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Palmer amaranth control, fecundity, and seed viability from soybean herbicides applied at first female inflorescence

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Research Article

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Abstract

Palmer amaranth is an extremely troublesome weed for soybean growers because of its aggressive growth, adaptability, prolific seed production, and widespread resistance to many herbicides. Studies were initiated to determine the effects of herbicide application at first female inflorescence on Palmer amaranth control, biomass, seed production, cumulative germination, and seed viability. Enlist (2,4-D–resistant) soybean and Xtend (dicamba-resistant) soybean were planted and various combinations of either 2,4-D or dicamba with and without glufosinate and/or glyphosate were applied at first visible female Palmer amaranth inflorescence. Mixtures of 2,4-D + glufosinate and 2,4-D + glufosinate + glyphosate provided the greatest control at 4 wk after treatment in Enlist soybean. Similarly, in Xtend soybean, combinations of dicamba + glufosinate and dicamba + glufosinate + glyphosate provided the greatest control. The greatest reductions in biomass were from combinations of auxin herbicides (2,4-D or dicamba) plus glufosinate with and without glyphosate. Seed production was reduced most by treatments containing at least one effective site of action: an auxin herbicide (2,4-D or dicamba) or glufosinate. In contrast to previous research, cumulative germination and seed viability were not affected by herbicide treatments. This research indicates the efficacy of auxin herbicides or glufosinate alone and in combination to reduce the seed production of Palmer amaranth when applied at first female inflorescence. More research is needed to evaluate the full potential for applications of these herbicides at flower initiation to mitigate the evolution of herbicide resistance.

Introduction

Palmer amaranth is one of the most prevalent and troublesome weeds in agronomic crops in the United States (Van Wychen 2019; Ward et al. 2013; Webster and Nichols 2012). Characteristics of Palmer amaranth, such as aggressive growth, high photosynthesis rate, and high phenotypic plasticity, allow for wide adaption and competitiveness in a variety of crops (Ward et al. 2013). In soybean, Palmer amaranth can reduce yields 68% to 79% (Bensch et al. 2006; Klingaman and Oliver 1994). Palmer amaranth is an obligate outcrosser, which allows for high genetic diversity and increases the probability of herbicide-resistant individuals to be found in fields (Franssen et al. 2001). Resistance to eight different sites of action (by acetolactate synthase inhibitors, microtubule inhibitors, synthetic auxins, photosystem II inhibitors, 5-enolpyruvylshikimate-3-phosphate synthase inhibitors, protoporphyrinogen oxidase inhibitors, long-chain fatty acid inhibitors, and 4-hydroxyphenylpyruvate dioxygenase inhibitors) has been confirmed in Palmer amaranth (Brabham et al. 2019; Culpepper et al. 2006; Gossett et al. 1992; Heap 2020; Horak and Peterson 1995; Jhala et al. 2014; Kumar et al. 2019; Oliveira et al. 2018; Salas et al. 2016). The extended germination window of Palmer amaranth seeds requires season-long management and increases the likelihood of weeds surviving to produce seed (Steckel et al. 2004). Palmer amaranth can produce up to 600,000 seeds plant^{-1}; thus, one mature plant can make large additions to the soil seedbank (Keeley et al. 1987; Steckel et al. 2004).

