^{-2}. Maize yield was greatest (13.1–14.2 tonnes ha^{-1}) with tillage followed by PRE and POST herbicide programme. The relationship between maize yield and late-season density of *A. trifida* escapes showed a 50% maize yield reduction irrespective of control measures when *A. trifida* density was 8.4 plants m^{-2}. It was concluded that the combination of tillage with PRE and/or POST herbicides reduced *A. trifida* density and biomass accumulation early in the season and provided an integrated approach for effective management.

Keywords: giant ragweed, competition, maize yield, late-season escapes, weed resistance management.

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Introduction

Ambrosia trifida L. (giant ragweed) is a natural coloniser in disturbed areas, a troublesome weed in arable lands and a threat to human health because of its allergenic pollen, a major cause of hay fever (Abul-fatih & Bazzaz, 1979; Baysinger & Sims, 1991). *Ambrosia trifida* dominates in common cropland plant communities, due to its early emergence, rapid growth rate, high leaf area index and ability to tolerate changing environmental factors by adjusting its resource utilisation response (Abul-fatih & Bazzaz, 1979; Bazzaz &

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Carlson, 1979). These characteristics of *A. trifida* result in crop shading, along with rapid consumption of water and nutrients, causing intense competition beginning from crop emergence, leading to significant yield losses (Abul-fatih & Bazzaz, 1979).

The commercialisation of glyphosate-resistant crops revolutionised weed management by providing excellent weed control and crop safety at a reduced cost. However, over-reliance and continuous use of glyphosate, along with declining trends in the use of other weed management practices, resulted in the evolution of glyphosate-resistant weeds, including *A. trifida* (Young, 2006; Givens *et al.*, 2009). Glyphosate-resistant *A. trifida* was first reported in Ohio in 2004 (Stachler, 2008) and as of 2016 has been confirmed in 12 US states (Heap, 2016) and in Ontario, Canada (Vink *et al.*, 2012). In addition, *A. trifida* populations resistant to both acetolactate synthase (ALS) inhibitors and glyphosate have been confirmed in Ohio, Minnesota and Missouri (Heap, 2016).

The evolution of herbicide-resistant weeds is primarily due to lack of diversity in weed management strategies (Norsworthy *et al.*, 2012). Therefore, one of the fundamental considerations for the management of glyphosate-resistant *A. trifida* and other herbicide-resistant weeds is the diversification of weed management strategies (Norsworthy *et al.*, 2012; Vencill *et al.*, 2012) using an integrated weed management (IWM) approach. Integrated weed management strategies should consider the use of cultural, mechanical and chemical control options that are both feasible in specific cropping systems and permitted by socio-economic conditions, to reduce selection pressure, delay the evolution of new resistant weeds and ensure effective management of existing herbicide-resistant weeds (Norsworthy *et al.*, 2012; Vencill *et al.*, 2012; Ganie *et al.*, 2014).

Integrated weed management practices are selected based on the biological and ecological characteristics of the weeds present (Harker & O'Donovan, 2013). However, current IWM systems mostly involve chemical plus physical and/or cultural methods, including tillage, cover crops and crop rotation (Harker & O'Donovan, 2013). Tillage is an important tool for managing herbicide-resistant weeds in agronomic crops (Jhala *et al.*, 2014b), and there is a need for more judicious, well-timed and precise use of tillage combined with other control methods (Shaner & Beckie, 2014). The success of tillage, like other weed control methods, is determined by several biological, physical and environmental factors (Vencill *et al.*, 2012). For example, early emerging weeds such as *A. trifida* are easy to control with tillage (Ganie *et al.*, 2016) compared with species that emerge simultaneously with crops and/or have a wide emergence period throughout the season (Hartzler *et al.*, 1999).

Ambrosia trifida competition has been assessed in several agronomic crops, including maize [*Zea mays* (L.)] (Harrison *et al.*, 2001), soyabean [*Glycine max* (L.) Merr.] (Baysinger & Sims, 1991) and cotton (*Gossypium hirsutum* L.) (Barnett & Steckel, 2013). Harrison *et al.* (2001) reported that *A. trifida* emerging simultaneously with maize resulted in 13 and 60% yield reduction at densities of 1.7 and 13.8 plants 10 m^{-2} respectively. *Ambrosia trifida* is even more competitive in soyabean, with 1 plant m^{-2} causing 45–77% yield loss (Baysinger & Sims, 1991; Webster *et al.*, 1994). However, there is no scientific literature available describing the impact of *A. trifida* that escapes weed control on crop yield, when there are increasing concerns about late-season *A. trifida* escapes in the eastern USA Corn Belt (Williams & Masiunas, 2006). A recent survey in Ohio and Wisconsin also reported *A. trifida* among the most common late-season escape weed species in glyphosate-dependent maize–soyabean cropping systems (Recker *et al.*, 2015). Most common causes of weed escapes have been reported by Bagavathiannan and Norsworthy (2012); however, the main reason for variable control of *A. trifida* with POST herbicides is emergence from different soil depths, resulting in variable plant sizes and leaf area. Small plants are sheltered by larger *A. trifida* plants, resulting in either zero or partial spray coverage of the POST herbicide, usually resulting in variable control (pers. obs. AJ Jhala). In addition, early-season management influences the size of *A. trifida* plants at the time of POST herbicide treatments. Loux *et al.* (2015) reported that at the time of POST herbicide application, 63% of *A. trifida* plants were >15 cm tall and 31% were >30 cm in untreated plots, whereas 95 and 99% of plants were <15 cm tall with a preplanting treatment (glyphosate plus 2,4-D ester) and preplant plus PRE herbicide programmes respectively.

