

Influence of glufosinate mixtures on waterhemp control and soybean canopy and yield

Nikola Arsenijevic¹ , Mark L. Bernards², Ryan P. DeWerff³, Nicholas J. Arneson⁴, Daniel H. Smith⁵ and Rodrigo Werle⁶ 

Research Article

Cite this article: Arsenijevic N, Bernards ML, DeWerff RP, Arneson NJ, Smith DH, Werle R (2025) Influence of glufosinate mixtures on waterhemp control and soybean canopy and yield. *Weed Technol.* **39**(e66), 1–10. doi: [10.1017/wet.2025.37](https://doi.org/10.1017/wet.2025.37)

Received: 22 December 2024

Revised: 7 April 2025

Accepted: 13 April 2025

Nomenclature:

Bentazon; glufosinate; flumiclorac-pentyl; fluthiacet-methyl; fomesafen; waterhemp; *Amaranthus tuberculatus* (Moq.) Sauer; soybean, *Glycine max* (L.) Merr.

Keywords:

Fomesafen; lactofen; PPO inhibitors; soybean; mixtures

Corresponding author:

Rodrigo Werle; Email: rwerle@wisc.edu

¹Graduate Research Assistant, Department of Plant and Agroecosystem Sciences, University of Wisconsin-Madison, Madison, WI, USA; ²Research Agronomist, Agricultural Research Service, U.S. Department of Agriculture, Morris, MN, USA; ³Research Specialist, Department of Plant and Agroecosystem Sciences, University of Wisconsin-Madison, Madison, WI, USA; ⁴Former Outreach Specialist, Department of Agronomy, University of Wisconsin-Madison, Madison, WI, USA; ⁵Southwest Outreach Specialist, Nutrient and Pest Management Program, University of Wisconsin-Madison, Madison, WI, USA and ⁶Associate Professor, Department of Plant and Agroecosystem Sciences, University of Wisconsin-Madison, Madison, WI, USA

Abstract

Glufosinate serves as both a primary herbicide option and a complement to glyphosate and other postemergence herbicides for managing herbicide-resistant weed species. Enhancing broadleaf weed control with glufosinate through effective mixtures may mitigate further herbicide resistance evolution in soybean and other glufosinate-resistant cropping systems. Two field experiments were conducted in 2020 and 2021 at four locations in Wisconsin (Arlington, Brooklyn, Janesville, and Lancaster) and one in Illinois (Macomb) to evaluate the effects of postemergence-applied glufosinate mixed with inhibitors of protoporphyrinogen oxidase (PPO) (flumiclorac-pentyl, fluthiacet-methyl, fomesafen, and lactofen; Group 14 herbicides), bentazon (a Group 6 herbicide), and 2,4-D (a Group 4 herbicide) on waterhemp control, soybean phytotoxicity, and yield. The experiments were established in a randomized, complete block design with four replications. The first experiment focused on soybean phytotoxicity 14 d after treatment (DAT) and yield in the absence of weed competition. All treatments received a preemergence herbicide, with postemergence herbicide applications occurring between the V3 and V6 soybean growth stages, depending on the site-year. The second experiment evaluated the effect of herbicide treatments on waterhemp control 14 DAT and on soybean yield. Lactofen, applied alone or with glufosinate, produced the greatest phytotoxicity to soybean at 14 DAT, but this injury did not translate into yield loss. Mixing glufosinate with 2,4-D, bentazon, and PPO-inhibitor herbicides did not increase waterhemp control, nor did it affect soybean yield compared to when glufosinate was applied alone, but it may be an effective practice to reduce selection pressure for glufosinate-resistant waterhemp.

Introduction

Waterhemp is one of the most common and troublesome weed species in corn and soybean production systems throughout the midwestern United States (Tranel et al. 2011; Van Wychen 2022, 2023). Waterhemp has evolved resistance to herbicides from seven different sites of action (SOAs) (Heap 2024). A population of waterhemp from Missouri demonstrated resistance to herbicides from six SOAs, limiting effective postemergence control options to only glufosinate and dicamba (Shergil 2018). Similarly, a comprehensive herbicide resistance screening of more than 80 waterhemp accessions from Wisconsin revealed glufosinate as the only herbicide providing complete control (>97% biomass reduction) of all accessions (Faleco et al. 2022). Glufosinate is a broad-spectrum, nonselective, light-dependent herbicide with limited translocation that targets glutamine synthetase and is primarily effective on annual weed species (Dayan et al. 2019; Steckel et al. 1997). However, its performance can vary in the field due to factors such as low humidity and temperature, time of day, and weed size (Coetzer et al. 2001; Kumaratilake and Preston 2005; Martinson et al. 2005; Tharp et al. 1999). Glufosinate-resistant crops were rarely adopted before glyphosate-resistant weeds became widespread in glyphosate-based systems, even though both technologies were commercialized around the same time, and delayed adoption was likely due to glufosinate's historically lower efficacy and consistency compared to glyphosate, as well as the limited availability of glufosinate-resistant soybean cultivars until 2020 (Takano and Dayan 2020). However, with the rising prevalence of multiple herbicide-resistant weeds, glufosinate's role in weed management is now expanding (Takano and Dayan 2020; USGS 2018). Currently six instances of glufosinate resistance have been reported, with one of the six weeds being a broadleaf species, Palmer amaranth (*Amaranthus palmeri*) (Heap 2024). Glufosinate should be used strategically to postpone further resistance evolution and to preserve it as a tool for effective broadleaf control.

