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Herbicide Interactions between Glufosinate and Three Fomesafen-Containing Herbicide Products as Affected by Weed Size and Spray Droplet Size

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Abstract

Two effective herbicides with different sites of action (SOA) are recommended for control of problematic weeds such as Palmer amaranth. When a LibertyLink® soybean variety is planted in the Midsouth, USA, glufosinate is often mixed with fomesafen to control Palmer amaranth and other common weed species. However, mixtures of glufosinate and fomesafen could be antagonistic, specifically when applied to grass species. A two-factor factorial experiment (herbicide treatment by weed size) was conducted at the Northeast Research and Extension Center in Keiser, Arkansas, to evaluate mixtures of glufosinate and three fomesafen-containing products for weed control and herbicide antagonism. Twelve herbicide treatments consisting of glufosinate and fomesafen herbicides alone, and in various combinations, were applied at two weed sizes (10 and 30 cm). Mixtures of glufosinate plus a fomesafen product, regardless of weed size, resulted in ≥ 96% control of PPO-inhibitor-susceptible Palmer amaranth. The addition of a fomesafen product to glufosinate had a negligible effect on control of barnyardgrass and tended to improve control of large crabgrass when compared to glufosinate alone (30 cm weed size). Most of the interactions between glufosinate and fomesafen were additive and it did not appear that one herbicide was negatively affecting the activity of the other. Generally, a premix of fomesafen+S-metolachlor provided better control than either Reflex or Flexstar herbicides alone and mixed with glufosinate. The improved control from the premix of fomesafen+S-metolachlor may partially be explained by droplet size; the premix produced smaller Dv50 (245 μm), compared to 289 and 303 μm, for Flexstar and Reflex herbicides, respectively. Smaller droplet sizes typically improve efficacy of contact herbicides such as glufosinate and fomesafen. To maximize efficacy of glufosinate plus fomesafen mixtures, use a premix of fomesafen+S-metolachlor, which also has the added benefit of multiple herbicide SOA for residual weed control.

Keywords: Antagonism; Barnyardgrass; Herbicide interactions; Palmer amaranth; Weed control

Introduction

Research into the confirmation and control of many glyphosate-resistant Palmer amaranth (*Amaranthus palmeri* S. Wats.) populations determined many protoporphyrinogen oxidase (PPO)-inhibiting herbicides, such as fomesafen, still provided excellent control [1,2]. As a result, adoption of PPO-inhibiting herbicides into weed management programs became a common recommendation in glyphosate-resistant soybean [3]. In glufosinate-resistant soybean, a PPO-inhibitor, such as fomesafen, can be mixed with glufosinate to provide multiple effective sites of action POST to reduce the likelihood of evolving herbicide resistance [4].

Prior to the widespread identification of PPO inhibitor-resistant [5-7], recommendations for Liberty Link soybean systems across the Midsouth included an early POST application of glufosinate plus a fomesafen-containing product, such as Flexstar or Prefix herbicide [8]. Glufosinate+fomesafen is effective on Palmer amaranth and other broadleaf weeds but may not achieve the same levels of control on grass species [8,9]. Culpepper et al. [9] showed the addition of fomesafen to glufosinate either increased, or did not change, control of many grass and broadleaf weeds [e.g., broadleaf signalgrass (*Urochloa platyphylla* Griseb.) and common lambsquarters (*Chenopodium album* L.)], however, the levels of control on the grass weeds were not always acceptable. Beyers et al. [10] reported foxtail biomass was greater when lactofen, another PPO-inhibiting herbicide, was added to glufosinate, indicating possible antagonism. However, no differences between visual control were detected and no herbicide interaction analysis was conducted.

Colby's method [11] is a common procedure used to investigate herbicide interactions and is well suited for evaluating mixtures of many products in the field. Glufosinate and fomesafen are both considered contact-type herbicides (vs. systemic or translocated herbicides); however, mixtures of two contact herbicides can still result in antagonism. Zhang et al. [12] compiled herbicide interaction results from 479 previously published cases and determined that an antagonistic interaction was just as likely for a combination of herbicides with similar transport mechanisms (i.e., both contact herbicides) as a mixture of a systemic and a contact herbicide.

Many fomesafen products are commercially available. Both Reflex® and Flexstar® herbicides contain the sodium salt formulations of fomesafen and are recommended for use PRE and POST in soybean. Prefix® herbicide also contains the sodium salt of fomesafen and is a premix of fomesafen and S-metolachlor. Reflex has a slightly higher concentration of the sodium salt of fomesafen than Flexstar (240 g L^{-1} *vs.* 226 g L^{-1}), includes a small amount of a preservative (1,2-benzisothiazolin-3-one), and is generally recommended as a PRE in soybean, whereas Flexstar is typically sprayed POST [8]. The formulations vary between Flexstar, Prefix, and Reflex herbicides, and a change in the adjuvant component of the formulation can impact herbicide efficacy [13]. Furthermore, Nalewaja and Matysiak [14] demonstrated identification of herbicide interactions can be dependent upon the formulation of the herbicides used.