Much research has been conducted investigating various herbicides and herbicide programs for Palmer amaranth control both PRE and POST when weeds were the most susceptible (approximately 10 cm tall) (Blake et al. 2018; Cahoon et al. 2015; Houston et al. 2019; Merchant et al. 2013). Limited research has been conducted on the use of herbicides to control large Palmer amaranth and their potential to limit seed production. Late-season applications of many herbicides, applied at flowering, have the potential to reduce seed production in a variety of weed species (Bennett and Shaw 2000; Brewer and Oliver 2007; Hill et al. 2016; Walker and Oliver 2008). Timing of these applications is critical because a 2-wk delay in herbicide application can result in little to no impact on viable seed production (Hill et al. 2016). Contact herbicides and systemic herbicides, applied at flowering, may reduce seed production (Bae et al. 2017; Bennett and Shaw 2000; Chang and Vanden Born 1971; Gougler and Geiger 1981). For example, glyphosate applied to flowering, glyphosate-susceptible Palmer amaranth can reduce seed production up to 90% compared with plants that did not receive a glyphosate application (Walker and Oliver 2008). In addition to reducing overall seed production, glyphosate inhibits protein accumulation in seeds of glyphosate-susceptible weeds, potentially reducing seed viability as well (Bennett and Shaw 2000; Cerdeira et al. 1985). Unfortunately, glyphosate resistance in Palmer amaranth is widespread in the United States (Heap 2020). Glyphosate-resistant Palmer amaranth treated at flowering with glyphosate resulted in a 62% to 81% reduction in seed production, whereas glufosinate, dicamba, or 2,4-D resulted in greater than 75% reduction in seed production (Jha and Norsworthy 2012). Seed viability was reduced relative to the nontreated by glufosinate, dicamba, 2,4-D, or glyphosate (46%, 52%, 58%, and 61%, respectively).

To our knowledge, combinations of glufosinate + 2,4-D or dicamba as well as 2,4-D or dicamba with and without glyphosate have not been evaluated for the reduction in seed production and viability when applied at first visible female inflorescence. Combining multiple effective herbicide sites of action improves control relative to applying individual herbicide sites of action (Lawrence et al. 2018; Vann et al. 2017). Seed fecundity has seldom been evaluated with herbicide combinations to determine if the same effect occurs. New or soon-to-be-released soybean herbicide–resistance traits make these herbicide combinations a potential option for growers. Therefore, research was conducted to evaluate the potential of combinations of dicamba or 2,4-D with glufosinate and/or glyphosate to provide enhanced Palmer amaranth control and decreased seed production and viability.

Materials and Methods

Study Sites

Two field sites were established in 2019. One site was in Blackstone, VA (37.084°N, 77.97°W) on an Appling sandy loam (fine, kaolinitic, thermic Typic Kanhapludults) and the second was in Blacksburg, VA (37.19°N, 80.58°W) on a Guernsey silt loam (fine, mixed, superactive, mesic Aquic Hapludalfs). Naturalized Palmer amaranth populations were present at both sites and known to be glyphosate and acetolactate-synthase resistant. Both sites were disked to reduce surface residue in December and early burndown herbicide applications of glyphosate (Roundup Powermax; Monsanto Company, St. Louis, MO) at 1.26 kg ae ha^{-1} plus glufosinate (Interline; United Phosphorus, Inc., King of Prussia, PA) at 0.656 kg ai ha^{-1} were made 2 wk before planting.

At both field sites, two separate experiments were conducted for each soybean trait (Enlist and Xtend), resulting in two replications in space for each experiment. Each experiment was designed as a randomized complete block with four replications. The plot size was 3 m wide by 8 m long. Herbicide treatments were made using CO_2 backpack spray equipment and four TeeJet 11002XR nozzles (Spraying Systems Co.; Wheaton, IL) for all treatments except dicamba- and 2,4-D–containing treatments, for which TeeJet 11002 TTI nozzles (Spraying Systems Co.) were used, as required by product labeling (Anonymous 2019a, 2019b). Spray equipment was calibrated to apply 140 L ha^{-1} at 207 kPa.

Treatments for both experiments are presented in Table 1 and consisted of auxin herbicides (dicamba or 2,4-D choline,) glufosinate, and glyphosate applied alone and in combinations; a nontreated check served as the control. Recommended adjuvants were used for each treatment; the brand of dicamba used in this trial does recommend an adjuvant when used by itself (Table 1).

