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# **Research Article**

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# Control of Palmer amaranth (*Amaranthus* palmeri) regrowth following failed applications of glufosinate and fomesafen

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### Abstract

Rapid vegetative growth and adverse application conditions are common factors leading to the failure of postemergence herbicides on Palmer amaranth. A sequential herbicide application, or respray, is often necessary to control weeds that have survived the initial herbicide application to protect crop yield and minimize weed seed production. The optimum timing after the initial application and the most effective herbicide for control of Palmer amaranth has not been characterized. The objectives of these experiments were to determine the optimum herbicide for treating Palmer amaranth regrowth, the optimum timing for each of those herbicides, and how the initial failed herbicide might affect efficacy of a second herbicide application. Bare ground field experiments were performed in 2017 and 2018 in which glufosinate or fomesafen herbicide failure was induced on Palmer amaranth plants that were 30 cm in height. Respray treatments of glufosinate, fomesafen, lactofen, 2,4-D, and dicamba were applied once at timings of 4 to 5 d, 7 d, or 11 d after the initial spray application. Nearly all herbicide treatment and timing combinations increased control by at least 13 percentage points compared to no respray herbicide treatment. Regardless of initial herbicide, glufosinate applied as a respray treatment was the most consistent and efficacious with up to 97% control. The specific herbicide used in the second application impacted final weed control more so than timing of the respray application. For instance, control by glufosinate respray treatments was 10 to 18 percentage points greater than control from lactofen respray treatments, whereas control decreased by 3 percentage points when respray applications of any herbicide were made 11 d after initial application of glufosinate compared to 4 to 5 and 7 d after initial application of glufosinate. In the event of failure to control Palmer amaranth with glufosinate or fomesafen, glufosinate should be applied in order to maximize control.

## Introduction

Palmer amaranth, native to the desert southwest United States, is a very troublesome weed in cotton and soybean growing regions in the United States, particularly the mid-south (Van Wychen 2019). Palmer amaranth seed can be transported long distances and quickly infest new areas (Li and Qiang 2009; Norsworthy et al. 2009). Once introduced to an area, Palmer amaranth demonstrates high levels of developmental plasticity such as completing a life cycle and producing seed even when emerging late in the year (Spaunhorst et al. 2018). Biotypes that are resistant to glyphosate and acetolactate synthase (ALS) inhibitors (categorized as a Group 2 herbicide by the Weed Science Society of America) are present in nearly every state with Palmer amaranth (Bagavathiannan and Norsworthy 2016; Heap 2020; Ward et al. 2013). Therefore, these herbicides cannot be reliably used for control of this weed. As a result, growers and land managers are increasingly relying on glufosinate, protoporphyrinogen oxidase (PPO) inhibitors (WSSA Group 14), dicamba, or 2,4-D (WSSA Group 4) in combination with herbicide-resistant crops.

Palmer amaranth exhibits rapid vegetative growth and can be extremely competitive with crops; 10 Palmer amaranth plants per meter of row can reduce soybean yield by 68% (Klingaman and Oliver 1994). In a short period of time, Palmer amaranth can outgrow the recommended weed height for POST herbicides, leading to potential herbicide failure. Larger weeds require a greater herbicide dose for complete control (Steckel et al. 1997a). Furthermore, Lillie et al. (2020) found that Palmer amaranth, sensitive to PPO-inhibiting herbicides, that was taller than 10 cm required more fomesafen to be controlled than waterhemp [(Amaranthus tuberculatus (Moq.) J. D. Sauer] at 10 cm in height that was resistant to PPO-inhibiting herbicides. In addition to weed size, environmental conditions and less than optimal spray application methods can also cause failure of a foliar herbicide application. Environmental conditions and sprayer configuration must be optimized in order to deliver the maximum dose

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to the target site, especially for a troublesome weed such as Palmer amaranth (Coetzer et al. 2001; Ritter and Coble 1981; Sikkema et al. 2008; Wichert et al. 1992).

Inevitably, some POST herbicide applications fail to completely kill Palmer amaranth plants, allowing compensatory growth from previously dormant axillary buds. Depending on the severity of plant injury, weeds that have regrown following the loss of growth from the shoot apical meristem can still produce as much biomass as plants that have not been sprayed (Mager et al. 2006b). Furthermore, weed seed production remains possible on plants surviving these herbicide applications. Thus, control of surviving Palmer amaranth is imperative so that crop competition is prevented, weed seed is not produced, and low-dose selection pressure for herbicide resistance does not cause a rapid shift to herbicide-resistant biotypes (Norsworthy et al. 2014).