When considering applications of contact herbicides, such as glufosinate and fomesafen, droplet size is an important consideration for maximizing efficacy. Glufosinate is known to perform better when applied with nozzles with a medium to coarse droplet designation compared to an ultra-coarse [15,16]. Applications of fomesafen and lactofen appear to be less sensitive to changes in droplet size compared to glufosinate, as the impact of nozzle selection seems appears to depend on species for the PPO-inhibiting herbicides [17,18]. The formulation itself can also influence the droplet size for different herbicide products containing the same active ingredient [19]. Thus, a change in droplet size as a result of mixing two herbicides could influence any potential antagonistic interactions.

Weed size influences efficacy of both glufosinate and PPO-inhibiting herbicides, with control declining when weeds are taller [20-22]. Antagonism can more easily be identified on large weeds compared to small weeds [23,24] typically because larger weeds are more likely to survive the application. The objective of this experiment was to determine the impact of weed size and fomesafen product on herbicide interactions between glufosinate and fomesafen. Of primary interest was the impact various formulations of fomesafen may have on the droplet size and subsequent identification of herbicide interactions with glufosinate.

Materials and Methods

Field experiments were conducted in 2015 and 2016 at the Northeast Research and Extension Center in Keiser, AR to evaluate the interaction between glufosinate and various fomesafen-containing products for control of small (10 cm) and large (30 cm) grass and broadleaf weeds. The experiment was a randomized complete block design with a factorial treatment structure: twelve herbicide treatments (factor 1) were applied at two application timings (factor 2). Application timings corresponded to weed height, approximately 10 and 30 cm weed sizes.

The twelve herbicide treatments consisted of glufosinate (Liberty herbicide, Bayer Crop Science, Research Triangle Park, NC) was applied alone at two rates (450 and 595 g ai ha^{-1}) and in combination with two formulations of fomesafen (Flexstar herbicide and Reflex herbicide, Syngenta Crop Protection LLC., Greensboro, NC), and one premix of fomesafen+S-metolachlor (Prefix Herbicide, Syngenta Crop Protection LLC., Greensboro, NC). Additionally, a nonionic surfactant (NIS) at 0.25% (v v^{-1}) (Induce, Helena Chemical Company, Collierville, TN) was added to all treatments that contained Flexstar or Reflex herbicides, as recommended by the product labels [25,26]. Any reference to Flexstar or Reflex herbicides alone or in mixture refers to a solution with NIS. Herbicide rates were selected based on those recommended on the product labels and were not rates that resulted in equal amounts of fomesafen being applied. A complete list of treatments, including the application timings, can be found in Tables 1 and 2.

Plots 3.9 by 9.1 m were established on a Sharkey clay (very fine, montmorillonitic, non-acid, thermic Vertic Haplaquept), with a pH 6.5 and 1.7% organic matter in both years. Each treatment was replicated four times in a given year. HBK 4950 Liberty Link soybean was planted at a rate of 313,000 seeds ha^{-1} on June 17, 2015 and June 10, 2016. Plots were furrow irrigated to soil saturation as needed throughout the growing season. Fertilizer and lime were applied based on a soil test and according to University of Arkansas recommendations.

Applications were made at 10:00 AM on July 16 and 5:00 PM on July 28 in 2015. In-field assessments at the time of application recorded a temperature of 32 C and 75% relative humidity for the first application and 36 C with 74% relative humidity for the second application in 2015. In 2016, applications were made at 7:30 AM on July 7 and 2:00 PM July 19. Temperatures were 29 and 25 C with a relative humidity of 75 and 49% for the first and second applications in 2016, respectively. Temperatures were 29 and 25 C with a relative humidity of 75 and 49% for the first and second applications in 2016, respectively.

Herbicide applications were made on the day the desired weed size was achieved and at a time that optimized herbicidal activity based on the product label (e.g., applications to occur between hours of dawn and two hours before sunset [27]) and minimized the opportunity for drift to adjacent plots (i.e., wind>8 km hr^{-1}). Weed heights at the time of herbicide application are listed by species in Table 1. For continuity, the two application timings will be referred to as 10 and 30 cm weeds. Soybean stages were V4-V5 at the first application and V7-V8 at the second application in both years.

Species	2015			2016		
	Height			Height		
	First[a]	Second	Density	First	Second	Density
	cm	Plants m^{-2}		cm	Plants m^{-2}	
Palmer amaranth	13	25	1	9	22	8
Prickly sida	5	18	6	3	12	1
Barnyard grass	10	35	9	10	26	20
Large crabgrass	10	31	2	9	22	3

[a]First and second application timing, to approximately 10 and 30 cm weeds, respectively.

Table 1: Weed sizes and densities of four weed species at both herbicide application timings evaluated in 2015 and 2016 in Keiser, AR.

A CO_2-pressurized backpack sprayer was used to make all herbicide applications calibrated to deliver 140 L ha^{-1} spray volume at 276 kPa at 4.8 km hr^{-1} through nozzles spaced 51 cm apart. The boom was equipped with Turbo TeeJet (TT) 110015 nozzles. (TeeJet Technologies, Springfield, Illinois). One day following the application of the herbicide treatments, all plots received an application of S-metolachlor (Dual Magnum®, Syngenta Crop Protection LLC., Greensboro, NC), except for those that already received an application as part of the experimental treatment.