Field Management and Data Collection

Soybean were planted in 76-cm wide rows at 66,800 seeds ha^{-1} on May 7, 2019, in Blackstone and on May 16, 2019, in Blacksburg. Two soybean herbicide–resistant traits were used: Enlist E3 '5219E' (Axis Seed, Adel, IA) and Xtend 'AG51X8' (Asgrow; Bayer Crop Science, St. Louis, MO). The Enlist E3 seeds were nontreated; Xtend seeds were treated with Acceleron (metalaxyl, fluxapyroxad, pyraclostrobin, imidacloprid; Bayer Crop Science). In Blacksburg, soybean had not been grown previously, so seed were inoculated with rhizobia (N-Dure; Verdesian Life Sciences, Cary, NC) at a rate of 1,051 g ha^{-1}.

At 3 wk after planting and before herbicide treatment application, a broadcast glyphosate application was made to the entire experiment (Roundup Powermax; Bayer Crop Science) to remove glyphosate-susceptible weeds. When sufficient Palmer amaranth emergence had occurred, 10 plants plot^{-1} (0.4 plants m^{-2}) centered between soybean rows were identified and shielded using cups; then a broadcast application was made of fomesafen at 0.42 kg ai ha^{-1} (Reflex; Syngenta Corp., Basel, Switzerland), glyphosate at 1.26 kg ae ha^{-1} (Roundup Powermax; Bayer Crop Science), and *S*-metolachlor at 0.59 kg ha^{-1} (Moccasin; United Phosphorus, Inc.) to control nonshielded weeds and reduce further weed establishment. The broadcast application was made on May 21 in Blackstone and June 26 in Blacksburg.

Of the 10 plants plot^{-1}, female plants were identified when flowers were approximately 1 cm tall by visual examination and touch for spiny bracts, which are characteristic of female flowers. Female plants were identified and flagged immediately before treatment application, which occurred at soybean stage R2 (Table 2). If fewer than 10 female plants plot^{-1} at the same growth stage were present, as many as possible were flagged. Because Xtend soybean is not glufosinate resistant, plots treated with dicamba + glufosinate had tarps placed over soybean rows immediately before treatment application to avoid herbicide contact and subsequent injury from glufosinate. These tarps were removed immediately after treatment application.

Visible control ratings, using a scale of 0 (no control) to 100 (complete plant death relative to the nontreated plants) (Frans et al. 1986), were conducted 2 and 4 wk after treatment (WAT). At soybean harvest, flagged plants that survived herbicide application were counted to yield a mortality rating, and entire plants were clipped at the soil surface to collect biomass. Soybean harvest and weed biomass collection occurred on October 15, 2019, in Blackstone. Biomass collection occurred on October 14 and soybean harvest occurred on November 5 in Blacksburg. Palmer amaranth retains 95% or greater seed at soybean harvest (Schwartz et al. 2016; Schwartz-Lazaro et al. 2017).

After collection, plants were bagged, dried at 50 C for 72 h, weighed, threshed, sieved, and seed cleaned using an air-column seed cleaner to collect total seed biomass, 100 seed weight, and total seed production on a per plant basis. Seed processing was completed by mid December 2019, and germination assays began in January 2020. This period is similar to that used by Jha et al. (2010) to achieve optimal Palmer amaranth germination.

Germination assays were conducted using random, homogenized, 100-seed subsamples from each plot. These seeds were placed on two layers of filter paper (Fisherbrand P5; Thermo

Table 1. Herbicide treatments for determination of Palmer amaranth control and fecundity after treatment at first female inflorescence.