© The Author(s), 2025. Published by Cambridge University Press on behalf of Weed Science Society of America. This is an Open Access article, distributed under the terms of the Creative Commons Attribution licence (<https://creativecommons.org/licenses/by/4.0/>), which permits unrestricted re-use, distribution and reproduction, provided the original article is properly cited.



Compelling evidence indicates that the rapid cell death in glufosinate-treated plants is mainly due to reactive oxygen species (ROS), which when produced in large quantities under light, cause severe lipid peroxidation of cell membranes leading to rapid phytotoxicity (Takano et al. 2019, 2020a). Herbicides that target protoporphyrinogen oxidase (PPO) lead to an accumulation of protoporphyrin IX, a compound that also produces ROS when exposed to light (Dayan et al. 2019). Combinations of glufosinate and PPO-inhibitor herbicides may be more advantageous in terms of weed control when compared to individual applications of these herbicides, because of the simultaneous inhibition of glutamine synthetase and PPO, leading to elevated accumulation of protoporphyrin IX and the concomitant accumulation of ROS (Takano et al. 2020a). Mixtures may also alleviate environmental effects on glufosinate performance (Takano et al. 2020b). Takano et al. (2020b) reported a synergistic effect in controlling Palmer amaranth and kochia [*Bassia scoparia* (L.) A.J. Scott] when a half rate of glufosinate (280 g ha⁻¹) was mixed with an extremely low dose of saflufenacil (1 g ha⁻¹). However, the utility of this mixture for postemergence weed control is limited because it caused >60% injury to both susceptible and glufosinate-resistant soybean and did not increase control of PPO inhibitor-resistant waterhemp. The strong synergistic effect initially observed on Palmer amaranth varied based on weed species treated, herbicide dosages, and PPO inhibitors tested (Takano et al. 2020b). For example, when flumioxazin, pyraflufen, lactofen, or fomesafen were mixed with glufosinate and applied to kochia, the synergistic effect was less than what was observed with saflufenacil (Takano et al. 2020b). The elevated soybean injury observed following postemergence applications of glufosinate + saflufenacil mixtures may portend increased soybean injury with mixtures of glufosinate with other PPO-inhibitor herbicides (Belfry et al. 2016; Takano et al. 2020b) and slow the development of canopy formation (Priess et al. 2020). This may discourage use of PPO-inhibitor chemistry when it may otherwise be a valuable part of an herbicide-resistance mitigation strategy.

Another potential glufosinate mix partner is 2,4-D (categorized as a Group 4 herbicide by the Weed Science Society of America [WSSA]). Craigmyle et al. (2013) indicated that addition of 2,4-D to either or both postemergence applications of glufosinate provided better waterhemp control compared to two postemergence applications of glufosinate alone. Furthermore, Joseph et al. (2018) reported an increased spectrum in control of sicklepod [*Senna obtusifolia* (L.) H.S. Irwin & Barneby], pitted morningglory [*Ipomoea lacunosa* L.], and Palmer amaranth when glufosinate was mixed with either 2,4-D or dicamba, compared to herbicides applied alone. Lanclous et al. (2002) reported a synergistic effect for control of spreading dayflower (*Commelina diffusa* Burm. f.) when glufosinate was mixed with propanil (WSSA Group 5, a photosystem II inhibitor), which also leads to accumulation of ROS. In contrast, acetyl-CoA carboxylase inhibitors and glyphosate have not always increased glufosinate control of some grass and broadleaf weed species (Besançon et al. 2018; Burke et al. 2005), warranting further investigation of the most effective partners with glufosinate to improve postemergence weed control in soybean production.