A recent study in Nebraska confirmed that early-spring tillage had no effect on the emergence pattern of *A. trifida* (Kaur *et al.*, 2016). The study reported here was initiated based on the hypothesis that tillage would provide effective early-season control of *A. trifida* to allow maize planting in a weed-free environment and improve the efficacy of PRE and POST herbicides. It was further hypothesised that *A. trifida* escapes under the management programmes evaluated in this study would have a direct impact on maize yield. The objectives of this study were (i) to evaluate the efficacy of integrated management of glyphosate-resistant *A. trifida* with or without tillage followed by PRE and/or POST herbicides and (ii) to determine the relationship between the density of *A. trifida* escapes under the evaluated management programmes and maize yield.

Materials and methods

Field experiments were conducted at Clay Center (40.52°N, 98.05°W) and David City (41.25°N, 97.13°W), Nebraska, in 2013 and 2014, respectively, in grower fields infested with glyphosate-resistant *A. trifida*. *Ambrosia trifida* populations from these sites were confirmed resistant to glyphosate in 2011, with the level of resistance [determined by the ratio of effective dose required for 90% (ED_{90}) control of glyphosate-resistant populations with the labelled rate of glyphosate (1050 g a.e. ha^{-1}) required for 90% control of the susceptible populations] ranging from 9× to 14× compared with susceptible populations (Rana *et al.*, 2013). The density of glyphosate-resistant *A. trifida* at these sites varied from 18 to 32 plants m^{-2}. The experimental area within the field was selected considering the uniform distribution of *A. trifida* plants and the experimental design was robust enough to contain the spatial variation. The soil type at Clay Center was silt loam with 17% sand, 58% silt, 25% clay, 2.5% organic matter and a pH of 6.5. The soil type at David City was silty clay loam with 18% sand, 50% silt, 32% clay, 2.1% organic matter and a pH of 5.4. The experiment was arranged in a split-plot design with four replications, where the main plot was tillage or no tillage and the subplot was PRE and/or POST herbicide treatments for a total of 16 treatment combinations (Table 1). Treatment with no tillage or herbicide application served as the untreated control and tillage alone as a no herbicide control. Application rates of herbicides were based on their labelled rates in maize. Glyphosate-resistant maize seeds (Cv. 'Pioneer 1151R' in 2013 and 'Mycogen 2V709' in 2014) were planted on 16 May 2013 and 17 May 2014. The seeds were planted 3 cm deep at a density of 79 000 seeds ha^{-1}. Individual plots were 3 m wide and 9 m long with four maize rows spaced 76 cm apart.

Tillage was accomplished using a tandem disc on 2 May 2013 and 3 May 2014. Herbicide treatments were applied as PRE (16 May 2013 and 17 May 2014) and POST (8 June 2013 and 9 June 2014) on 6–15 cm tall (two- to six-leaf stage) *A. trifida* plants. Herbicides were applied with a CO_2-pressurised backpack sprayer equipped with a four-nozzle boom fitted with AIXR 110015 flat-fan nozzles (TeeJet, Spraying Systems Co., Wheaton, IL, USA), calibrated to deliver 140 L ha^{-1} at 276 kPa. The experimental locations were under rain-fed/non-irrigated conditions during both years.

Control of *A. trifida* by different treatments was visually assessed based on comparison with the untreated control plots with respect to symptoms such as chlorosis, necrosis, stand loss and stunting using a scale of 0–100% (0 being no control and 100 being complete control) at 7 and 14 days after preplant treatments (tillage/no tillage); 7, 14 and 21 days after PRE herbicide treatments (DAPRE); 30 and 60 days after POST herbicide treatments (DAPOST) and at harvest. Herbicide injury on maize was recorded using a scale of 0–100% (0 being no injury and 100 being plant death) at 14 and 21 days after PRE and POST herbicide treatments. *Ambrosia trifida* density was recorded from three randomly placed 0.25 m^2 quadrats per plot at 21 days after PRE herbicide treatments, 60 days after POST herbicide treatments and 2 weeks before maize harvest. The density of *A. trifida* recorded at 60 days after POST herbicide treatments was used to derive the crop yield loss function with escaped *A. trifida* plants. Glyphosate-resistant *A. trifida* biomass was assessed from three randomly placed 0.25 m^2 quadrats per plot at 60 days after POST herbicide application. *Ambrosia trifida* plants that survived herbicide treatment were cut at the stem base close to the soil surface, placed in paper bags, dried in an oven for 70 h at 55°C and weighed (g). Two centre rows of maize were harvested using a plot combine and yields were adjusted to 15% moisture content (Harrison *et al.*, 2001). *Ambrosia trifida* biomass data were converted into per cent biomass reduction compared to the untreated control (Ganie *et al.*, 2016) as:

$$\text{per cent biomass reduction} = \left[\frac{c - B}{c}\right] \times 100, \quad (1)$$

where c is the biomass of untreated control plots and B is the biomass of an individual treated experimental unit.

Statistical analysis

Data were subjected to ANOVA using the PROC GLIMMIX procedure in SAS version 9.3 (SAS Institute Inc, Cary, NC, USA). As subplot treatments (PRE/POST herbicides) were not applied until 14 days after tillage/no tillage, least square means for the visual control estimates of *A. trifida* at 7 and 14 days after tillage/no tillage were analysed as a randomised complete block design with preplant control (tillage or no tillage), year and their interactions considered as fixed effects, and replication as a random effect in the model. All other data were analysed as a split-plot design with preplant control, PRE and/or POST herbicides and their interactions considered as fixed effects and the year and replications as a random effect in the model. The

Table 1 Herbicide treatments, application timing, rates and products used in a field study for control of glyphosate-resistant *Ambrosia trifida* in glyphosate-resistant maize in Nebraska in 2013 and 2014 (acid equivalent = a.e., active ingredient = a.i., followed by = *fb*)

Herbicide common name	Timing	Rate g a.e. or a.i. ha^{-1}	Trade name	Manufacturer	Adjuvant*
2,4-D amine	POST	534	2,4-D amine	Winfield Solutions, LLC, St Paul, MN 55164; www.winfield.com	AMS + NIS
Saflufenacil + dimethenamid-*P fb*	PRE	780	Verdict	BASF Corporation, 26 Davis, Research Triangle Park, NC; www.basf.com	AMS + MSO
Glyphosate	POST	1260	Roundup PowerMax	Monsanto Company, 800 North, Lindberg Ave., St. Louis, MO; www.monsanto.com	AMS
Atrazine + saflufenacil + dimethenamid-*P fb*	PRE	2470 + 780	Aatre + Verdict	Syngenta Crop Protection, Inc, Greensboro, NC 27419 + BASF	AMS + MSO
Glyphosate	POST	1260	Roundup PowerMax	Corporation; Monsanto Company	AMS
Saflufenacil + dimethenamid-*P fb*	PRE	780	Verdict	BASF Corporation	AMS + MSO
2,4-D amine + glyphosate	POST	534 + 1260	2,4-D amine + Roundup PowerMax	Winfield Solutions + Monsanto Company	AMS
Halosulfuron + dicamba + glyphosate	POST	380 + 1260	Yukon + Roundup PowerMax	Gowan Company, Yuma, AZ 85366; www.gowanco.com + Monsanto Company	AMS + NIS
Saflufenacil + dimethenamid-*P fb*	PRE	780	Verdict	BASF Corporation	AMS + MSO
Halosulfuron + dicamba + glyphosate	POST	380 + 1260	Yukon + Roundup PowerMax	Gowan Company + Monsanto Company	AMS + NIS
Saflufenacil + dimethenamid-*P fb*	PRE	780	Verdict	BASF Corporation	AMS + MSO
Tembotrione + atrazine	POST	92 + 560	Laudis + Aatrex	Bayer Crop Science, Research Triangle Park, NC 27709; www.cropscience.bayer.com + Syngenta Crop Protection	

*AMS (ammonium sulphate, DSM Chemicals North America Inc., Augusta, GA) at 2% (wt/v), COC (crop oil concentrate, Agridex, Helena Chemical Co., Collierville, TN) or MSO (methylated seed oil, Southern Ag Inc., Suwanee, GA) at 1% (v/v) and NIS (non-ionic surfactant, Induce, Helena Chemical Co., Collierville, TN) at 0.25% (v/v) were mixed with herbicides.

treatment combinations with zero response variables were not included in the data analyses. Before analyses, data were tested for normality of residuals using the PROC UNIVARIATE procedure in SAS. Visual estimates of *A. trifida* control, density and biomass data were arcsine square root transformed before analysis; however, back-transformed data are presented with mean separation based on transformed data. When the ANOVA indicated treatment effects were significant, means were separated at $P \leq 0.05$ using Tukey–Kramer's pairwise comparison test. Pre-planned single degree-of-freedom contrast statements were used to compare management programmes by testing specific hypotheses, including tillage followed by PRE vs. PRE, tillage followed by POST vs. POST and tillage followed by PRE + POST vs. PRE + POST.