Soybean trait	Herbicide	Trade name	Rate	Manufacturer[a]	Location
			g ai or ae ha^{-1}		
Enlist®	Nontreated	Not applicable			
	Glyphosate	Roundup Powermax®	1,260	Bayer Crop Science	St. Louis, MO
	2,4-D	Freelexx®	1,060	Corteva Agriscience	Wilmington, DE
	2,4-D + glyphosate	Freelexx® + Roundup Powermax®	1,060 + 1,260		
	Glufosinate[b]	Interline®	656	United Phosphorus	King of Prussia, PA
	Glufosinate + glyphosate[b]	Interline® + Roundup Powermax®	656 + 1,260		
	2,4-D + glufosinate[b]	Freelexx® + Interline®	1,060 + 656		
	2,4-D + glufosinate + glyphosate[b]	Freelexx® + Interline® + Roundup Powermax®	1,060 + 656 + 1,260		
Xtend®	Nontreated	Not applicable			
	Glyphosate	Roundup Powermax®	1,260		
	Dicamba	Xtendimax®	560	Bayer Crop Science	St. Louis, MO
	Dicamba + glyphosate[c]	Xtendimax® + Roundup Powermax®	560 + 1,260		
	Dicamba + glufosinate[c,d]	Xtendimax® + Interline®	560 + 656		
	Dicamba + glufosinate + glyphosate[c,d]	Xtendimax® + Interline® + Roundup Powermax®	560 + 656 + 1,260		

[a]Only listed at first mention.
[b]Included ammonium sulfate (3.36 kg ha^{-1}; DSM Chemicals, Augusta, GA), per product label recommendation.
[c]Included Reign (0.5% vol/vol; Loveland Products, Loveland, CO), per product label recommendation.
[d]Tarps were used to shield Xtend soybean from glufosinate.

Table 2. Treatment application data and corresponding crop and weed data in 2019.

Site characteristic	Blackstone, VA	Blacksburg, VA
Treatment application date	July 2	July 19
Temperature at application, C	33	35
Humidity at application, %	52	69
Cloud cover at application, %	0	20
Crop stage at application	R2	R2
Crop height at application, cm	46	Enlist: 71; Xtend: 91
Palmer amaranth height at application, cm	81	66

Fisher Scientific, Waltham, MA) with 5 mL of deionized water. Petri dishes (Fisherbrand 100 mm × 15 mm; Thermo Fisher Scientific) were wrapped in parafilm (Parafilm PM-992; Amcor, Zurich, Switzerland) and placed under 12-h photoperiod at 23 C for 3 wk for the initial light period, followed by a 3-wk dark, cold stratification period at 3 C (Buhler and Hoffman 1999). This process was then repeated for a total of three light periods and two dark, cold stratification periods. Germination was evaluated weekly during the light periods with counts and removal of germinated seed. The appearance of a radicle was considered germination. After germination assays, nongerminated imbibed seeds were pressure tested, and seeds withstanding gentle pressure with forceps were determined viable as described by Borza et al. (2007).

All data were subjected to ANOVA and subsequent means separation using Fisher protected LSD test ($P \leq 0.05$). Data were analyzed by soybean trait using JMP Pro 15 (SAS Institute, Inc; Cary, NC). The nontreated plants were excluded from analysis of control and mortality data. Site year and replications were considered random effects to permit inferences to be made over a range of conditions (Blouin et al. 2011; Carmer et al. 1989). All seed production data were square-root transformed to improve normality. Back-transformed data are presented. Mortality rate (%) was calculated from counts of surviving weeds divided by the number of flagged weeds. Germination was summed across stratification periods for analysis. All germinated seed was considered viable and summed with seeds deemed viable from the imbibed pressure test for viability analysis.

Results and Discussion

Palmer Amaranth Control and Mortality

In Enlist soybean treatments, the three-way mixture of 2,4-D + glufosinate + glyphosate or the two-way mixture of 2,4-D + glufosinate resulted in the greatest control (88%) at 2 WAT (Table 3). Application of glufosinate + glyphosate or glufosinate alone resulted in similar control (approximately 73%). Results from application of 2,4-D alone and with glyphosate (approximately 30% control) were slightly better than from glyphosate alone (17%). Similar results were seen in Xtend soybean, where the three-way mixture of dicamba + glyphosate + glufosinate (85%) and the two-way mixture of dicamba + glufosinate (78%) performed best. Dicamba + glyphosate and dicamba alone again performed similarly (31% to 38%) and glyphosate alone performed worst (11%). The poor performance of glyphosate alone was expected because of the level of glyphosate resistance in the weed population. Overall, auxins alone resulted in less control than glufosinate alone, a result that also was expected because auxin herbicides must be translocated, and full efficacy is not typically visible 2 WAT.