The proportion of herbicide-resistant weeds in the field will rapidly increase with repeated use of the same herbicide SOA (Beckie 2006). The strategic use of both preemergence and postemergence herbicide mixtures containing multiple effective SOAs is crucial to delaying herbicide resistance, preserving the effectiveness of new herbicide-resistant crops, and ensuring the long-term economic sustainability of agriculture (Norsworthy

et al. 2012). The combination of glufosinate with PPO inhibitors and other alternative herbicide SOAs (i.e., Group 4 or Group 6) is one research area that requires additional studies to understand their interactions and effect on weed control and crop injury (Takano et al. 2020b). Our objectives were to measure the efficacy of glufosinate applied alone and mixed with other active ingredients on 1) waterhemp control and 2) soybean injury and yield.

Materials and Methods

Two separate field experiments were conducted in Illinois and Wisconsin to investigate glufosinate combinations with various herbicides on soybean phytotoxicity and yield (hereafter referred to as the *crop response study*), and waterhemp control (hereafter referred to as the *waterhemp response study*). The crop response study was conducted in 2020 and 2021 in Macomb, IL (40.4900°N, 90.6888°W), and in 2020 and 2021 at the Arlington Agricultural Research Station in Arlington, WI (43.3034°N, 89.3455°W), and the Rock County Research farm in Janesville, WI (42.7262°N, 89.0235°W), in fields with a known history of low weed infestation and no waterhemp presence (R.P. DeWerff and M.L. Bernards personal observations). The waterhemp response study was conducted in 2021 at a site in Macomb, IL (40.4795°N, 90.7208°W), and in 2020 and 2021 at the Lancaster Agricultural Research Station in Lancaster, WI (42.8313°N, 90.7880°W), and the O'Brien Family Farm near Brooklyn, WI (42.8768°N, 89.3980°W), in fields that were naturally infested with waterhemp. Experiments were established in a randomized complete block design with four replications, using experimental units that measured 3 m wide by 9.1 m long with four soybean rows planted 76 cm apart. Both studies included a preemergence herbicide–nontreated control (receiving only postemergence herbicides), while only the waterhemp response study contained a complete nontreated control (no preemergence or postemergence herbicides). In contrast, the whole-crop response study was maintained weed-free throughout the season. A more effective preemergence herbicide combination, flumioxazin + pyroxasulfone (70.4 and 89.3 g ai ha⁻¹, respectively [Fierce; Nufarm, Morrisville, NC]), was applied at soybean planting for the crop response study to aid in weed-free maintenance during the growing season, such that any measured effects on soybean development and yield resulted solely from the effect of a postemergence herbicide treatments. In the waterhemp response study, a preemergence application of flumioxazin alone (112 g ai ha⁻¹, Valor; Valent, San Ramon, CA) was made to all treatments at soybean planting, except for the nontreated control. The postemergence herbicide treatments were identical across both studies (Table 1). postemergence herbicide treatments were applied using a CO₂-pressurized backpack sprayer, equipped with AIXR11015 spray nozzles (TeeJet Technologies, Glendale Heights, IL) on a 2.54-m-wide spray boom, calibrated to deliver 140 L ha⁻¹ of carrier volume. Weather information for the soybean growing season at each location is presented in Table 2. Soil characteristics, soybean variety and planting dates, and soybean growth and waterhemp density and height at post-emergence herbicide application for all experimental locations are displayed in Table 3.

Soybean Phytotoxicity and Soybean Green Cover

A visual evaluation of soybean phytotoxicity in the crop response study was made 14 DAT on a scale from 0% to 100%, where 0% represented no injury and 100% represented plant death. The most

Table 1. Postemergence herbicide treatments used in both field experiments, along with herbicide group numbers, active ingredients, and their application rates.^{a-c}

Herbicide	Trade name	Manufacturer	WSSA group number	Application rate g ai ha ⁻¹
Glufosinate	Liberty 280 SL [®]	BASF	10	657
2,4-D	Enlist One [®]	Corteva	4	1,067
Bentazon	Basagran 4L [®]	BASF	6	897
Flumiclorac-pentyl	Resource [®]	Valent	14	60
Fluthiacet-methyl	Cadet [®]	FMC	14	7.2
Fomesafen	Flexstar [®]	Syngenta	14	264
Lactofen	Cobra [®]	Valent	14	219
Glufosinate + 2,4-D			10 + 4	657 + 1,067
Glufosinate + bentazon			10 + 6	657 + 897
Glufosinate + flumiclorac-pentyl			10 + 14	657 + 60
Glufosinate + fluthiacet-methyl			10 + 14	657 + 7.2
Glufosinate + fomesafen			10 + 14	657 + 264
Glufosinate + lactofen			10 + 14	657 + 219
No PRE (nontreated control)				

^aAbbreviations: POST, postemergence; PRE, preemergence; WSSA, Weed Science Society of America.