A two-parameter hyperbolic regression model (Eqn 2) was fitted to determine the relationship between maize yield and density of *A. trifida* escapes under the management approaches evaluated in this study (Barnett & Steckel, 2013) using R software (R statistical software, R Foundation for Statistical Computing, Vienna, Austria).

$$y = \frac{ab}{b + x}, \quad (2)$$

where y is maize yield (tonnes ha^{-1}), a is the upper asymptote or estimate of maximum yield, b is the estimate of *A. trifida* density (plants m^{-2}) that causes 50% reduction in maize yield and x is *A. trifida* density (plants m^{-2}).

Results

Year-by-treatment interactions for *A. trifida* visual control, density, biomass reduction, maize injury and yield were not significant; therefore, data were combined over years. However, the interaction between main plot treatments (tillage/no tillage) and subplot treatments (PRE and/or POST herbicides) was significant ($P < 0.05$) for all variables; therefore, mean separation for the simple effects is presented.

Ambrosia trifida control

Tillage resulted in 80–85% control of glyphosate-resistant *A. trifida* at 7 and 14 days after tillage compared with no tillage (data not shown). However, *A. trifida* control following tillage without PRE or POST herbicides declined to 55 and 46% at 30 and 60 DAPOST respectively (Table 2). The results of contrast analysis indicated that tillage + PRE herbicides provided 99% *A. trifida* control compared with 85% control with PRE-only herbicides at seven DAPRE (Table 3). For example, tillage + PRE application of saflufenacil plus dimethenamid-*P* with or without atrazine resulted in 99% control of *A. trifida* at 21 DAPRE treatment (Table 2). However, without tillage, the same treatment resulted in 95–96% control (Table 2). Tillage + POST herbicides resulted in 95–97% control of *A. trifida* compared with 90–95% control with POST-only herbicide programmes at 30 and 60 days after application and at harvest (Table 3).

Contrast statements to test the hypothesis that *A. trifida* control would be greater in tillage + PRE + POST herbicide programmes compared with PRE + POST herbicide programmes were not significant ($P > 0.05$) (Table 3). Both management programmes including herbicide mixtures such as saflufenacil plus dimethenamid-*P* with or without atrazine as PRE and glyphosate or halosulfuron plus dicamba plus glyphosate or tembotrione plus atrazine applied POST resulted in 99% control of glyphosate-resistant *A. trifida* at 30, 60 DAPOST and at harvest, regardless of tillage (Tables 2 and 3).

Ambrosia trifida density and biomass

The greatest *A. trifida* density at harvest was observed in the untreated control (≥26 plants m^{-2}). The next highest density was observed in plots where tillage alone had been applied (≥12 plants m^{-2}) (Table 4). Contrast analysis between tillage + POST herbicide and POST-only herbicide programme was significant ($P < 0.001$) at 60 DAPOST; however, tillage + PRE + POST herbicide vs. PRE + POST herbicide was not significant ($P = 0.819$). PRE + POST herbicide programmes reduced *A. trifida* density to <2.0 plants m^{-2} irrespective of tillage. However, density varied from 2 to 3 plants m^{-2} with tillage + POST herbicides, and 2 to 5 plants m^{-2} with POST-only herbicide programme (Table 5).

Contrast analysis to test the hypothesis that reduction in *A. trifida* biomass would be greater with tillage + PRE + POST compared with PRE + POST herbicide programme was not significant ($P = 0.262$). PRE + POST herbicide programmes resulted in >98% biomass reduction in *A. trifida* at 60 DAPOST irrespective of tillage (Table 5). Contrasts between tillage + POST and a POST-only herbicide programme were significant ($P = 0.04$). Results indicated that tillage + POST herbicide programmes reduced *A. trifida* biomass by 92% compared with 85% for the POST-only herbicide programmes (Table 5). However, 90–94% reduction in *A. trifida* biomass was observed among POST-only treatments irrespective of tillage, except for a 77% biomass reduction with 2,4-D applied POST without tillage (Table 4). Biomass reduction with only tillage was 24%, indicating the failure of tillage alone to control *A. trifida* later in the season (Table 4).

Maize injury and yield

Maize injury was 2–4% at 14 day after PRE herbicide treatments; however, injuries were transient and not visible at 30 day after treatment (data not shown). Contrast analyses to test the hypothesis that tillage + POST and tillage + PRE + POST herbicide programmes would result in greater maize yield compared with POST-only and PRE + POST herbicide programmes, respectively, were significant ($P < 0.003$) (Table 5). Tillage + POST herbicides resulted in average maize yield of 12.63 tonnes ha^{-1} compared with 8.50 tonnes ha^{-1} with POST-only programme (Table 5). Similarly, tillage + PRE + POST herbicides resulted in average maize yield of 13.71 tonnes ha^{-1} compared with 12.40 tonnes ha^{-1} with PRE + POST herbicide programme, indicating the importance of tillage for control of *A. trifida* (Table 5). However, the comparison of individual treatments suggested a comparable yield with tillage + PRE + POST and tillage + POST or PRE + POST (Table 4). The POST-only application of 2,4-D or halosulfuron plus dicamba plus glyphosate resulted in maize yields ranging from 7.90–9.10 tonnes ha^{-1}, which was greater than the yield with only tillage (4.60 tonnes ha^{-1}) (Table 4).