In the event of herbicide failure, few research-based recommendations exist on the course of action to guide respray timing or herbicide active ingredient to use. Following a failed glufosinate application, glufosinate and fomesafen as respray treatments resulted in 90% to 100% control of waterhemp, regardless of application timing (Haarmann et al. 2020). After a failed application of fomesafen, applying glufosinate or 2,4-D resulted in 87% to 99% control of waterhemp. However, the application timing of respray herbicide applications relative to the initial failed application had little effect on waterhemp efficacy (Haarmann et al. 2020). Waterhemp plants clipped to simulate herbicide failure were more susceptible to lactofen, but there was no change in susceptibility to glyphosate (Mager et al. 2006a). The same study resulted in a different response in giant ragweed (Ambrosia trifida L.) and ivyleaf morningglory (Ipomoea hederacea Jacq.) demonstrating that response of herbicide-injured plants is different across species. Herbicide response can also change with application timing. Palmer amaranth control was greater when lactofen was applied 14 d after an initial application of lactofen rather than at 7 d (Sperry et al. 2017). Another study by Randell et al. (2020) found that Palmer amaranth control was greatest when glufosinate was applied sequentially at 1 to 10 d after the initial application of glufosinate compared to when it was applied 10 to 14 d after the initial application. The authors did not provide an explanation why timing was important, and there is also no commonly accepted explanation across weed species or herbicides. Presumably, delaying the respray application for too long results in a weed that is too large for the subsequent herbicide. An application that is performed too early may not have sufficient actively growing leaf tissue for herbicide absorption to occur, especially if the first herbicide was a nonsystemic herbicide resulting in rapid necrosis of leaf tissue. However, nonsystemic herbicides may result in near complete tissue necrosis, but meristems may still be active.

We hypothesize that respray applications to Palmer amaranth should be delayed to approximately 7 d after Palmer amaranth has regrown so as to increase herbicide interception on actively growing leaf tissue, but before a large amount of biomass accumulation and full coverage becomes difficult (Steckel et al 1997a). Such a specific timing of 7 d will be variable by region, speed of herbicide activity, and many other soil and environmental factors that influence plant growth. One of the major contributors to plant grow and development is the growing degree day (GDD; Spaunhorst et al. 2018). The accumulation of GDD is dependent on the high and low temperature for that particular day. Therefore, late summer months and southern climates will accumulate more GDD than spring months and northern climates.

Most current herbicide labels offer limited guidance as to when herbicides should be reapplied if necessary. The Cobra® herbicide label states that sequential applications can be made "after a minimum of 14 days have passed following the first application" (Anonymous 2015). However, such an interval may be to prevent excessive crop injury rather than maximize weed control. The Liberty® herbicide label states that some weeds may require a sequential application (Anonymous 2019a). The label also lists minimum sequential application timings that vary from 5 to 14 d depending on labeled crop indicating that the timing is meant to minimize crop injury. A 24(c) label has been issued for Liberty® herbicide only in the state of Georgia that allows two POST applications in a season with a minimum of a 5-d interval between applications (Anonymous 2020).

The application timing of respray herbicides will likely be especially important for nonsystemic herbicides such as PPO inhibitors and glufosinate, because efficacy is often limited by the lack of translocation out of treated plant tissue (Ritter and Coble 1981; Steckel et al. 1997b). The objectives of this research were to determine the most effective herbicides and timing of the herbicide respray following an initial application of glufosinate or fomesafen that failed to control Palmer amaranth. These herbicides were chosen because they were the most commonly used POST herbicides for control of glyphosate- and ALS inhibitor-resistant Palmer amaranth at the time the experiments were designed and initiated.

### **Materials and Methods**

Field trials were conducted in 2017 and 2018 near Medaryville, Indiana (41.0487°N, 87.0088°W) on glyphosate-resistant Palmer amaranth (susceptible to PPO-inhibitors). Palmer amaranth density was approximately 2 to 3 plants m<sup>-2</sup>. Large crabgrass [Digitaria sanguinalis (L.) Scop.] was the only other weed present in the field trial areas at a density of approximately 40 plants m<sup>-2</sup>. The soil type was Rensselaer fine sandy loam with organic matter content of 5.6%, pH 6.7. The experiment used a randomized complete block design with four replications and non-crop plots measuring 3 m wide by 9 m long. Palmer amaranth plants within the plots were allowed to grow until the average height reached approximately 30 cm, at which point five randomly selected 30-cm-tall plants in each plot were marked. The relatively large plant size was targeted to simulate a late commercial application that may result in incomplete control.

Two separate experiments were conducted which focused on initial applications to all plots of either glufosinate (Liberty, Bayer Crop Science, Research Triangle Park, NC) or fomesafen (Flexstar, Syngenta, Greensboro, NC). Glufosinate was applied at 450 g ai ha<sup>-1</sup> with a liquid ammonium sulfate (AMS) product (NPAK AMS Winfield Solutions, St. Paul, MN) at 3.4 kg ha<sup>-1</sup>. Fomesafen was applied at 280 g ai ha<sup>-1</sup> with AMS at 2.5% vol/ vol and methylated seed oil (MSO; MSO Ultra Precision Laboratories, Waukegan, IL) at 1% vol/vol to all plots. These herbicide rates were selected to simulate a failed commercial application and induce severe defoliation and herbicide injury, but ensure Palmer amaranth survival and regrowth. Treatments within each experiment (first application of glufosinate or fomesafen) were a factorial of the subsequent "respray" herbicide application timing (3) and the specific herbicide active ingredient (7) used in the respray. Following initial herbicide application, respray applications were targeted at three separate timings of 3, 7, or 11 d after the initial application (DAI) with one of seven herbicide treatments

Table 1. Respray herbicide treatments applied 4 to 5, 7, and 11 d after initial applications of glufosinate or fomesafen.