^bHerbicides in WSSA Group 14 (protoporphyrinogen inhibitor) and Group 6 (photosystem II inhibitor) applied solely were combined with a crop oil concentrate (10 mL L⁻¹; CHS Agronomy Inc., Willmar, MN) as a surfactant, while mixes with glufosinate excluded a crop oil concentrate. Ammonium-sulfate (2,243 g ha⁻¹) was added to all herbicide treatments.

^cBoth studies included a nontreated control (No PRE). However, only the waterhemp response study had a true weedy nontreated control (No PRE nor a POST herbicide application). In contrast, the whole-crop response study was maintained weed-free throughout the season.

common symptoms observed were necrosis (bronzing) and stunting of soybean growth. A digital estimation of soybean canopy development was conducted to estimate soybean green cover percentage, also at 14 DAT. Three photographs, each capturing approximately 1.7 m of row of both the second and third row, were taken in each plot. A wooden L-shaped pole measuring 1.93 m in height was used to support a GoPro Hero 8 Black camera (GoPro Inc., San Mateo, CA) above soybean canopy, which was paired with an iPhone 6s (Apple Inc., Cupertino, CA) via the GoPro Quik app and used as an electronic viewfinder for the camera. Resolution of the images captured with GoPro 8 Hero Black camera was 4,000 × 3,000 pixels (aspect ratio 4:3), with linear distortion setting. The images were processed using the Canopeo add-on (Canopeo software [<https://canopeoapp.com/>]) was developed by staff and researchers in Oklahoma State University's Division of Agricultural Sciences and Natural Resources Soil Physics Program) with MATLAB software (MathWorks, Natick, MA). This allowed for the estimation of fractional soybean green cover within each image and served as a proxy of herbicide-induced crop injury, where a higher green cover percentage indicated lower soybean injury (Arsenijevic et al. 2021; Liang et al. 2012; Paruelo et al. 2000; Patrignani and Ochsner 2015).

Visual Assessment of Waterhemp Control and Biomass Collection

In the waterhemp control study a visual estimate of waterhemp control was made 14 DAT, using a scale ranging from 0% to 100%, where 0% represented no control, and 100% represented complete control of all waterhemp. Waterhemp biomass was collected at 14 DAT by harvesting all waterhemp plants within two 0.25-m² quadrats in each plot. Harvested plants were dried to a constant weight at 60 C, and waterhemp biomass reduction was compared with that of the nontreated control was calculated using Equation 1:

$$R = 100 - \left(\frac{H}{C} * 100 \right) \quad [1]$$

where biomass reduction (R) was estimated by comparing the dry biomass of a treated plot (H) to the average dry biomass of the nontreated control (C).

Soybean Yield

At crop maturity, the center two rows of each experimental plot were mechanically harvested using a plot combine for both studies. The soybean yield data obtained were adjusted to 13% moisture content and are presented in kilograms per hectare (kg ha⁻¹).

Statistical Analyses

All response variables (waterhemp response study: visual assessment of waterhemp control [%], waterhemp biomass reduction [%], soybean yield [kg ha⁻¹]; crop response study: soybean phytotoxicity [%], soybean green cover [%], and soybean yield [kg ha⁻¹]) were analyzed using R Statistical Software (v. 4.4.1; R Core Team 2021). Data were pooled across site-years (year and location were treated as random factors). Herbicide treatment was the main effect, and replications nested within site-years were treated as random effects.

A generalized linear mixed model with Template Model Builder with beta distribution and logit link (GLMMTMB package, v. 1.1.9) (Brooks et al, 2017) was fit to soybean injury, soybean green cover percentage, visual assessment of waterhemp control, and waterhemp biomass reduction. A Pearson chi-square test (using the NORTTEST package, v. 1.0-4) and Levene's test (with the CAR package, v. 3.1-2) were used to check normality and homogeneity of variance, respectively. Response variables were logit-transformed to improve normality assumptions (Barnes et al. 2020; Davies et al. 2019; Striegel et al. 2020). The analysis of variance type II Wald chi-square test was performed followed by Tukey's honestly significant difference (HSD) test ($\alpha = 0.05$) and pairwise comparisons using the EMMEANS package (v. 1.10.3). Back transformed means are presented for ease of result interpretation.

A linear mixed model with a normal distribution using the LME4 package (v. 1.1-35.5) was fit to soybean yield data. To better meet the normality and variance homogeneity assumptions, response variables were square root-transformed. When ANOVA results indicated a significant herbicide effect, means were compared using Tukey's HSD test ($\alpha = 0.05$). Means were separated when herbicide treatment effect was less than $P = 0.05$ using Tukey's HSD test. Back-transformed means are presented for ease of interpretation.