Impact of *A. trifida* escapes on maize yield

Size and density of *A. trifida* plants varied at the time of POST herbicide application, depending on prior control measures. For example, tillage alone resulted in <50% reduction in *A. trifida* density compared with ≥90% reduction with tillage + PRE herbicides (Table 4). Regrowth of partially controlled *A. trifida* plants and new emergence resulted in a mixed stand of

Table 2 Effect of tillage and/or herbicides for control of glyphosate-resistant *Ambrosia trifida* at 7 and 21 DAPRE; 30 and 60 DAPOST; and at harvest in glyphosate-resistant maize in 2013 and 2014 at Clay Centre and David City, NE respectively (acid equivalent = a.e., days after PRE = DAPRE, days after POST = DAPOST, followed by = *fb*)*,†

Treatment	Application timing	Rate g a.e. or a.i. ha−1	*Ambrosia trifida* control after PRE and POST treatments ‡,§				
			7 DAPRE	21 DAPRE	30 DAPOST	60 DAPOST	At harvest
Tillage	Preplant	–	75 c	66 c	55 c	45 c	36 c
Tillage *fb*	Preplant	–	73 c	64 c	95 ab	97 ab	97 a
2,4-D amine	POST	534					
Tillage *fb*	Preplant	–	99 a	99 a	99 a	99 a	99 a
Saflufenacil + dimethenamid-*P fb*	PRE	780					
Glyphosate	POST	1260					
Tillage *fb*	Preplant	–	99 a	99 a	99 a	99 a	98 a
Atrazine + saflufenacil + dimethenamid-*P fb*	PRE	2470 + 780					
Glyphosate	POST	1260					
Tillage *fb*	Preplant	–	99 a	99 a	99 a	99 a	99 a
Saflufenacil + dimethenamid-*P fb*	PRE	780					
2,4-D amine + glyphosate	POST	534 + 1260					
Tillage *fb*	Preplant	–	73 c	66 c	96 ab	97 ab	96 ab
Halosulfuron + dicamba + glyphosate	POST	380 + 1260					
Tillage *fb*	Preplant	–	99 a	99 a	99 a	99 a	99 a
Saflufenacil + dimethenamid-*P fb*	PRE	780					
Halosulfuron + dicamba + glyphosate	POST	380 + 1260					
Tillage *fb*	Preplant	–	99 a	99 a	99 a	98 ab	99 a
Saflufenacil + dimethenamid-*P fb*	PRE	780					
Tembotrione + atrazine	POST	92 + 560					
2,4-D amine	POST	534	0	0	91 b	92 b	90 b
Saflufenacil + dimethenamid-*P fb*	PRE	780	86 b	96 b	99 a	99 a	99 a
Glyphosate	POST	1540					
Atrazine + saflufenacil + dimethenamid-*P fb*	PRE	2470 + 780	84 b	95 b	99 a	99 a	99 a
Glyphosate	POST	1260					
Saflufenacil + dimethenamid-*P fb*	PRE	780	85 b	96 b	99 a	99 a	99 a
2,4-D amine + glyphosate	POST	534 + 1260					
Halosulfuron + dicamba + glyphosate	POST	380 + 1260	0	0	95 ab	93 ab	90 b
Saflufenacil + dimethenamid-*P fb*	PRE	780	85 b	96 b	99 a	99 a	99 a
Halosulfuron + dicamba + glyphosate	POST	380 + 1260					
Saflufenacil + dimethenamid-*P fb*	PRE	780	85 b	95 b	99 a	99 a	99 a
Tembotrione + atrazine	POST	92 + 560					
P-value			0.04	<0.001	0.014	<0.001	<0.001

*The treatments were arranged in a split-plot design, but to reduce the size of the table, main plot (tillage/no tillage) and subplot (PRE/POST herbicides) treatments are presented in the same column and when PRE/POST herbicides were applied alone, no-preplant control was mentioned in the table. Additionally, untreated control treatment with zero response variables was not included in analysis or in this table.

†Year-by-treatment interaction was not significant; therefore, data for both years were combined.

‡Data were arcsine square root transformed before analysis; however, data presented are the means of actual values for comparison based on interpretation from the transformed data.

§Means within columns with no common letter(s) are significantly different according to the Tukey–Kramer's pairwise comparison test at $P \leq 0.05$.