Herbicide	Rate <sup>a</sup>	Trade name	Formulation <sup>c</sup>	Manufacturer	Location	Adjuvant <sup>bc</sup>
	g ai ha <sup>-1</sup>					
Glufosinate (low)	450	Liberty®	SL	Bayer CropScience	Research Triangle Park, NC	AMS
Glufosinate (high)	736	Liberty®	SL	Bayer CropScience	Research Triangle Park, NC	AMS
Fomesafen	450	Flexstar®	SL	Syngenta Crop Protection LLC	Greensboro, NC	AMS + MSO
Lactofen	220	Cobra <sup>®</sup>	EC	Valent U.S.A. Corporation	Walnut Creek, CA	AMS + MSO
2,4-D	1120	Enlist One®	SL	Dow AgroSciences	Indianapolis, IN	COC
Dicamba	560	Engenia <sup>®</sup>	SL	BASF Corporation	Research Triangle Park, NC	MSO

<sup>&</sup>lt;sup>a</sup>Rate for 2,4-D and dicamba expressed as g ae ha<sup>-1</sup>.

Table 2. Environmental conditions at the time of initial and respray herbicide applications.

	2017					2018		
			Respray interv	al		Respray interval		
	Initial application	4 da	7 d	11 d	Initial application	5 d <sup>a</sup>	7 d	11 d
Application date	June 27	July 1	July 4	July 8	June 18	June 23	June 25	June 29
Start time	2:10	12:40	9:55	10:30	2:05	12:21	1:10	10:45
End Time	2:35	1:10	10:15	11:00	2:51	12:52	1:58	11:40
Temp	23	26	28	26	33	20	28	31
Relative humidity	44%	58%	81%	41%	61%	79%	59%	71%
Wind speed	7	8	4	6.5	12	5	2	5
Wind direction	W	W	W	N	W	W	W	N
Dew present	no	no	yes	yes	no	no	no	no
Soil moisture	dry	dry	adequate	adequate	dry	wet	adequate	adequate
Cloud cover	35%	25%	25%	60%	25%	95%	25%	10%
Height, cm	30	29	32	34	33	32	27	25

<sup>&</sup>lt;sup>a</sup>The first respray timing targeted a 3-d interval, but was delayed in both trial years due to rain.

described in Table 1. All herbicide applications were made with a  $\rm CO_2$ -pressurized backpack sprayer, equipped with XR11002 (Teejet Technologies, Wheaton, IL) flat-fan nozzles calibrated to deliver 140 L ha<sup>-1</sup> at 117 kPa. Inclement weather on the day of the 3 DAI timing in both 2017 and 2018 resulted in delayed applications to 4 and 5 DAI in 2017 and 2018, respectively. Environmental conditions at the time of herbicide applications are listed in Table 2.

## Data Collection and Analysis

Visual estimates of weed control ratings were taken for the entire plot at 7, 14, and 21 d after respray treatment based on a 0 to 100 scale (0 indicating no inhibition of plant growth and 100 corresponding to complete plant death). Individual Palmer amaranth survival was assessed for the marked plants by counting the number of new branches at 7 and 14 d after respray treatment. Branching data prior to initial application was not recorded, but in general, Palmer amaranth had retained apical dominance and branches were assumed to have a score of 0. Data were subjected to repeated measures analysis of variance (rmANOVA) using the GLIMMIX procedure in SAS 9.4 (SAS Institute Cary, NC). Control data were transformed using arcsine square root transformation, and branches were natural log transformed in order to better meet constant variance assumptions. Data were analyzed as a four factor (herbicide, timing, year, and block) repeated measures design with block as a random effect. In independent models, the repeated measure was visual estimate of control and number of branches. Means presented are a repeated measures average from all

evaluation timings in order to account for the fact that applications and data collections take place at staggered timings and is shown to be more informative than traditional ANOVA for a single time point (Nkurunziza and Milberg 2007). Means were separated using Tukey-Kramer adjusted HSD at  $\alpha=0.05$ . Data were pooled over nonsignificant factors when appropriate throughout the analysis.