Table 2. Monthly average air temperature and precipitation for experimental sites in 2020 and 2021 growing seasons.^{a,b}

	Location														
	Arlington			Brooklyn			Janesville			Lancaster			Macomb		
	2020	2021	30 yr	2020	2021	30 yr	2020	2021	30 yr	2020	2021	30 yr	2020	2021	30 yr
	Air temperature C														
May	12.9	13.5	14	13.6	14.4	14	13.9	14.8	16	11.1	12.8	15	16.1	15.5	17
June	20.1	21.4	20	21.3	22.5	20	21.3	22.8	21	18.7	20.2	21	23.3	22.7	22
July	22.2	20.6	23	23.5	21.8	23	24.1	22.1	24	21.3	20.1	23	25.5	23.8	25
August	14.3	20.9	22	21.4	21.9	22	21.8	22.6	23	18.7	19.0	22	22.7	24.2	24
September	14.3	16.4	18	15.7	18.1	18	15.7	18.6	19	12.8	14.8	18	18.3	21.8	20
Season ^c	16.8	18.5	19.4	19.1	19.7	19.4	19.4	20.2	20.6	16.5	17.4	19.8	21.0	21.6	21.6
	Precipitation mm														
May	113	66	89	119	60	91	107	74	94	139	72	91	126	185	102
June	110	96	104	111	133	107	82	55	107	198	43	109	161	134	114
July	142	38	97	118	76	102	148	53	102	131	121	104	129	43	107
August	97	90	102	20	63	104	79	79	104	94	132	107	12	59	109
September	76	59	91	122	31	94	87	18	97	187	50	94	37	45	99
Season ^c	538	349	483	490	363	498	503	279	504	749	418	505	465	466	531

^aAir, soil, and rainfall data were collected with WatchDog 2000 Series ground weather stations from an Enviro-weather station.

^bThirty-year air temperature and precipitation averages for the period 1991 to 2021 were obtained with R statistical software (v. 4.4.1) using daily Daymet weather data for 1-km grids (Correndo et al. 2021; Thornton et al. 2016; *daymetr* package).

^cCumulative precipitation and average monthly temperature throughout the growing season.

Table 3. Information for each experimental location covering soybean variety and its planting date, herbicide application dates, herbicide application dates, soybean growth stages, the height and density of waterhemp, and soil information.^{a,b}

	Waterhemp response study					Crop response study					
	Brooklyn		Lancaster		Macomb	Arlington		Janesville		Macomb	
	2020	2021	2020	2021	2021	2020	2021	2020	2021	2020	2021
Planting date	May 22	May 25	May 20	May 17	June 5	May 1	May 12	May 8	April 29	May 25	May 24
PRE herbicide application	May 22	May 26	May 20	May 19	June 6	May 1	May 12	May 8	April 29	May 29	May 26
POST herbicide application	June 24 (V4)	June 30 (V5)	July 1 (V6)	June 17 (V6)	July 14 (V4)	June 25 (V4)	June 26 (V4)	July 2 (V4)	June 18 (V4)	June 29 (V4)	July 2 (V5)
Waterhemp height at POST ^c	2–20	2–22	7–28	4–13	2–20						
Waterhemp density at POST ^d	16–33	12–40	18–34	1–13	12–36						
	Soil information										
% Sand	40	40	10	10	11	8	4	7	8	3	3
% Silt	41	41	76	76	79	56	71	70	66	76	72
% Clay	19	19	14	18	10	36	25	23	26	21	25
% Organic matter	2	2	2.5	3.1	2.4	2.9	3.3	3.1	4.1	3.4	2.0
pH	7.1	7.1	6.6	5.3	7.5	6.5	6.4	6.4	6.7	6.8	6.4
Textural class	Loam	Loam	Silt loam	Silt loam	Keomah silt loam	Silty clay loam	Silt loam	Silt loam	Silt loam	Osco silt loam	Osco silt loam

^aAbbreviations: POST, postemergence; PRE, preemergence; V4, V5, and V6 refer to soybean growth stage.

^bSoybean P22T86E was planted in Wisconsin in 2020 and 2021. Syngenta S33E3 was planted in 2020, and NuTech 35N03E was planted in 2021 in Illinois.

^cWaterhemp height at the time of POST herbicide application is measured in centimeters and shown as a range.

^dWaterhemp density at the time of POST herbicide application is measured in square meters (m⁻²) and shown as a range.