Table 3 Contrast means for control of glyphosate-resistant *Ambrosia trifida* in maize in different management programmes in field experiments conducted in Nebraska in 2013 and 2014 (days after PRE = DAPRE, days after POST = DAPOST, followed by = *fb*)*

Treatment	*Ambrosia trifida* control†				
	7 DAPRE	21 DAPRE	30 DAPOST	60 DAPOST	At harvest
Tillage *fb* PRE	99	99	–	–	–
PRE	85	96	–	–	–
Tillage *fb* POST	–	–	96	97	97
POST	–	–	93	92	90
Tillage *fb* PRE fb POST	–	–	99	99	99
PRE *fb* POST	–	–	99	99	99
Tillage *fb* PRE vs. PRE only	$P < 0.001$	$P < 0.001$	–	–	–
Tillage *fb* POST vs. POST only	–	–	$P = 0.014$	$P = 0.001$	$P < 0.001$
Tillage *fb* PRE fb POST vs. PRE *fb* POST	–	–	$P = 0.521$	$P = 0.499$	$P = 0.328$

*Year-by-treatment interaction was not significant; therefore, data for both years were combined.
†*P*-values are based on single degree-of-freedom contrast analysis.

plants varying from 8 to 15 cm in height at the time of POST herbicide application. Therefore, maize yield and the density of *A. trifida* escapes under different management approaches were correlated and this relationship was explained by a two-parameter hyperbolic regression model. The estimated parameters of the model for yield vs. *A. trifida* density at 60 DAPOST were $y = \left[\frac{14.4 \times 8.44}{8.44 + x}\right]$ with a root-mean-square error (RMSE) of 3.3, where y represents yield (tonnes ha^{-1}) and x represents *A. trifida* density (plants m^{-2}). The model predicted that *A. trifida* density of 8.44 plants m^{-2} allowed to compete up to 60 DAPOST herbicide application has the potential to cause 50% reduction in maize yield (Fig. 1).

Discussion

Tillage before crop planting provided effective control of *A. trifida*; however, the control following tillage without PRE or POST herbicides declined due to new emergence and regrowth of partially controlled *A. trifida*. Similarly, Ganie *et al.* (2016) reported >90% early-season control of *A. trifida* with tillage in soyabean, although the maintenance of effective control was dependent on follow-up applications of PRE and/or POST herbicide treatments. PRE treatments with multiple site-of-action herbicides including saflufenacil plus dimethenamid-*P* with or without atrazine were effective for control of *A. trifida*. Previous studies also reported ≥87% control of glyphosate-resistant *A. trifida* with a tank-mixture of glyphosate, saflufenacil and dimethenamid-*P* (Belfry & Sikkema, 2015) and 63% control with saflufenacil plus dimethenamid-*P* (Soltani *et al.*, 2011) at 28 DAPRE. Recently, Ganie *et al.* (2016) reported >96% control of glyphosate-resistant *A. trifida* in soyabean with tillage or 2,4-D followed by sulfentrazon plus cloransulam applied PRE compared with ≤86% control with the same herbicide treatments without tillage or 2,4 D at 21 DAPRE. Results of this study suggested the importance of tillage to supplement the PRE herbicides for effective early-season management of *A. trifida* in maize.

Although herbicide mixtures based on different biochemical sites-of-action applied PRE + POST or POST-only programmes provided >90% control irrespective of tillage, tillage + PRE + POST or tillage + POST is more desirable because of its potential to allow maize planting under less *A. trifida* competition during maize emergence. The most competitive weed species causing greatest yield loss in any crop including maize are those that emerge with or before the crop (McDonald *et al.*, 2010; Swanton *et al.*, 2015). Tillage is favourable to diversify the management approach, reduce dependence on herbicides and mitigate herbicide selection pressure for resistance by exposing fewer plants to herbicide(s) (Gressel & Levy, 2006; Norsworthy *et al.*, 2012).

The results of *A. trifida* control estimates were reflected in *A. trifida* density and biomass (Tables 4 and 5). Tillage + PRE and/or POST treatments resulted in greater reduction in *A. trifida* density and biomass compared with POST-only treatments. Similarly, Riley and Bradley (2014) reported the greatest reduction in *A. trifida* density (<6 plants m^{-2}) with a preplanting application of glyphosate plus 2,4-D, dicamba, or saflufenacil + glyphosate alone or glyphosate plus fomesafen or cloransulam or chlorimuron in glyphosate-resistant soyabean. Likewise, previous studies reported 75–100% reduction in *A. trifida* biomass with preplanting or PRE + POST herbicide programmes (Jhala *et al.*, 2014a; Kaur *et al.*, 2014).