Weed control ratings were collected 4 weeks after treatment (WAT) for barnyardgrass [Echinochloa crus-galli (L.) Beauv], Palmer amaranth, prickly sida (*Sida spinosa* L.), and large crabgrass (*Digitaria sanguinalis* L). The Palmer amaranth population was a glyphosate and acetolactate synthase-inhibitor-resistant population and was still sensitive to PPO-inhibiting herbicides, including fomesafen. Weed control was visually evaluated on a scale of 0 (no control) to 100% (complete death of all plants) relative to the nontreated check. Weed height and density data were collected 4 WAT. At the end of the season, plots were machine harvested and yield data collected. Soybean yields from each plot were corrected to 13% moisture.

A low-speed wind tunnel located at the University of Nebraska-Lincoln West Central Research and Extension Center in North Platte, NE was used to analyze the droplet spectra for herbicide treatments used in the field experiment. The wind tunnel was equipped with a Sympatec Helos Vario KR particle-size analyzer (Sympatec GmbH, Clausthal-Zellerfeld, Germany) which utilized a laser and R7 lens with a particle size detection range from 18 to 3,500 µm. The nozzle was attached 30 cm from the laser and width of the nozzle plume was moved across the laser via a linear actuator. The tunnel was set to produce a wind speed of 24 km h^{-1} to mitigate spatial sampling bias and each herbicide treatment was replicated three times. The volume median diameter (D_{v50}) was determined for each treatment as well as the D_{v10}, D_{v90}, relative span (RS), and the percentage of fine droplets. The D_{v50} is the droplet diameter below which 50% of the liquid volume is contained in droplets smaller than that value and the D_{v10} and D_{v90} are similar values for 10% and 90% of the volume, respectively. The percentage of fine droplets in this experiment was considered a fraction of the total volume of the spray containing droplets with a diameter <150 µm ($\%_{vol}$ fines). The range in droplet sizes is typically described with the relative span (RS) calculated using Equation 1.

$$RS=(D_{v90}-D_{v10})D_{v50}^{-1} \text{ ………… [1]}$$

Statistical analysis

For analysis and interpretation, the data were subject to an analysis of variance (ANOVA) using JMP 13 (SAS Institute Inc., Cary, NC). The statistical model included replication and year as random effects. The experimental design for the particle-size data did not include a blocking factor and a more-conservative Tukey adjustment (α=0.05) was used to identify differences between the means. The ANOVA was used to test for model effects and a treatment means were separated using a Fisher's protected least significant difference (LSD) test (α=0.05). In addition, herbicide interactions were tested for using Colby's method [11]. Both Colby's method the ANOVA were used to interpret the data. The results from the ANOVA were used to directly compare treatment means whereas Colby's method compares the mixture to the calculated Expected value for that mixture (Table 2).

Product Name	Common Name	Rate g ai ha^{-1}	Size cm	Control		Height reduction		Density reduction	
				%	SE	%	SE	%	SE
Liberty	Glufosinate	451	10	99	1	90	7	97	2
			30	89	3	86	14	96	4
Liberty	Glufosinate	595	10	99	1	92	8	93	6
			30	95	2	79	14	85	12
Flexstar	Fomesafen	264	10	98	3	74	17	86	12
			30	93	3	54	18	76	19
Reflex	Fomesafen	280	10	98	1	73	14	92	4
			30	93	3	63	14	93	3
Prefix	Fomesafen+S-metolachlor	266+1189	10	97	2	75	16	95	4
			30	95	2	60	16	90	4
Liberty+Flexstar	Glufosinate+fomesafen	451+264	10	99	1	90	8	98	2
			30	96	1	87	12	90	10
Liberty +Flexstar	Glufosinate+fomesafen	595+264	10	100	0	100	0	100	0
			30	100	0	100	0	100	0
Liberty+Reflex	Glufosinate+fomesafen	451+280	10	100	0	100	0	100	0
			30	96	2	87	13	96	4
Liberty+Reflex	Glufosinate+fomesafen	595+280	10	100	0	100	0	100	0
			30	98	2	87	11	90	10
Liberty+Prefix	Glufosinate+fomesafen +S-metolachlor	451+266+1189	10	100	0	100	0	100	0
			30	97	2	89	7	79	19

Liberty+Prefix	Glufosinate+fomesafen +S-metolachlor	595+266+1189	10	100	0	100	0	100	0
			30	100	0	100	0	100	0
Liberty+Dual Magnum	Glufosinate+S-metolachlor	451+1389	10	99	1	88	10	95	4
			30	93	2	93	8	98	2

[a]Data did not meet the assumptions of ANOVA and are reported as means followed by the standard error (SE) of the mean. [b]Height and density reduction is expressed as a percent of the nontreated control.

Table 2: The effect of weed size and mixtures of glufosinate plus fomesafen-containing products on Palmer amaranth control, height reduction and density reduction at 4 weeks after treatment in 2015 and 2016 in Keiser, AR[ab].