Table 4. Soybean visible phytotoxicity and green cover (Canopeo) 14 d after treatment, and soybean final yield for crop response (weed-free) study.^{a-e}

Herbicide treatment	Visible phytotoxicity	Green cover	Soybean yield
	%		kg ha ⁻¹
PRE only	2 (0.1–2.5) a	78 (70–85) ab	4,359 (3,501–5,310) ab
Glufosinate	2 (1–4) a	78 (70–85) ab	4,576 (3,686–5,560) a
2,4-D	2 (1.0–3) a	81 (73–87) a	4,532 (3,647–5,513) ab
Bentazon	5 (3–6) c	75 (66–82) b	4,505 (3,623–5,483) ab
Flumiclorac-pentyl	18 (15–22) e	73 (64–81) b	4,453 (3,576–5,425) ab
Fluthiacet-methyl	14 (11–17) de	76 (67–83) ab	4,622 (3,728–5,612) a
Fomesafen	13 (10–17) d	75 (66–82) b	4,421 (3,547–5,390) ab
Lactofen	27 (23–32) f	59 (48–69) c	4,376 (3,517–5,329) ab
Glufosinate + 2,4-D	3 (2–4) abc	76 (68–83) ab	4,479 (3,599–5,453) ab
Glufosinate + bentazon	4 (3.0–6) bc	78 (69–84) ab	4,430 (3,556–5,400) ab
Glufosinate + flumiclorac-pentyl	18 (15–22) e	73 (63–80) b	4,455 (3,578–5,428) ab
Glufosinate + fluthiacet-methyl	13 (10–16) d	75 (65–82) b	4,395 (3,524–5,361) ab
Glufosinate + fomesafen	17 (14–20) de	75 (66–82) b	4,336 (3,471–5,295) ab
Glufosinate + lactofen	27 (23–32) f	60 (49–70) c	4,201 (3,350–5,146) b
No PRE (nontreated control)	1.5 (1–2) a	79 (70–85) ab	4,556 (3,669–5,539) ab
P-value	<0.0001	<0.0001	0.0174

^aAbbreviation: PRE, preemergence.

^bMeans with the same letters are not statistically different from each other according to Tukey's honestly significant difference test ($\alpha = 0.05$).

^cInformation presented in parentheses refers to 95% confidence intervals.

^dThe data presented in the table are from experimental locations in Wisconsin and Illinois during 2020 and 2021.

^eGreen cover refers to Canopeo data (see text).

To assess the relationship between soybean visual injury and soybean green cover (Canopeo data), a linear mixed-effects model was used (LME4 package). Soybean visual injury was the response variable, soybean green cover was the fixed effect, and replications were nested within site-years. The model was fit using maximum likelihood estimation. Predicted soybean visual injury values were calculated based on the fitted model. A simple linear regression was conducted and the predicted soybean visual injury was calculated. The goodness-of-fit of the models was assessed using the R-squared statistic (PIECEWISESEM package), which represents the proportion of variance in phytotoxicity that can be explained by the models (marginal and conditional R^2). The relationship between soybean visual injury and soybean green cover was calculated according to Equation 2:

$$V = \beta_0 + \beta_1 \times C + r + \varepsilon \quad [2]$$

where V = visual injury (dependent variable); β_0 = intercept; β_1 = slope for soybean green cover (independent variable); C = green cover; r = random effect of site-year nested within rep; ε = error term.

Results and Discussion

Crop Response Study

Soybean Visible Phytotoxicity and Soybean Green Cover

The main effect of herbicide treatment was significant for visual soybean phytotoxicity and green cover ($P < 0.05$). Greater visible phytotoxicity indicates more severe soybean herbicide injury, while greater green cover suggests less herbicide injury. Herbicide treatments that caused the greatest soybean injury (27%) were lactofen and glufosinate + lactofen (Table 4). All PPO-inhibitor herbicides and PPO inhibitor + glufosinate mixtures caused greater than 10% injury (Table 4). Glufosinate, 2,4-D, and bentazon caused less than 5% soybean injury (Table 4).

Soybean green cover was reduced 25% by lactofen and glufosinate + lactofen when compared with the nontreated

control (Table 4). Soybean is susceptible to injury from PPO inhibitors, particularly under hot and humid conditions following herbicide application (Sarangi and Jhala 2015; Whitaker et al. 2010). This injury could hinder the development of the soybean canopy (Nelson and Renner 2001). Differential soybean tolerance to some of the PPO-inhibitor herbicides has been reported as (least injurious to most injurious): fomesafen < acifluorfen < lactofen (Harris et al. 1991). The recovery of soybean from injury that delays canopy formation depends on factors such as planting date, soybean phenology, maturity group, growth habit, and soil moisture availability (Priess et al. 2020). However, even when these herbicides (fomesafen, acifluorfen, lactofen) were applied to soybean at several rates between growth stages V1 and V5 and caused up to 20% of foliar injury, there was no yield loss at the end of the season (Beam et al. 2018; Kapusta et al. 1986; Riley and Bradley 2014; Wichert and Talber 1993; Young et al. 2003).