Control 4 WAT was generally greater than at the 2 WAT rating timing, but similar trends were observed (Table 3). In Enlist soybean treatments, 2,4-D + glufosinate + glyphosate or 2,4-D + glufosinate resulted in the greatest control (94% to 95%), although the control with these treatments was not significantly different than the control achieved with glufosinate alone (86%) or glufosinate + glyphosate (88%). Results from the 2,4-D + glyphosate treatment (78%) were better than with 2,4-D alone (62%). Lawrence et al. (2018) reported similar results: 2,4-D + glufosinate

Table 3. Palmer amaranth visible control and end-of-season mortality rate in field experiments in Blackstone and Blacksburg, VA, in 2019[a].

Soybean trait	Herbicide	Rate	Control 2 WAT[b]	Control 4 WAT	Mortality Rate
		g ai or ae ha^{-1}	——%——		
Enlist®[c]					
	Glyphosate	1,260	17 d	16 d	28 d
	2,4-D	1,060	30 c	62 c	53 c
	2,4-D + glyphosate	1,060 + 1,260	33 c	78 b	68 bc
	Glufosinate	656	73 b	86 ab	80 ab
	Glufosinate + glyphosate	656 + 1,260	74 b	88 ab	88 a
	2,4-D + glufosinate	1,060 + 656	88 a	95 a	93 a
	2,4-D + glufosinate + glyphosate	1,060 + 656 + 1,260	88 a	94 a	95 a
Xtend®[c]					
	Glyphosate	1,260	11 C	9 C	19 B
	Dicamba	560	31 B	72 B	79 A
	Dicamba + glyphosate	560 + 1,260	38 B	87 A	73 A
	Dicamba + glufosinate	560 + 656	78 A	93 A	79 A
	Dicamba + glufosinate + glyphosate	560 + 656 + 1,260	85 A	94 A	75 A

[a]Means within a column followed by the same case letter are not significantly different. Data were analyzed by soybean trait used.
[b]Abbreviation: WAT, wk after treatment.
[c]Data were analyzed separately by soybean trait. Varieties used were Enlist E3 '5219E' and Xtend 'AG51X8'.

with and without glyphosate resulted in the most Palmer amaranth control, although their studies targeted 5- to 10-cm tall weeds.

Glyphosate alone resulted in similar control as the 2 WAT rating (16%). Trends similar to those reported with the Enlist soybean were seen in the Xtend soybean 4 WAT, with dicamba + glufosinate + glyphosate (94%), dicamba + glufosinate (93%), or dicamba + glyphosate (87%) performing best. It is interesting to note that dicamba + glyphosate (87%) performed in the top statistical grouping and resulted in significantly better control than dicamba alone (72%). Results with the auxin + glyphosate combination were better than for the auxin alone in both the Enlist and the Xtend systems, possibly because of the adjuvant loading of the glyphosate used (Roundup PowerMax). Glyphosate resulted in only 9% control in the Xtend soybean, similar to the low level of control seen in the Enlist soybean. Glyphosate overall resulted in poor control and provided no additive benefit to glufosinate-containing mixes. Interestingly, an additive benefit was seen in both systems when comparing an auxin herbicide alone to an auxin herbicide + glyphosate. Jha and Norsworthy (2012) found similar results, with dicamba, 2,4-D, or glufosinate being the most effective for controlling one glyphosate-resistant Palmer amaranth biotype (52% to 74%). They reported greatest control from dicamba or 2,4-D (40% to 47%) in a different glyphosate-resistant biotype in their study and low control from glufosinate and glyphosate (14% to 18%) (Jha and Norsworthy 2012).