Colby's method uses an equation to calculate an Expected Value (E) as shown in Equation 2.

$$E=(X+Y)-(XY)/100 \ldots\ldots [2]$$

Where E is the expected level of control of two herbicides mixed together and variables X and Y represent the level of control provided by each herbicide applied individually. The observed value for the mixture was compared to the E calculated for that mixture using a two-sided t-test ($\alpha=0.05$). If the t-test was significant and E was greater than the observed value for a given mixture, it was deemed antagonistic. When E was less than the observed value, the mixture was considered synergistic and when no difference between E and observed was identified, additive. Some mixtures included three herbicides, one being S-metolachlor as part of a premix of fomesafen +S-metolachlor. As S-metolachlor is considered to have no POST activity, the calculation of E for the mixture proceeded as if the premix was a single product.

Results and Discussion

Palmer amaranth

All treatments of glufosinate and fomesafen-products alone or in combination provided >96% control of 10 cm Palmer amaranth and >88% control of 30 cm Palmer amaranth 4 weeks after treatment (WAT) (Table 2). Seven treatments, all of which were mixtures of glufosinate plus a fomesafen product, provided 100% Palmer amaranth control. Thus, the percent control, height reduction, and density reduction data did not meet the assumptions of ANOVA. Therefore, no ANOVA was conducted for these data. Instead, means for all treatments are presented in Table 2 and include a standard error for reference. The Palmer amaranth population evaluated in this study was sensitive to PPO-inhibiting herbicides and all three fomesafen products alone resulted in ≥ 93% control.

Prickly sida

In general, fomesafen was not effective at controlling prickly sida, whereas all treatments that contained glufosinate provided ≥ 87% control 4 WAT (Table 3). No fomesafen product alone applied to small (~10 cm) prickly sida provided more than 40% control, 42% height reduction, or 51% density reduction 4 WAT. Control with glufosinate at 451 g ai ha^{-1} was 87% when applied to large (~30 cm) prickly sida, and all mixtures of glufosinate plus a fomesafen product provided ≥ 90% control of prickly sida.

Antagonism was noted for three treatments applied to small prickly sida: glufosinate (451 g ai ha^{-1})+Flexstar herbicide, glufosinate (451 g ai ha^{-1})+Reflex herbicide, and glufosinate (595 g ai ha^{-1})+Reflex herbicide (Table 3).

Product Name	Common Name	Rate g ai ha^{-1}	Size cm	Control[c]				Height reduction				Density reduction			
				Obs %		Exp[d] %	p	Obs %		Exp %	p	Obs %		Exp %	p
Liberty	Glufosinate	451	10	97	--	--	--	75	--	--	--	95	--	--	--
			30	87	--	--	--	67	--	--	--	82	--	--	--
Liberty	Glufosinate	595	10	99	--	--	--	88	--	--	--	97	--	--	--
			30	90	--	--	--	85	--	--	--	83	--	--	--
Flexstar	Fomesafen	264	10	36	--	--	--	32	--	--	--	39	--	--	--
			30	30	--	--	--	34	--	--	--	24	--	--	--
Reflex	Fomesafen	280	10	28	--	--	--	17	--	--	--	46	--	--	--
			30	21	--	--	--	24	--	--	--	22	--	--	--
Prefix	Fomesafen+S-metolachlor	266+1189	10	40	--	--	--	35	--	--	--	51	--	--	--
			30	25	--	--	--	42	--	--	--	30	--	--	--

Liberty+Flexstar	Glufosinate+Fomesafen	451+264	10	92	NS	98	*	86	NS	88	NS	93	NS	98	NS
			30	92	NS	90	NS	73	NS	78	NS	83	NS	86	NS
Liberty+Flexstar	Glufosinate+fomesafen	595+264	10	98	NS	99	NS	95	NS	91	NS	97	NS	99	NS
			30	94	NS	93	NS	87	NS	90	NS	84	NS	86	NS
Liberty+Reflex	Glufosinate +fomesafen	451+280	10	93	NS	97	*	85	NS	79	NS	94	NS	97	NS
			30	95	NS	89	NS	74	NS	76	NS	87	NS	85	NS
Liberty+Reflex	Glufosinate+fomesafen	595+280	10	93	NS	99	*	95	NS	90	NS	88	NS	98	*
			30	94	NS	92	NS	86	NS	88	NS	84	NS	87	NS
Liberty+Prefix	Glufosinate+fomesafen+S-metolachlor	451+266+1189	10	98	NS	98	NS	94	^	84	NS	92	NS	97	NS
			30	90	NS	90	NS	71	NS	81	NS	87	NS	88	NS
Liberty+Prefix	Glufosinate+fomesafen+S-metolachlor	595+266+1189	10	99	NS	99	NS	93	NS	92	NS	97	NS	99	NS
			30	94	NS	92	NS	89	NS	91	NS	93	NS	88	NS
Liberty+Dual Magnum	Glufosinate+S-metolachlor	451+1389	10	95	NS	--	--	85	NS	--	--	96	NS	--	--
			30	90	NS	--	--	62	NS	--	--	75	NS	--	--
LSD				9				13				13			

[a]Abbreviation: Obs, observed value; Exp, Expected value; NS, Not Significant. [b]Height and density reduction is expressed as a percent of the nontreated control. [c]A "^" indicates a mixture that provided significantly greater control than both herbicides alone based on the LSD. NS indicates the mixture was similar to both of the herbicides alone. [d]A "*" denotes significant antagonism based on a two-sided t-test between observed and expected values. Expected values are based on Colby's equation [E=(X+Y)-(XY)/100]. Expected values can only be calculated when two herbicides in the mixture have POST activity on the species. [e]Rate is in g acid equivalent ha^{-1}.