Relationship Between Soybean Green Cover and Visible Soybean Injury

Our analysis revealed a negative correlation between soybean green cover (the Canopeo data) and visual injury (Figure 1). This negative correlation is intuitive; as visual injury increases soybean green cover decreases, which is reflected by the downward slope of the regression line. The marginal R^2 value was 0.51, indicating that soybean green cover alone accounted for approximately 51% of the observed variation in soybean visible injury. The remaining 26% of the variation (yielding a conditional R^2 value of 0.77) was attributed to differences across site-years.

Soybean Yield

The main effect of herbicide treatment was significant for soybean yield ($P < 0.05$; Table 4). However, no herbicide treatment was different when compared to the no-preemergence (nontreated) and preemergence-only treatments. When herbicides are applied within labeled rates early in the season, soybean injury is generally transitory with minimal impact on grain yield (Beam et al. 2018; Kapusta et al. 1986; Riley and Bradley 2014; Wichert and Talber 1993; Young et al. 2003). However, Priess et al. (2020) found that

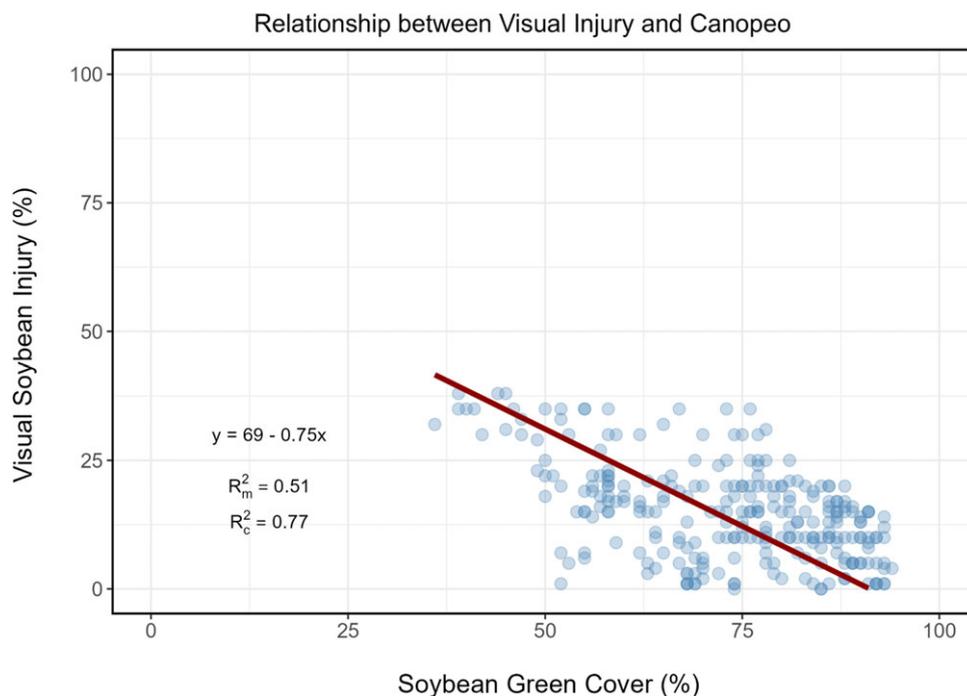


Figure 1. Relationship between visual soybean injury and soybean green cover (Canopeo data). R_m^2 signifies that site-year as a random effect is not considered (marginal); R_c^2 signifies that site-year as a random effect is considered (conditional).

soybean injured when herbicide was applied at the V2 stage exhibited slower canopy formation. Delaying application of injurious herbicides until near the flowering stage or when moisture availability limits canopy growth may have more lasting negative effects because grain yield is linked to the canopy present at the onset of reproductive development (Edwards and Purcell, 2005). PPO-inhibitor herbicides should be applied early enough to allow the crop to reach full canopy closure, which is crucial for end-of-season weed suppression and maximizing soybean yield (Arsenijevic et al. 2022; Edwards and Purcell 2005; Jha and Norsworthy 2009).