End-of-season mortality data again revealed three-way mixes of auxin + glufosinate + glyphosate and auxin + glufosinate yielded best control results along with other treatments in the top statistical grouping, depending on soybean trait (Table 3). In Enlist soybean, 2,4-D + glufosinate + glyphosate (95%), 2,4-D + glufosinate (93%), glufosinate + glyphosate (88%), or glufosinate alone (80%) resulted in the greatest mortality rate. Glyphosate + 2,4-D (68%) and 2,4-D alone (53%) resulted in similar control, which was greater than results with glyphosate alone (28%). In the Xtend soybean, dicamba + glufosinate + glyphosate (75%), dicamba + glufosinate (79%), dicamba + glyphosate (73%), or dicamba alone (79%) all resulted in the greatest mortality rates, which were more than that with glyphosate alone (19%).

These results suggest there is no benefit to controlling Palmer amaranth at the reproductive stages by applying glyphosate in tank mixtures with glufosinate or dicamba, but there was a benefit with 2,4-D. The consistently best control and mortality rates across rating timings was seen with 2,4-D + glufosinate combinations in Enlist soybean and dicamba + glyphosate or glufosinate in Xtend soybean.

Palmer Amaranth Biomass

Palmer amaranth biomass at soybean harvest averaged 45 g plant^{-1} in the Enlist soybean nontreated plants and was reduced by all Enlist soybean treatments except glyphosate (Table 4). Glyphosate reduced biomass 36%, to 29 g plant^{-1}, and was not significantly different from the nontreated control, 2,4-D (22 g plant^{-1}; 51% reduction), 2,4-D + glyphosate (12 g plant^{-1}; 73% reduction), or glufosinate (10 g plant^{-1}; 78% reduction). Glufosinate + glyphosate (7 g plant^{-1}; 84% reduction), 2,4-D + glufosinate (7 g plant^{-1}; 84% reduction), or 2,4-D + glufosinate + glyphosate (6 g plant^{-1}; 87% reduction) all resulted in less biomass than the nontreated control and glyphosate alone, but results were not significantly different than with 2,4-D alone, 2,4-D + glyphosate, or glufosinate alone. Treatment with dicamba + glyphosate (14 g plant^{-1}; 48% reduction) resulted in less biomass than the nontreated control (27 g plant^{-1}), but that was not significantly different from all other treatments. Dicamba + glufosinate (12 g plant^{-1}; 56% reduction) or dicamba + glufosinate + glyphosate (11 g plant^{-1}; 59% reduction) treatment resulted in the greatest decrease in biomass compared with the nontreated control, but these decreases were not significantly different than decreases recorded for other treatments besides glyphosate.

These data indicate that, overall, auxins or glufosinate treatment result in significant decreases in Palmer amaranth biomass when applied at first female inflorescence. Glyphosate alone did not significantly reduce biomass in either soybean system and no additive effect was seen when combining glyphosate with glufosinate alone or an auxin + glufosinate mixture, which is to be expected of a glyphosate-resistant population. Previous research into glyphosate + auxin tank mixes applied to flowering common ragweed revealed similar results: glyphosate treatment alone did not reduce biomass and the addition of glyphosate to 2,4-D or dicamba did not improve biomass reduction (Bae et al. 2017). Bae et al. (2017) only found biomass reductions after treatments with 2,4-D and 2,4-D plus glyphosate. In their research, biomass

Table 4. Palmer amaranth biomass and seed production in field experiments in Blackstone and Blacksburg, VA in 2019[a].