Table 3: The effect of weed size and mixtures of glufosinate plus fomesafen-containing product on prickly sida control, height reduction, and density reduction at 4 weeks after treatment in 2015 and 2016 in Keiser, AR[ab].

Although the mixtures of glufosinate plus a premix of fomesafen+S-metolachlor did not provide greater control than other mixtures, all observed values for glufosinate+fomesafen+S-metolachlor were either greater than, or equal to, expected values (i.e., no antagonism was identified).

The premix of fomesafen+S-metolachlor tended to provide superior control compared to the other fomesafen products, whether alone or in mixture, indicating the premix may be the best product to utilize when applying fomesafen in mixture with glufosinate.

Barnyardgrass

Fomesafen products alone only provided suppression of 10 and 30 cm tall barnyardgrass at 4 WAT (28 to 46% control), while glufosinate alone controlled barnyardgrass 84 to 96% (Table 4).

Product name	Common name	Rate g ai ha^{-1}	Size cm	Control[c]				Height reduction				Density reduction			
				Obs %		Exp[d] %	p	Obs %		Exp %	p	Obs %		Exp %	p
Liberty	Glufosinate	451	10	95	--	--	--	68	--	--	--	95	--	--	--
			30	84	--	--	--	73	--	--	--	90	--	--	--
Liberty	Glufosinate	595	10	96	--	--	--	88	--	--	--	96	--	--	--
			30	88	--	--	--	73	--	--	--	91	--	--	--
Flexstar	Fomesafen	264	10	36	--	--	--	14	--	--	--	56	--	--	--
			30	42	--	--	--	17	--	--	--	44	--	--	--
Reflex	Fomesafen	280	10	28	--	--	--	17	--	--	--	56	--	--	--
			30	34	--	--	--	23	--	--	--	41	--	--	--

Prefix	Fomesafen+S-metolachlor	266+1189	10	41	--	--	--	18	--	--	--	53	--	--	--
			30	46	--	--	--	16	--	--	--	39	--	--	--
Liberty +Flexstar	Glufosinate +fomesafen	451+264	10	95	NS	97	NS	84	^	83	NS	95	NS	98	NS
			30	85	NS	91	NS	69	NS	77	*	87	NS	94	NS
Liberty +Flexstar	Glufosinate +fomesafen	595+264	10	98	NS	97	NS	94	NS	91	NS	97	NS	97	NS
			30	90	^	92	NS	69	NS	76	NS	87	NS	95	NS
Liberty +Reflex	Glufosinate +fomesafen	451+280	10	96	NS	97	NS	84	^	75	NS	97	NS	97	NS
			30	86	NS	89	NS	66	NS	78	NS	86	NS	95	NS
Liberty +Reflex	Glufosinate +fomesafen	595+280	10	98	NS	97	NS	95	NS	91	NS	99	NS	98	NS
			30	88	NS	92	NS	79	NS	79	NS	91	NS	95	NS
Liberty +Prefix	Glufosinate +fomesafen+S-metolachlor	451+266+1189	10	99	NS	97	NS	81	NS	75	NS	98	NS	98	NS
			30	89	NS	91	NS	63	NS	76	NS	89	NS	94	NS
Liberty +Prefix	Glufosinate +fomesafen+S-metolachlor	595+266+1189	10	99	NS	98	NS	91	NS	91	NS	98	NS	98	NS
			30	91	^	93	NS	77	NS	77	NS	86	NS	94	NS
Liberty+Dual Magnum	Glufosinate+S-metolachlor	451+1389	10	96	NS	--	--	86	^	--	--	97	NS	--	--
			30	88	NS	--	--	65	NS	35	--	86	NS	--	--
LSD				6				15				14			

[a]Abbreviation: Obs, Observed value; Exp, Expected value; NS, Not Significant, [b]Height and density reduction is expressed as a percent of the nontreated control, [c]A "^" indicates a mixture that provided significantly greater control than both herbicides alone based on the LSD. NS indicates the mixture was similar to both of the herbicides alone, [d]A "*" denotes significant antagonism based on a two-sided t-test between observed and expected values. Expected values are based on Colby's equation [E=(X+Y)-(XY)/100]. Expected values can only be calculated when two herbicides in the mixture have POST activity on the species, [e]Rate is in g acid equivalent ha^{-1}.

Table 4: The effect of weed size and mixtures of glufosinate plus fomesafen-containing product on barnyardgrass control, height reduction and density reduction at 4 weeks after treatment in 2015 and 2016 in Keiser, AR[ab].

No differences in control were observed between any mixtures of glufosinate plus a fomesafen product and the equivalent rate of glufosinate alone. Two treatments, glufosinate at 451 g ai ha^{-1}+Flexstar herbicide, and glufosinate at 451 g ai ha^{-1}+Reflex herbicide, showed significantly greater height reduction on 10 cm tall barnyardgrass compared to glufosinate alone, but was not reflected in the control ratings.