## **Results and Discussion**

## Initial Application of Glufosinate

Palmer amaranth control from the single initial glufosinate application was 51% and plant regrowth consisted of 8.6 branches per plant when pooled across timings and evaluations. (Tables 3 and 4). The desired efficacy from the failed initial application for our research was approximately 50% control to allow for adequate detection of any increase or decrease in weed control from the subsequent respray application. Furthermore, herbicide failure of this level allows for vigorous regrowth while still observing a severe phytotoxic effect similar to that reported by Mager et al. (2006a). In that experiment, herbicide failure was simulated physically by clipping plants at the middle node. Clipping at the middle node corresponds to approximately 50% control because a plant's propensity for regrowth is related to the height of plant cutting, with lower cutting heights producing less regrowth than greater cutting heights (Andreasen et al. 2002; Mager et al. 2006a). In the present study, the number of branches per plant was inversely associated with that of the control. The number of branches in the plots receiving no respray herbicide indicated how much regrowth

bAdjuvant rates: AMS, 3.4 kg ai ha<sup>-1</sup> for glufosinate and 2.5% vol/vol for lactofen and fomesafen (N-Pak, Winfield Solutions LLC, St Paul, MN); MSO, 1% vol/vol (MSO Ultra, Precision Laboratories, Waukegan, IL); COC, 1% vol/vol (Prime Oil, Winfield Solutions LLC, St. Paul, MN).

<sup>&</sup>lt;sup>c</sup>Abbreviations: AMS, ammonium sulfate; MSO, methylated seed oil; COC, crop oil concentrate; SL, soluble liquid; EC, emulsifiable concentrate

**Table 3.** Control of Palmer amaranth after herbicide respray treatments applied at 4 to 5, 7, or 11 d after a failed application of glufosinate in field research conducted in 2017 and 2018.<sup>a</sup>

Factor	Control <sup>b</sup>
Herbicide	%
None	51 e
2,4-D	91 cd
Dicamba	88 d
Fomesafen	93 bc
Glufosinate (low) <sup>c</sup>	95 ab
Glufosinate (high) <sup>c</sup>	97 a
Lactofen	87 d
Timing (days after initial application)	
4–5	87 a
7	87 a
11	84 b

<sup>&</sup>lt;sup>a</sup>Data are presented as main effects and pooled by year due to insignificant interactions of year, herbicide, and application timing. Means presented are a repeated measures statistic derived from evaluation timings of 7, 14, and 21 d after respray applications.

was occurring through axillary meristems. The reduction in branches in a resprayed treatment compared with the treatment receiving no respray herbicide indicated how much regrowth has been controlled.

All resprayed herbicide treatments resulted in greater control of 36 to 46 percentage points than treatments receiving no respray application (Table 3). Fomesafen and glufosinate (both rates) as respray treatments resulted in control increase of 5 to 10 percentage points compared to lactofen and dicamba treatments. Treatments applied 4 to 5 and 7 d after the initial application (DAI) on average resulted in 3 percentage points greater control than respray treatments applied 11 DAI.

Branch data followed very similar trends as the control data. All herbicide respray treatments resulted in 5.2 to 8.2 fewer branches (60% to 95% reduction) than treatments receiving no herbicide respray (Table 4). Treatments receiving glufosinate at either rate as a respray treatment resulted in 1.7 to 3 (63% to 88%) fewer branches than dicamba and lactofen respray treatments. Respray treatments applied 4 to 5 and 7 DAI resulted in fewer branches than when applied 11 DAI by 1.7 to 2.5 (40% to 58% reduction) branches.

The herbicide timing effect is consistent with that reported by Randell et al. (2020) who observed that Palmer amaranth control declined when the respray herbicide applications exceeded 10 d after initial application. Merchant et al. (2014), however, observed that Palmer amaranth control increased when sequential applications of 2,4-D followed by (fb) 2,4-D, 2,4-D fb glufosinate, and 2,4-D + glufosinate fb 2,4-D + glufosinate were delayed from 5 d to 10 or 15 d after initial application. To our knowledge there is no precedent in the literature to compare the efficacy of glufosinate or fomesafen followed by applications of various other foliar herbicides other than our own similar research using waterhemp (Haarmann et al. 2020). Herbicide efficacy trends between the two species were similar.

Respray applications of glufosinate were most effective for control of Palmer amaranth that survived a POST glufosinate application. Fomesafen and 2,4-D respray applications were also quite effective (control >90%), but less so than glufosinate applied at the high rate. Timing respray applications prior to 11 DAI also slightly improved control of the herbicides applied.

**Table 4.** Average number of Palmer amaranth branches per marked plant 7 and 14 d after herbicide respray treatments applied at 4 to 5, 7, or 11 d after a failed application of glufosinate in field research conducted in 2017 and 2018.<sup>a</sup>

Factor	No. of branches <sup>b</sup>
Herbicide	
None	8.6 a
2,4-D	2.6 bc
Dicamba	3.4 b
Fomesafen	1.5 bc
Glufosinate (low) <sup>c</sup>	1.0 cd
Glufosinate (high) <sup>c</sup>	0.4 d
Lactofen	2.7 b
Timing (days after initial application)	
4–5	2.6 b
7	1.8 b
11	4.3 a

<sup>a</sup>Data are presented as main effects and were pooled by year due to nonsignificant year, herbicide, and timing interactions. Means presented are a repeated measures statistic derived from evaluation timings of 7 and 14 d after respray applications.