Different plant height at the time of POST treatments often results in variable control and the escape

Table 4 Effect of different management programmes on glyphosate-resistant *Ambrosia trifida* density, biomass reduction, maize injury and seed yield in field experiments conducted in 2013 and 2014 at Clay Centre and David City, NE, respectively (acid equivalent = a.e., days after PRE = DAPRE, days after POST = DAPOST, followed by = *fb*).*,†,‡

		Rate	*Ambrosia trifida*,§,¶				
			Density (No. m^{-2})			Biomass reduction (%)	Maize yield
Herbicide	Application timing	g a.e. or a.i. ha^{-1}	21 DAPRE	60 DAPOST	At harvest	60 DAPOST	(Tonnes ha^{-1})§
Untreated control		–	31 a	29 a	26 a	–	0
Tillage	Preplant	–	14 b	14 b	12 b	24 d	4.60 d
Tillage *fb*	Preplant	–	13 b	3 cd	2 c	90 b	12.80 ab
2,4-D amine	POST	534					
Tillage *fb*	Preplant	–	1 c	1 d	0 c	99 a	13.44 a
Saflufenacil + dimethenamid-*P fb*	PRE	780					
Glyphosate	POST	1260					
Tillage *fb*	Preplant	–	1 c	1 d	0 c	99 a	13.82 a
Atrazine + saflufenacil+	PRE	2470 + 780					
Dimethenamid-*P fb*		–					
Glyphosate	POST	1260					
Tillage *fb*	Preplant	–	1 c	1 d	0 c	99 a	14.03 a
Saflufenacil + dimethenamid-*P fb*	PRE	780					
2,4-D amine + glyphosate	POST	534 + 1260					
Tillage *fb*	Preplant	–	10 b	2 d	2 c	94 ab	12.50 ab
Halosulfuron + dicamba + glyphosate	POST	380 + 1260					
Tillage *fb*	Preplant	–	1 c	1 d	0 c	99 a	13.12 a
Saflufenacil + dimethenamid-*P fb*	PRE	780					
Halosulfuron + dicamba + glyphosate	POST	380 + 1260					
Tillage *fb*	Preplant	–	1 c	1 d	0 c	99 a	14.20 a
Saflufenacil + dimethenamid-*P fb*	PRE	780					
Tembotrione + atrazine	POST	92 + 560					
2,4-D amine	POST	534	27 a	6 c	2 c	77 c	7.90 c
Saflufenacil + dimethenamid-*P fb*	PRE	780	2 c	1 d	0 c	99 a	12.54 ab
Glyphosate	POST	1260					
Atrazine + saflufenacil + dimethenamid-*P fb*	PRE	2470 + 780	2 c	1 d	0 c	97 ab	13.00 ab
Glyphosate	POST	1260					
Saflufenacil + dimethenamid-*P fb*	PRE	780	2 c	1 d	0 c	99 a	12.00 ab
2,4-D amine + glyphosate	POST	534 + 1260					
Halosulfuron + dicamba + glyphosate	POST	380 + 1260	26 a	4 cd	2 c	92 b	9.10 bc
Saflufenacil + dimethenamid-*P fb*	PRE	780	2 c	1 d	0 c	99 a	12.20 ab
Halosulfuron + dicamba + glyphosate	POST	380 + 1260					
Saflufenacil + dimethenamid-*P fb*	PRE	780	3 c	1 d	0 c	99 a	12.25 ab
Tembotrione + atrazine	POST	92 + 560					
P-value			<0.001	0.001	<0.001	0.044	0.030

*Treatments with 0% maize injury and no maize yield (0 tonnes ha^{-1}) were not included in the analysis.

†Treatments were arranged in a split-plot design but to reduce the size of the table main (tillage/no tillage) and subplot (PRE/POST herbicides) treatments were presented in same column and when PRE/POST herbicides were applied alone, no-preplant control was mentioned in the table.

‡Year-by-treatment interaction was not significant; therefore, data for both years were combined.

§*Ambrosia trifida* density and biomass data presented were collected at 60 DAPOST, and the data were arcsine square root transformed before analysis; however, data presented are the means of actual values for comparison based on interpretation from the transformed data.

¶Means within columns with no common letter(s) are significantly different according to the Tukey–Kramer's pairwise comparison test at $P \leq 0.05$.

Table 5 Contrast means for density and biomass reduction in glyphosate-resistant *Ambrosia trifida* in maize and maize seed yield under different management programmes in field experiments conducted in Nebraska in 2013 and 2014 (days after PRE = DAPRE, days after POST = DAPOST, followed by = *fb*) *,†

	Ambrosia trifida				
	Density (No. m^{-2})			Biomass reduction (%)	Maize yield (Tonnes ha^{-1})
Treatment	21 DAPRE	60 DAPOST	At harvest		
Tillage *fb* PRE	1	–	–	–	–
PRE	2	–	–	–	–
Tillage *fb* POST	–	3	2	92	12.63
POST	–	5	2	85	8.50
Tillage *fb* PRE *fb* POST	–	1	0	99	13.71
PRE *fb* POST	–	1	0	99	12.40
Tillage *fb* PRE vs. PRE only	$P = 0.024$		–		
Tillage *fb* POST vs. POST only	–	$P = 0.001$	$P = 0.819$	$P = 0.040$	$P < 0.001$
Tillage *fb* PRE *fb* POST vs. PRE *fb* POST	–	$P = 0.819$	$P = 1.000$	$P = 0.262$	$P < 0.003$

*Year-by-treatment interaction was not significant; therefore, data for both years were combined.
†*P*-values are based on single degree-of-freedom contrast analysis.