As such, fomesafen does not appear to antagonize glufosinate activity on barnyardgrass, regardless of weed size. Only one case of antagonism was identified across all parameters. Height reduction for glufosinate (451 g ai ha^{-1})+Flexstar on 30 cm barnyardgrass was antagonistic, but control and density reduction for the same treatment were considered additive based on Colby's method.

Large crabgrass

Stark reductions in large crabgrass control were observed when the same treatment was applied to 30 cm large crabgrass, compared to 10 cm (Table 5). For example, large crabgrass control with glufosinate at 595 g ai ha^{-1} was 95% 4 WAT, whereas the same treatment only provided 67% control when the application was made to 30 cm large crabgrass. Monks and Schultheis [28] also reported the ability to control large crabgrass with herbicides is diminished after it begins to form adventitious roots at the stem internodes. Large crabgrass evaluations generally responded positively when a fomesafen product was added to glufosinate. Control with glufosinate (595 g ai ha^{-1})+fomesafen+S-metolachlor was 82% compared to 70% with glufosinate alone.

Product name	Common name	Rate g ai ha^{-1}	Size cm	Control[c]				Height Reduction				Density reduction			
				Obs %		Exp[d] %	p	Obs %		Exp %	p	Obs %		Exp %	p
Liberty	Glufosinate	451	10	95	--	--	--	53	--	--	--	93	--	--	--
			30	67	--	--	--	52	--	--	--	51	--	--	--

Liberty	Glufosinate	595	10	98	--	--	--	51	--	--	--	95	--	--	--
			30	70	--	--	--	53	--	--	--	46	--	--	--
Flexstar	Fomesafen	264	10	41	--	--	--	15	--	--	--	32	--	--	--
			30	22	--	--	--	14	--	--	--	24	--	--	--
Reflex	Fomesafen	280	10	34	--	--	--	10	--	--	--	40	--	--	--
			30	13	--	--	--	14	--	--	--	28	--	--	--
Prefix	Fomesafen+S-metolachlor	266+1189	10	44	--	--	--	9	--	--	--	38	--	--	--
			30	20	--	--	--	18	--	--	--	16	--	--	--
Liberty +Flexstar	Glufosinate +fomesafen	451+264	10	95	NS	98	NS	85	^	54	*	98	NS	97	NS
			30	76	^	75	NS	59	NS	58	NS	54	NS	61	NS
Liberty +Flexstar	Glufosinate +fomesafen	595+264	10	97	NS	99	NS	79	^	58	*	98	NS	96	NS
			30	81	^	76	NS	65	NS	60	NS	50	NS	60	NS
Liberty+Reflex	Glufosinate +fomesafen	451+280	10	94	NS	96	NS	85	^	58	*	96	NS	95	NS
			30	77	^	73	*	52	NS	58	NS	51	NS	61	NS
Liberty+Reflex	Glufosinate +fomesafen	595+280	10	96	NS	99	NS	72	^	56	NS	97	NS	97	NS
			30	81	^	75	*	61	NS	59	NS	47	NS	61	NS
Liberty+Prefix	Glufosinate +fomesafen+S-metolachlor	451+266+1189	10	98	NS	97	NS	98	^	57	*	99	NS	96	NS
			30	78	^	74	NS	61	NS	60	NS	51	NS	55	NS
Liberty+Prefix	Glufosinate +fomesafen+S-metolachlor	595+266+1189	10	95	NS	99	NS	88	^	55	*	97	NS	97	NS
			30	82	^	76	*	59	NS	57	NS	59	NS	54	NS
Liberty+Dual Magnum	Glufosinate+S-metolachlor	451+1389	10	97	NS	--	--	58	NS	--	--	97	NS	--	--
			30	70	NS	--	--	42	NS	--	--	48	NS	--	--
	LSD			6				15				14			

[a]Abbreviation: Obs, Observed value; Exp, Expected value; NS, Not Significant, [b]Height and density reduction is expressed as a percent of the nontreated control, [c]A "^" indicates a mixture that provided significantly greater control than both herbicides alone based on the LSD. NS indicates the mixture was similar to both of the herbicides alone, [d]A "*" denotes significant antagonism based on a two-sided t-test between observed and expected values. Expected values are based on Colby's equation [E=(X+Y)-(XY)/100]. Expected values can only be calculated when two herbicides in the mixture have POST activity on the species, [e]Rate is in g acid equivalent ha^{-1}.

Table 5: The effect of weed size and mixtures of glufosinate plus fomesafen-containing product on large crabgrass control, height reduction, and density reduction at 4 weeks after treatment in 2015 and 2016 in Keiser, AR[ab].

When applied to 10 cm large crabgrass, a synergistic response was detected for height reduction for all mixtures of glufosinate plus a fomesafen product. However, neither percent control nor density reduction detected a synergistic response on 10 cm large crabgrass. These findings are likely due to the overall high levels of control (≥ 94%) and density reduction (96%) that were observed when applications were made to 10 cm weeds. Height assessments were only collected on plants that survived the application and it is possible the addition of fomesafen to glufosinate inhibited the survivors' ability to regrow new tissue compared to glufosinate alone.

Grain yield

Overall, soybean yield was greatest for treatments that provided superior control of all species. The interaction between herbicide and weed size and the main effect of weed size was not significant in the

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ANOVA model at α=0.050 (p=0.0730 and p=0.2973 for the interaction and weed size main effect, respectively).