**Table 5.** Control of Palmer amaranth after herbicide respray treatments applied at 4 to 5, 7, or 11 d after a failed application of fomesafen in field research conducted in 2017 and 2018.<sup>a</sup>

	Days after initial fomesafen application <sup>b</sup>			
Respray herbicide	4 to 5	7	11	
		%		
None	63 fg	62 fg	52 g	
2,4-D	88 a-e	91 a-d	83 c-e	
Dicamba	87 a-e	89 a-e	83 c-e	
Fomesafen	86 c-e	86 b-e	86 a-e	
Glufosinate (low) <sup>c</sup>	93 a-c	91 a-d	91 a-d	
Glufosinate (high) <sup>c</sup>	94 ab	90 a-d	96 a	
Lactofen	76 ef	80 de	78 de	

<sup>a</sup>Data were analyzed as a two-way interaction of respray herbicide and application timing pooled by year due to significant two-way interaction of herbicide and application timing and insignificant three-way interaction of respray herbicide, application timing and year. Means presented are a repeated measures statistic derived from evaluation timings of 7, 14, and 21 d after respray applications.

## Initial Application of Fomesafen

Control data were analyzed as a two-way interaction of application timing and respray herbicide (P = 0.0235) and pooled by year due to insignificant three-way interaction (P = 0.1907). Palmer amaranth control from a single application of fomesafen was 52% to 63% (Table 5). Our research objective for the targeted control was 50% control of Palmer amaranth, which is similar to that of glufosinate. Control of 96% was achieved with glufosinate applied at the high rate 11 d after initial application. Similar levels of control were achieved with glufosinate at both rates applied at any timing as well as with 2,4-D and dicamba applied up to 7 DAI. The main source of the interaction was in 2,4-D and dicamba application timings. While control of Palmer amaranth with each herbicide was similar across timings, herbicides applied 11 DAI were less than the greatest control in the experiment, unlike applications made 4 to 7 DAI. Lactofen applied 4 to 5 DAI did not improve Palmer amaranth control compared to no respray application,

 $<sup>^{</sup>b}$ Mean separation for control was based on arcsin square root transformation, Data presented are means from nontransformed data. Means within a column followed by the same letter are not different based on the Tukey HSD test at  $\alpha = 0.05$ .

 $<sup>^{\</sup>rm c}$ Low and high rates of glufosinate were 450 and 736 g ai ha $^{-1}$ , respectively

 $<sup>^</sup>b$ Mean separation for branches was based on natural log transformation. Data presented are means from nontransformed data. Means within a column followed by the same letter are not different based on the Tukey HSD test at  $\alpha=0.05$ .

<sup>&</sup>lt;sup>c</sup>Low and high rates of glufosinate were 450 and 736 g ai ha<sup>-1</sup>, respectively.

 $<sup>^{</sup>b}$ Mean separation for control was based on arcsin square root transformation. Data presented are means from nontransformed data. Means followed by the same letter are not different based on the Tukey HSD test at  $\alpha = 0.05$ .

<sup>&</sup>lt;sup>c</sup>Low and high rates of glufosinate were 450 and 736 g ai ha<sup>-1</sup>, respectively.

**Table 6.** Average number of Palmer amaranth branches per marked plant after herbicide respray treatments applied at 4 to 5, 7, or 11 d after a failed application of fomesafen in field research conducted in 2017 and 2018.<sup>a</sup>

		Branches <sup>b</sup>				
Respray herbicide		2017		2018  Days after initial fomesafen application		
	Days af	ter initial fomesafen app	olication			
	4	7	11	4	7	11
	-	No			No	
None	8.6 a	6.3 ab	9.3 a	8.7 a	1.2 a-e	1.7 a-e
2,4-D	1.5 a-d	0.3 cd	4.7 a-d	2.5 a-d	0.8 a-e	4.1 a-d
Dicamba	2.9 a-c	0.3 d	5.5 a-d	2.6 a-d	0.9 a-e	4.2 a-c
Fomesafen	4.4 ab	2.8 a-c	2.4 a-d	1.6 a-d	0.5 a-e	2.9 a-e
Glufosinate (low) <sup>c</sup>	2.8 a-d	0.7 b-d	2.0 a-d	0.0 e	0.5 b-e	0.6 de
Glufosinate (high) <sup>c</sup>	1.4 a-d	1.1 a-d	1.9 a-d	0.0 e	0.5 c-e	0.3 c-e
Lactofen	4.6 ab	4.1 a-c	2.1 a-d	1.3 a-e	0.5 c-e	7.7 ab

<sup>&</sup>lt;sup>a</sup>Data were separated by year due to significant three-way interaction of year, herbicide, and application timing. Means presented are a repeated measures statistic derived from evaluation timings of 7 and 14 d after respray applications.

whereas lactofen applied 7 or 11 DAI improved control compared to no respray application.

High variability in individual plant response produced few treatment differences for branch data, especially in 2018 (Table 6). Interactions occurred because little to no regrowth had occurred on the marked plants in 2018 with the exception of a few seemingly random treatments having high numbers of branches. However, similar numerical trends are present compared to visual ratings.