Soybean trait	Herbicide	Rate	Biomass	Seed production	Cumulative germination[b]	Viability[c]
		g ai or ae ha^{-1}	g plant^{-1}	seeds plant^{-1}	—————%—————	
Enlist®[d]						
	Nontreated	Not applicable	45 c	8,260 c	47	96
	Glyphosate	1,260	29 bc	2,810 b	52	94
	2,4-D	1,060	22 ab	393 a	52	91
	2,4-D + glyphosate	1,060 + 1,260	12 ab	243 a	74	97
	Glufosinate	656	10 ab	208 a	44	96
	Glufosinate + glyphosate	656 + 1,260	7 a	38 a	52	92
	2,4-D + glufosinate	1,060 + 656	7 a	40 a	29	62
	2,4-D + glufosinate + glyphosate	1,060 + 656 + 1,260	6 a	21 a	27	94
Xtend®[d]						
	Nontreated	Not applicable	27 C	4,537 B	40	92
	Glyphosate	1,260	24 BC	2,380 B	54	94
	Dicamba	560	20 ABC	75 A	73	96
	Dicamba + glyphosate	560 + 1,260	14 AB	101 A	50	96
	Dicamba + glufosinate	560 + 656	12 A	16 A	37	98
	Dicamba + glufosinate + glyphosate	560 + 656 + 1,260	11 A	96 A	50	96

[a]Means within a column followed by the same case letter are not significantly different. Data were analyzed by soybean trait used.
[b]Germination data were not significant ($P = 0.189$ and 0.401 for Enlist and Xtend, respectively).
[c]Seed viability data were not significant ($P = 0.227$ and 0.871 for Enlist and Xtend, respectively).
[d]Data were analyzed separately by soybean trait. Varieties used were Enlist E3 '5219E' and Xtend 'AG51X8'.

reductions with dicamba or dicamba + glyphosate were not different from that of the nontreated, the glyphosate alone, or from the 2,4-D–containing treatments.

Palmer Amaranth Seed Production

All treatments in the Enlist soybean reduced seed production relative to the nontreated control, which produced 8,260 seeds plant^{-1} (Table 4). Glyphosate alone reduced seed production 66%, to 2,810 seeds plant^{-1}.Treatments with 2,4-D (393 seeds plant^{-1}; 95% reduction), 2,4-D + glyphosate (243 seeds plant^{-1}; 97% reduction), glufosinate (208 seeds plant^{-1}; 97% reduction), glufosinate + glyphosate (38 seeds plant^{-1}; >99% reduction), 2,4-D + glufosinate (40 seeds plant^{-1}; >99% reduction), or 2,4-D + glufosinate + glyphosate (21 seeds plant^{-1}; >99% reduction) all resulted in significantly less seed production than treatments with glyphosate alone or in the nontreated control.

In the Xtend soybean, all treatments besides glyphosate (2,380 seeds plant^{-1}) resulted in less seed production than the nontreated control (4,537 seeds plant^{-1}). Seed production of the remaining treatments ranged from 16 seeds plant^{-1} (dicamba + glufosinate) to 101 seeds plant^{-1} (dicamba + glyphosate), with total reductions relative to the nontreated control being greater than 98%. Jha and Norsworthy (2012) found similar results, with glyphosate reducing seed production 62% to 81% and dicamba, 2,4-D, or glufosinate resulting in greater reductions of 75% to 95%. No additional effect was found for the combination of glufosinate and an auxin herbicide or the inclusion of glyphosate with dicamba, 2,4-D, or glufosinate. Glyphosate in Xtend soybean resulted in a similar number of seeds produced (2,380 seeds plant^{-1}) versus the nontreated, possibly due to less overall seed production of the nontreated in Xtend soybean (4,537 seeds plant^{-1}) due to variability in the data. Overall, seed production data closely followed biomass production data and 2,4-D, dicamba, or glufosinate were effective at consistently reducing seed production.