Only the main effect of herbicide was interpreted for grain yield (Table 6). Overall, the presence of glufosinate was the most important factor for maximizing grain yield. The treatment of glufosinate alone (451 g ai ha^{-1}) produced 3286 kg ha^{-1} of soybean, averaged over weed sizes, and was not different from any of the mixtures with fomesafen products. The lowest yields were obtained from treatments composed of only a fomesafen-product. No differences in yield were identified between Flexstar, Prefix, and Reflex herbicides, averaged over weed size. The lowest yields from the fomesafen only treatments were likely due to the intense grass pressure (Table 1) and lack control of barnyardgrass, large crabgrass, and prickly sida with those treatments.

Product name	Herbicide treatment	Rate g ai ha^{-1}	Yield[a] kg ha^{-1}
Liberty	Glufosinate	451	3286^{ab}
Liberty	Glufosinate	595	3253^{ab}
Flexstar	Fomesafen	264	2814^{c}
Reflex	Fomesafen	280	2810^{c}
Prefix	Fomesafen+S-metolachlor	266+1189	2934^{c}
Liberty+Flexstar	Glufosinate+fomesafen	451+264	3174^{b}
Liberty+Flexstar	Glufosinate+fomesafen	595+264	3337^{a}
Liberty+Reflex	Glufosinate+fomesafen	451+280	3285^{ab}
Liberty+Reflex	Glufosinate+fomesafen	595+280	3287^{ab}
Liberty+Prefix	Glufosinate+fomesafen+S-metolachlor	451+266+1189	3239^{ab}
Liberty+Prefix	Glufosinate+fomesafen+S-metolachlor	595+266+1189	3324^{a}
Liberty+Dual Magnum	Glufosinate+S-metolachlor	451+1389	3233^{ab}

[a]Means followed by the same letter within a column are not statistically different according to Fisher's protected LSD (α=0.05).

Table 6: The effect of mixtures of glufosinate plus fomesafen-containing product on soybean grain yield, averaged over application timing in 2015 and 2016 in Keiser, AR.

One interesting difference was obtained with mixtures of Flexstar herbicide with the high and low rates of glufosinate; a significantly greater yield (3,340 kg ha-1) was obtained when glufosinate at 595 g ai ha^{-1} was mixed with Flexstar herbicide (264 g ai ha^{-1} fomesafen) compared to the mixture with the low rate (451 g ai ha^{-1}) of glufosinate (3,170 kg ha^{-1}). The improved yield from the higher rate of glufosinate in mixture highlights the importance of using full herbicide rates, even in mixtures (it should be noted that both 451 and 595 g ai ha^{-1} are labeled use rates).

Droplet size analysis

The droplet spectra analysis (Table 7) provides some insight to the differing performance between fomesafen products and herbicide treatments on weed control. Of the fomesafen products alone, the premix of fomesafen+S-metolachlor generally provided superior control to Reflex herbicide and was either equal to, or greater than, Flexstar herbicide. This generalization correlates with the droplet data, where the premix of fomesafen+S-metolachlor, Flexstar herbicide, and Reflex herbicide had D_{v50} values of 245, 289, and 303 µm, respectively.

Product Name	Herbicide Treatment	Rate g ai ha^{-1}	Droplet spectra parameters[a]				
			D_{v10}	D_{v50}	D_{v90}	Relative span[b]	<150 µm
			µm				%vol
	Water		143^{a}	307^{a}	488^{a}	1.12^{de}	11.1^{e}
Liberty	Glufosinate	451	136^{ab}	296^{bc}	478^{ab}	1.15^{cde}	12.4^{de}
Liberty	Glufosinate	595	126^{cd}	280de	470^{ab}	1.23^{abc}	14.9^{bc}
Flexstar	Fomesafen	264	141^{a}	289^{cd}	462^{b}	1.11^{de}	11.7^{de}
Reflex	Fomesafen	280	140^{ab}	303^{ab}	483^{ab}	1.13^{cde}	11.8^{de}
Prefix	Fomesafen+S-metolachlor	266+1189	122^{cd}	245^{g}	393^{c}	1.1d^{e}	17.1^{ab}
Liberty+Flexstar	Glufosinate+fomesafen	451+264	131^{bc}	283^{d}	470^{ab}	1.2^{bcd}	14^{cd}
Liberty+Flexstar	Glufosinate+fomesafen	595+264	126^{cd}	280de	485^{a}	1.28^{ab}	15.1^{bc}
Liberty+Reflex	Glufosinate+fomesafen	451+280	120^{d}	270^{ef}	472^{ab}	1.3^{a}	16.8^{ab}
Liberty+Reflex	Glufosinate+fomesafen	595+280	122^{d}	267^{f}	462^{b}	1.27^{ab}	16.6^{ab}
Liberty+Prefix	Glufosinate+fomesafen+S-metolachlor	451+266+1189	124^{cd}	241^{g}	382^{c}	1.07^{e}	16.8^{ab}
Liberty+Prefix	Glufosinate+fomesafen+S-metolachlor	595+266+1189	122^{cd}	241^{g}	383^{c}	1.08^{e}	17.2^{ab}

Liberty+Dual Magnum	Glufosinate+S-metolachlor	451+1389	121[d]	238[g]	374[c]	1.06[e]	17.5[a]

[a]Means followed by the same letter within a column are not statistically different according to Fisher's protected LSD with a Tukey adjustment (α=0.05), [b]Relative span is a unitless index of the uniformity of droplet size distribution. Smaller values represent more uniformity in droplet size distribution.