When fomesafen was the initial herbicide, there was an overall lack of an herbicide timing effect. Noteworthy is the efficacy of lactofen and fomesafen, which was not different across timings. Such a result contrasts with results reported by Sperry et al. (2017) when lactofen efficacy on Palmer amaranth improved with delayed sequential application timings. In that experiment, lactofen applied to Palmer amaranth in sequence was most effective when applied 15 d after the initial lactofen application in comparison to 5 d after initial application. The presence of a timing effect observed by Sperry et al. (2017) may be due to recovery from plant stress and accumulation of biomass, making further control difficult. When an herbicide is applied, a stress response is induced in plants, which causes induction of detoxification and ROS quenching pathways (Salas-Parez at al 2018). During such a time, the plant is likely to be less susceptible to herbicides (Zhou et al. 2007). Velvetleaf under abiotic stress was less susceptible to glyphosate than nonstressed velvetleaf (Zhou et al. 2007). After a plant survives the sublethal dose, growth will resume. At some point, the plant may fully recover and resume growth depending on the severity of injury (Mager et al. 2006a). Large plants are less susceptible to foliarapplied herbicides, so every passing day makes it more difficult to control of weeds that have survived a herbicide application (Steckel at al. 1997a). Glufosinate and PPO inhibitors differ in their mechanisms of action, so the course of events from herbicide injury to recovery may vary between the two herbicides (personal observation).

Herbicide respray timings in the present study only extended to 11 d after the initial application and were spaced 3 d apart. Such a treatment structure may have been inadequate to capture the full breadth of plant response especially the treatment differences observed by other research groups. Other research groups typically used treatment structures with longer sequential application

intervals (Merchant et al. 2014; Randell et al. 2020; Sperry et al. 2017). In addition, other researchers were also located in the southern United States, whereas Indiana is substantially farther north. Therefore, the application intervals used by other researchers are likely spaced even further apart than the present study in terms of GDD accumulation. Whereas each research group's observed responses are different, one thing that is consistent is that the studies were not discrete with each timing, but more of a binary response where after a particular number of days, the plant response to the sequential herbicide changed (Merchant et al. 2014; Randell et al. 2020; Sperry et al. 2017).

Overall, the data partially support our hypothesis that there is an optimum timing for herbicide applications. Respray herbicide applications had a timing effect when glufosinate was the initial herbicide, but not when fomesafen was the initial herbicide. The glufosinate timing effect is consistent with that reported by Randell et al. (2020), who found that sequential glufosinate applications are more effective when applied sooner than 10 d after the initial glufosinate application compared to more than 10 d.

The results presented here illustrate that scouting fields soon (5 to 7 d rather than 14 to 21 d) after POST herbicide applications is critically important to ensure a successful application. If glufosinate application fails and Palmer amaranth regrows, producers should reapply it within 7 d of noticing the herbicide failure. When reapplying the herbicide, be especially cognizant of factors that can compromise a spray application and plan applications around optimal spray conditions.

Herbicide selection in a respray situation is often limited by crop herbicide tolerance, calendar date, and label restrictions. However, recent herbicide resistance trait combinations in crops such as Enlist E3\* and Xtendflex\* are allowing growers to use more herbicides POST than ever before. In both experiments, the effect of specific respray herbicide was much greater than the effect of respray application timing. Nearly all respray treatment and timing combinations improved control compared to no respray. Across both trials, glufosinate, particularly at the 736 g ha<sup>-1</sup> rate, was the most efficacious and consistent herbicide treatment. Despite glufosinate being very effective when applied as a respray following a failed glufosinate application, repeated applications of the same herbicide mechanism of action should be avoided to delay selection for herbicide-resistant biotypes (Norsworthy et al. 2012).

<sup>&</sup>lt;sup>b</sup>Mean separation for branches was based on natural log transformation. Data presented are means from nontransformed data. Means within a trial year followed by the same letter are not different based on the Tukey HSD test at  $\alpha = 0.05$ .

<sup>&</sup>lt;sup>c</sup>Low and high rates of glufosinate were 450 and 736 g ai ha<sup>-1</sup>, respectively

One important consideration in respray scenarios is the herbicide label limitations. Many situations will lead to lactofen being the only legal respray option due to advancing crop growth stage, advancing calendar date, and crop herbicide tolerance. Glufosinate and dicamba may be applied to soybeans only before the R1 growth stage (Anonymous 2018a; Anonymous 2019a). Similarly, 2,4-D must be applied before the R2 growth stage (Anonymous 2019c). Calendar date restrictions are a concern for both fomesafen and dicamba applications based on geography (Anonymous 2018a; Anonymous 2019b). Additional dicamba restrictions include presence of a temperature inversion, wind speeds below 4.8 or above 24 km h<sup>-1</sup>, rain forecast within 4 h, and sensitive neighboring crops.