Soybean Yield

Soybean yield averaged 1.80 Mg ha^{-1} in the Enlist soybean and 2.04 Mg ha^{-1} in the Xtend soybean. There were no differences among treatments with respect to yield. The Xtend soybeans were taller (91 cm) at application, compared with the Enlist soybeans (71 cm). The taller height at herbicide application and greater yield of the Xtend soybean variety indicate greater competitiveness with Palmer amaranth than do the results of the Enlist soybean variety between experiments. This difference explains the lower Palmer amaranth biomass and seed production between experiments.

Palmer Amaranth Seed Viability

The weight of a 100 seed sample averaged 0.040 g in the Enlist soybean and 0.031 g in the Xtend soybean, although there were no differences among treatments. Palmer amaranth germination ranged from 27% to 74% in Enlist soybean and 37% to 73% in Xtend soybean, although no differences were detected among treatments ($P \geq 0.189$) (Table 4). Palmer amaranth seed viability included the number of seeds germinated plus the number deemed viable from the imbibed pressure test.

Palmer amaranth viability ranged from 62% to 97% in the Enlist system and 92% to 98% in the Xtend system, but no differences were found among treatments ($P \geq 0.227$). These results differ from those reported by Jha and Norsworthy (2012), who saw reductions in cumulative emergence from glyphosate, glufosinate, 2,4-D, or dicamba (14%, 3%, 23%, and 22% of nontreated plants, respectively). Jha and Norsworthy (2012) also observed differences in seed viability. In their research, plants treated with glyphosate, glufosinate, 2,4-D, or dicamba resulted in 46% to 61% viability. But similar to our results, where viability of 96% and 92% was observed in the nontreated control in Enlist and Xtend experiments, respectively (Table 4), Jha and Norsworthy (2012) reported 97% viability in the nontreated control. Findings in our experiments may differ due to a slightly different application timing. Jha and Norsworthy (2012) applied treatments at first visible inflorescence rather than waiting until male and female plants could be distinguished, which, in our experience, could result in a 7- to 14-d difference in application timing. Therefore, more research is needed to further refine optimal application timing.

Results may have also differed because inflorescences were bagged in other experiments. If a treatment caused early shatter of seeds that were less viable, those seeds would not be captured in our experiments. However, weed-seed shattering timing in relation to herbicide application has not been examined in the literature, to our knowledge. In addition, our cleaning method for Palmer amaranth also removed small, immature seed. If such seed were included in germination assays and pressure tests, cumulative germination and viability may have been less. These results suggest that Palmer amaranth exposed to auxins, glufosinate, or glyphosate at first visible female inflorescence may still produce viable seed. More research is needed to evaluate the potential for these seed to possess or develop herbicide-resistant traits from these applications.

Palmer Amaranth Management Implications

Our findings suggest that auxin or glufosinate herbicides applied alone and in combination at first visible female inflorescence may be a viable option to reduce the amount of viable seed reaching the soil from uncontrolled or emerging Palmer amaranth. Hand removing weeds can also reduce additions to the soil seedbank, but adoption of this method is low and can cost as high as \$371 ha^{-1} (Riar et al. 2013). Harvest weed-seed control (HWSC) provides another opportunity to manage weed seeds at harvest, although challenges to adoption exist. The use of herbicides to manage these late-season weeds in crop is known as "crop-topping" in Australia and is primarily accomplished with nonselective herbicides (Walsh and Powles 2009). The use of these nonselective herbicides in a growing (not herbicide-resistant) crop often affected yield but reduced seed production of wild radish (*Raphanus raphanistrum* L.) 80% to 95% using either glyphosate or paraquat + diquat (Walsh and Powles 2009). With soybean herbicide–-resistant trait technologies such as XtendFlex and Enlist soybean, similar reductions in weed-seed production can be seen without the detriment to crop yields if these applications can be made in accordance with product labels. In addition, this method of reducing soil seedbank contributions can be accomplished with equipment a grower already uses, without the costs and complexity of hand weeding or adopting HWSC. Future research should evaluate the impacts of these herbicide applications on herbicide-resistance management and the risks associated with herbicide-resistance development from seed that remains viable after these applications.

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