Table 7: Spray characteristics of various herbicide combinations for glufosinate, three fomesafen products and various mixtures used in the field experiment including D_{v10}, D_{v50}, D_{v90}, relative span, and % of the volume (%vol) containing droplets with diameters <150 µm.

Mixtures of glufosinate and fomesafen containing products produced similar or significantly smaller droplets in comparison to the individual components of the mixture. For example, glufosinate (451 g ai ha^{-1}) plus Flexstar herbicide had a volume median diameter (D_{v50}) of 283 µm which was equivalent to Flexstar herbicide alone (289 µm) and less than glufosinate alone (296 µm). A premix of fomesafen+S-metolachlor produced the smallest droplets (D_{v50}=245 µm) and was equal to mixtures with glufosinate that included the premix, as well as glufosinate+S-metolachlor.

In general, treatments that produced the smallest D_{v50} also produced the greatest number of fine droplets (%vol fines), although mixtures of glufosinate+Reflex herbicide had $\%_{vol}$ fines equal to that of the treatments that contained S-metolachlor (either in Prefix or Dual herbicides). It is possible for one treatment to produce a similar number of percent fines as another and have a smaller D_{v50}. The relative span (RS) of a treatment is a unitless index that represents the range or spread in droplet sizes for a given treatment and can explain discrepancies between D_{v50} and $\%_{vol}$ fines. For example, glufosinate (451 g ai ha^{-1})+Reflex herbicide produced a D_{v50}=270 µm, $\%_{vol}$ fines=16.8, and RS=1.30, compared to glufosinate (451 g ai ha^{-1})+fomesafen+S-metolachlor with a D_{v50}=241 µm, $\%_{vol}$ fines=16.8, and RS=1.07. In regard to efficacy, an ideal mixture of two contact herbicides would result in a smaller D_{v50}, larger $\%_{vol}$ fines, and a narrow relative span, indicating the mixture is producing smaller and more uniform droplets than either of its components.

Although the droplet data may explain some of the differences observed between the fomesafen products on weed control, it is not the only contributing factor. For example, Flexstar herbicide had a droplet spectra closer to that of Reflex than the fomesafen+S-metolachlor premix. Thus, differences between Reflex and Flexstar may also be associated with the adjuvant system in each product. When considering Prefix herbicide, S-metolachlor unlikely impacted efficacy, as all plots that did not already have an application of S-metolachlor had it applied 24 h later. In addition to the droplet size, the most likely explanation for the differences between fomesafen products is the adjuvant system associated with the product itself [13]. Although few differences were observed between fomesafen products when they were mixed with glufosinate, the premix of fomesafen+S-metolachlor appears to be the better mixture partner with glufosinate in regards to optimizing spray droplet parameters for efficacy, and has the added benefit of already including S-metolachlor. If spray drift is a concern, large droplets with a smaller $\%_{vol}$ fines is preferred, and Flexstar herbicide would be a better mix partner than a premix of fomesafen+S-metolachlor in such cases.

Conclusion and Practical Implications

Fomesafen does not appear to be interacting negatively with activity of glufosinate on grass species. In fact, the addition of a fomesafen product to glufosinate appeared to improve control of large crabgrass compared to glufosinate alone. Even so, the highest rates of both herbicides should be used to maximize control and reduce the likelihood of yield loss. No severe cases of antagonism or synergism were identified in this experiment, although the identification of an herbicide interaction does depend on weed size and parameter investigated. Overall, the results from these experiments agree with the findings of Culpepper et al. [9] that the addition of fomesafen does not antagonize the efficacy of glufosinate.

Even though the prevalence of PPO-inhibitor resistant (PPO-resistant) Palmer amaranth populations across the Midsouth may diminish applications of currently labeled PPO-inhibiting herbicides, chemical companies are bringing new PPO-inhibitors to market [29]. Furthermore, Umphres et al. [30] determined soil-applied PPO-inhibiting herbicides, including fomesafen, still have activity on a PPO-resistant Palmer amaranth population. Even if fomesafen lost some of its utility as a POST herbicide on PPO-inhibitor resistant populations, it may still provide value as a POST residual option, in addition to other species it may control POST.

Because of the presence of Palmer amaranth populations resistant to two or more sites of action (e.g., ALS, EPSPS, PPO-inhibitors; ALS and EPSPS; EPSPS and PPO-inhibitors) [7,31] fomesafen may still be of some value in soybean production. Fomesafen does not appear to severely antagonize the activity of glufosinate on grass or broadleaf weeds and can improve control when mixed with glufosinate, compared to glufosinate alone. Furthermore, the residual activity of fomesafen on PPO-resistant Palmer amaranth may reduce the selection pressure on very long chain fatty acid synthesis-inhibitors (e.g., S-metolachlor) which are frequently applied PRE and POST in soybean.

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