Although statistical comparisons were not made between the two experiments, fomesafen and lactofen efficacy were generally greater when applied after an initial application of glufosinate rather than initial application of fomesafen. Vila-Aiub and Ghersa (2005) observed a similar effect in Lolium with repeated nonlethal applications of diclofop. The repeated herbicide applications may have induced sustained upregulation of a diclofop detoxification pathway (Burns et al. 2018). The effect was not a heritable resistance mechanism (Vila-Aiub and Ghersa 2005). Interestingly, the present study did not observe the same effect with sequential applications of glufosinate. Glufosinate covalently modifies the target enzyme glutamine synthetase rather than transient inhibition like most herbicides. Therefore, the conventional mechanism of xenobiotic detoxification via cytochrome P450 and glutathione S-transferase may not apply since glufosinate is permanently bound to glutamine synthetase (Takano et al. 2020).

Synthetic auxins applied as respray herbicides were neither the most effective nor least effective and can be suitable respray herbicides in some situations. A systemic herbicide has the benefit of translocation since leaf area and interception are limited in herbicide failure scenarios, but the issue of reduced herbicide interception is still a problem regardless or respray herbicide.

Future research should investigate more combinations of sequential herbicide applications, particularly instances of synthetic auxin herbicide failure, as well as herbicide tank mix combinations. Recent experiments investigating planned sequential POST applications demonstrate that herbicide combinations are necessary in order to control large weeds (Craigmyle et al. 2013; Merchant et al. 2014; Vann et al. 2017). Other research should focus on the physiological basis of plant regrowth such as the contribution of dormant axillary meristems and the status of regrowth at the time of the sequential application.

These data are valuable for the foundation of extension recommendations for herbicide respray applications. Recommendations based on our research for glufosinate herbicide failure on Palmer amaranth are to apply fomesafen, 2,4-D or dicamba prior to 11 d after the initial herbicide application when crop tolerance, calendar date, and other label restrictions allow. A timing of 11 d after initial application generally corresponds to fresh regrowth of 1 to 2 cm. Glufosinate resprays are also effective, but should be avoided for resistance management purposes. In the case of POST PPO inhibitor failure and Palmer amaranth regrowth, glufosinate, 2,4-D, or dicamba should be applied with glufosinate as the preferred respray herbicide where possible. Respray applications should be made as soon as failure is detected so as to minimize Palmer amaranth regrowth.

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## **References**

- Anonymous (2015) Cobra® herbicide product label. Walnut Creek, CA: Valent USA Corporation
- Anonymous (2018a) Engenia® herbicide product label. Research Triangle Park, NC: BASF Corporation
- Anonymous (2018b) Xtendimax® herbicide product label. St. Louis, MO: Bayer Crop Science
- Anonymous (2019a) Liberty® herbicide product label. Research Triangle Park, NC: BASF Corporation
- Anonymous (2019b) Flexstar® herbicide product label. Greensboro, NC: Syngenta Crop Protection LLC
- Anonymous (2019c) Enlist One® herbicide product label. Indianapolis, IN: Corteva Agriscience
- Anonymous (2020) Liberty® herbicide FIFRA Sec. 24(c) Special Local Need Label. Research Triangle Park, NC: BASF Corporation
- Andreasen C, Hansen CH, Moller C, Kjaer-Pedersen NK (2002) Regrowth of weed species after cutting. Weed Technol 16:873–879
- Bagavathiannan MV, Norsworthy JK (2016) Multiple-herbicide resistance is widespread in roadside Palmer amaranth populations. PLoS One 11: e0148748
- Burns EE, Barbara KK, Mohammed YR, Bothner B, Dyer WE (2018).
  Constitutive Redox and Phosphoproteome Changes in Multiple Herbicide Resistant Avena fatua L. Are Similar to Those of Systemic Acquired Resistance and Systemic Acquired Acclimation. J Plant Physiol 220:105–114
- Coetzer E, Al-Khatib K, Loughin TM (2001) Glufosinate efficacy, absorption, and translocation in amaranth as affected by relative humidity and temperature. Weed Sci 49:8–13
- Craigmyle BD, Ellis JM, Bradley KW (2013) Influence of weed height and glufosinate plus 2,4-D combinations on weed control in soybean with resistance to 2,4-D. Weed Technol 27:271–280
- Haarmann JA, Young BG, Johnson WG (2020). Control of waterhemp (Amaranthus tuberculatus) regrowth after failed applications of glufosinate or fomesafen. Weed Technol doi: 10.1017/wet.2020.58
- Heap I (2020) International Survey of Herbicide Resistant Weeds. http://www.weedscience.org. Accessed: March 25, 2019
- Klingaman TE, Oliver LR (1994) Palmer amaranth interference in soybeans (*Glycine max*). Weed Sci 42:523–527
- Li RH, Qiang S (2009) Composition of floating weed seeds in lowland rice fields in China and the effects of irrigation frequency and previous crops. Weed Res 49:417–427
- Lillie KJ, Giacomini DA, Tranel PJ (2020) Comparing Responses of Sensitive and Resistant Populations of Palmer amaranth (*Amaranthus palmeri*) and Waterhemp (*Amaranthus tuberculatus* var. *rudis*) to PPO Inhibitors. Weed Technol 34:140–146
- Mager HJ, Young BG, Preece JE (2006a) Characterization of compensatory weed growth. Weed Sci 54:274–281
- Mager HJ, Young BG, Preece JE (2006b) Efficacy of POST herbicides on weeds during compensatory growth. Weed Sci 54:321–325
- Merchant RM, Culpepper AS, Eure PM, Richburg JS, Braxton LB (2014) Salvage Palmer Amaranth Programs Can Be Effective in Cotton Resistant to Glyphosate, 2,4-D, and Glufosinate. Weed Technol 28:316–322
- Nkurunziza L, Milberg, P (2007) Repeated grading of weed abundance and multivariate methods to improve efficacy in on-farm weed control trials: technical report. Weed Biol Manag 7:132–139
- Norsworthy JK, Griffith G, Griffin T, Bagavathiannan M, Gbur EE (2014) Infield movement of glyphosate-resistant Palmer amaranth (*Amaranthus palmeri*) and its impact on cotton lint yield: evidence supporting a zero-threshold strategy. Weed Sci 62:237–249
- Norsworthy JK, Smith KL, Steckel LE, Koger CH (2009) Weed seed contamination of cotton gin trash. Weed Technol 23:574–580
- Norsworthy JK, Ward SM, Shaw DR, Llewellyn RS, Nichols RL, Webster TM, Bradley KW, Frisvold G, Powles SB, Burgos NR, Witt WW, Barrett M (2012) Reducing the risks of herbicide resistance: Best management practices and recommendations. Weed Sci 60:31–62
- Randell TM, Hand LC, Vance JC, Culpepper AS (2020) Interval between Sequential Glufosinate Applications Influences Weed Control in Cotton. Weed Technol doi: 10.1017/wet.2020.16