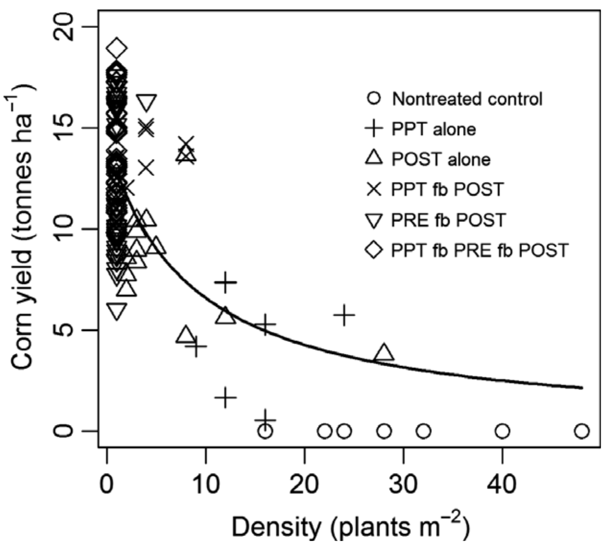

Fig. 1 Maize yield relative to density of glyphosate-resistant *Ambrosia trifida* (fb = followed, PPT = preplant tillage, POST = post-emergence, PRE = pre-emergence). The fitted line is calculated from the two-parameter hyperbolic model, $y = \frac{ab}{b+x}$, where y is maize yield (tonnes ha^{-1}), a is the upper asymptote or estimate of maximum yield, b is the estimate of *A. trifida* density (plants m^{-2}), which causes 50% reduction in maize yield, and x is the *A. trifida* density (plants m^{-2}). The estimated parameters were $y = \left[\frac{14.4 \times 8.44}{8.44 + x}\right]$ and root-mean-square error (RMSE) of 3.3.

of *A. trifida*. Loux *et al.* (2015) reported that without early-season control, 63% of *A. trifida* plants were >15 cm tall and 31% of plants were >30 cm tall at the time of POST herbicide application compared with tillage + PRE herbicide programmes, where 99% of plants were <15 cm tall. In glasshouse studies, Chahal *et al.* (2015) and Ganie *et al.* (2015) reported that *A. trifida* control declined and higher herbicide rates were needed for effective control at 20 cm height compared with 10 cm height following application of 2,4-D choline plus glyphosate and fluthiacet-methyl plus mesotrione respectively. Rapid growth rate, larger leaf size and the ability to grow taller than the crop, enable *A. trifida* to compete with crops even at lower densities and often require a second POST herbicide application for effective control and to prevent seed production (Loux *et al.*, 2015). Earlier studies have reported *A. trifida* as the most competitive weed in maize, soyabean and cotton (Baysinger & Sims, 1991; Harrison *et al.*, 2001; Barnett & Steckel, 2013). Harrison *et al.* (2001) reported 13.6% yield loss in maize with 1 *A. trifida* plant 10 m^{-2}. Additionally, they also reported a reduction in *A. trifida* interference with a 4-week delay in emergence compared with maize. However, the results of this study indicated that *A. trifida* escapes are competitive and cause yield losses, irrespective of control measures. Previous studies reported that the critical period of weed control in soyabean extended from 4 to 6 weeks after planting (Coble *et al.*, 1981; Bloomberg *et al.*, 1982) to 8–10 weeks in the presence of *A. trifida* (Baysinger & Sims, 1991).

Practical implications

Herbicide programmes exist for effective control of glyphosate-resistant *A. trifida* in maize, although it may not be true for the multiple herbicide-resistant *A. trifida* populations. Diversity in management approaches is needed for a true integrated weed management programme (Harker & O'Donovan, 2013). Results of this study suggested that tillage before sowing provided effective (>80%) early-season control of already emerged *A. trifida* and allowed maize to be planted under reduced *A. trifida* pressure (<20%).

In conclusion, this study indicated that tillage provides effective early-season control of *A. trifida* and supplements follow-up herbicides, but the use of PRE and/or POST herbicides or herbicide mixtures with different sites-of-action is indispensable, because *A. trifida* escapes can result in yield loss. A similar study in glyphosate-resistant soyabean reported that preplant tillage or 2,4-D + PRE and/or POST herbicides provided effective (>95%) *A. trifida* control compared to PRE + POST herbicide programme (Ganie *et al.*, 2016). Thus, tillage can be a potential tool for the integrated management of glyphosate-resistant *A. trifida* in maize–soyabean cropping systems. Future studies should consider integrating herbicides with additional non-chemical control strategies, including cover crops, harvest weed seed destruction and narrow-row planting to reduce selection pressure while providing an effective integrated weed resistance management strategy.

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