- Ritter RL, Coble HD (1981) Influence of temperature and relative humidity on the activity of acifluorfen. Weed Sci 29:480–485
- Salas-Perez RA, Saski CA, Noorai RE, Srivastava SK, Lawton-Rauh AL, Nichols RL, Roma-Burgos N (2018) RNA-Seq Transcriptome Analysis of *Amaranthus palmeri* with Differential Tolerance to Glufosinate Herbicide. PLoS One 13:e0195488
- Sikkema PH, Brown L, Shropshire C, Spieser H, Soltani N (2008) Flat fan and air induction nozzles affect soybean herbicide efficacy. Weed Biol Manag 8:31–38
- Spaunhorst DJ, Devkota P, Johnson WG, Smeda RJ, Meyer CJ, Norsworthy JK (2018) Phenology of five Palmer amaranth (*Amaranthus palmeri*) populations grown in northern Indiana and Arkansas. Weed Sci 66:457–469
- Sperry BP, Ferrell JA, Smith HC, Fernandez VJ, Leon RG, Smith CA (2017) Effect of sequential applications of protoporphyrinogen oxidase-inhibiting herbicides on Palmer amaranth (*Amaranthus palmeri*) control and peanut response. Weed Technol 31:46–52
- Steckel GJ, Hart SE, Wax LM (1997b) Absorption and translocation of glufosinate on four weed species. Weed Sci 45:378–381
- Steckel GJ, Wax LM, Simmons FW, Phillips WH (1997a) Glufosinate efficacy on annual weeds is influenced by rate and growth Stage. Weed Technol 11:484–488

- Takano HK, Beffa R, Preston C, Westra P, Dayan FE (2020) A novel insight into the mode of action of glufosinate: how reactive oxygen species are formed. Photosynth Res 144:361–372
- Vila-Aiub MM, Ghersa CM (2005) Building up resistance by recurrently exposing target plants to sublethal doses of herbicide. Eur J Agron 22: 195–207
- Vann RA, York AC, Cahoon CW, Buck TB, Askew MC, Seagroves RW (2017) Glufosinate plus dicamba for rescue Palmer amaranth control in XtendFlexTM cotton. Weed Technol 31:666–674
- Van Wychen L (2019) 2019 Survey of the most common and troublesome weeds in broadleaf crops, fruits & vegetables in the United States and Canada. Weed Science Society of America National Weed Survey Dataset. http://wssa.net/wp-content/uploads/2019-Weed-Survey\_broadleaf-crops. xlsx. Accessed: August 29, 2020
- Ward SM, Webster TM, Steckel LE (2013) Palmer amaranth (*Amaranthus palmeri*): A review. Weed Technol 27:12–27
- Wichert RA, Bozsa R, Talbert RE, Oliver LR (1992) Temperature and relative humidity effects on diphenylether herbicides Weed Technol 6:19–24
- Zhou J, Tao B, Messersmith CG, Nalewaja JD (2007) Glyphosate efficacy on velvetleaf (*Abutilon theophrasti*) is affected by stress. Weed Sci 55: 240–244