Waterhemp Response Study

Visual Assessment of Waterhemp Control and Dry Biomass at 14 DAT

The main effect of herbicide treatment was significant for visual assessment of waterhemp control and dry biomass reduction ($P < 0.05$; Table 5). All glufosinate mixtures provided $\geq 88\%$ control of waterhemp, equal to glufosinate applied solo (90%). In addition, 2,4-D, fomesafen, and lactofen applied alone provided $\geq 88\%$ control (Table 5). Flumiclorac-pentyl (73%) and fluthiacet-methyl (71%) applied individually showed limited activity on waterhemp and were similar to the preemergence-only flumioxazin treatment (60%). Bentazon applied alone (54%) showed the lowest control of waterhemp.

Waterhemp biomass reduction measurements generally paralleled the visual assessments of waterhemp control results (Table 5). Effective control was defined as herbicide treatments achieving an efficacy of $\geq 90\%$ (Arneson et al. 2020; Etheridge et al. 2001; Werle et al. 2023). Three treatments resulted in 91% waterhemp biomass reduction: glufosinate + fomesafen, glufosinate + lactofen, and glufosinate + bentazon. Glufosinate applied alone was the only single active ingredient treatment that resulted $\geq 90\%$ waterhemp biomass reduction. However, the only postemergence treatment to

provide less waterhemp biomass reduction than glufosinate applied alone was bentazon applied alone, which provided no biomass reduction (55%; Table 5), similar to the preemergence-only treatment (56%; Table 5).

Although glufosinate, 2,4-D, fomesafen, and lactofen applied individually resulted in high levels of waterhemp control in this study, repeated use of single SOA herbicides increases the risk of herbicide resistance evolution (Norsworthy et al. 2012). In bareground trials conducted in Wisconsin, Werle et al. (2023) reported that 2,4-D, dicamba, lactofen, and fomesafen applied alone provided variable waterhemp control (74% to 87%). The absence of crop competition in these systems likely contributed to the inability of any solo herbicide treatment to achieve the $\geq 90\%$ control threshold for an excellent rating in University of Wisconsin–Madison Extension guidelines (Arneson et al. 2020). These results highlight both the inherent limitations of bareground systems (lacking crop-weed competition) and the practical need for mixtures to achieve commercially acceptable waterhemp control in production fields.

Takano and Dayan (2020) reported that mixing glufosinate and PPO inhibitors enhanced the herbicidal activity, although other reports showed that the degree of enhancement varied depending on the weed species, herbicide dosage, and the PPO-inhibitor herbicide being evaluated (Takano et al. 2020b). However, in our experiment, the herbicide combinations did not increase waterhemp control compared to glufosinate alone (Table 5). We used labeled rates of both glufosinate and the mix partners, which is encouraged to reduce the risk of herbicide-resistance evolution (Norsworthy et al. 2012). Postemergence applications of glufosinate mixtures, specifically with PPO inhibitors in XtendFlex® (Bayer CropScience, St. Louis, MO) soybean or with 2,4-D in Enlist E3® (Corteva Agriscience, Indianapolis, IN) soybean, may provide an effective herbicide resistance management strategy when combined with effective preemergence herbicides. Furthermore, other glufosinate-resistant platforms such as LibertyLink® GT27

Table 5. Visible assessment of waterhemp control and waterhemp dry biomass reduction 14 d after treatment, and soybean final yield for the waterhemp response study.^{a-d}

Herbicide treatment	Waterhemp control	Biomass reduction	Soybean yield
	%		kg ha ⁻¹
PRE only	60 (44–74) bc	56 (41–70) cd	2,904 (2,325–3,548) c
Glufosinate	90 (81–95) a	90 (81–95) ab	3,669 (3,013–4,389) a
2,4-D	90 (81–95) a	87 (77–93) ab	3,568 (2,923–4,279) a
Bentazon	54 (38–68) c	55 (39–69) d	3,264 (2,647–3,944) abc
Flumiclorac-pentyl	73 (57–84) b	79 (66–88) ab	3,343 (2,719–4,031) abc
Fluthiacet-methyl	71 (55–82) bc	77 (64–87) bc	3,365 (2,738–4,055) abc
Fomesafen	88 (77–94) a	87 (78–94) ab	3,460 (2,826–4,158) ab
Lactofen	90 (81–95) a	87 (77–93) ab	2,914 (2,335–3,549) bc
Glufosinate + 2,4-D	92 (84–96) a	89 (80–94) ab	3,713 (3,053–4,437) a
Glufosinate + bentazon	90 (82–95) a	91 (83–95) a	3,661 (3,007–4,380) a
Glufosinate + flumiclorac-pentyl	88 (78–94) a	79 (81–95) ab	3,532 (2,890–4,239) a
Glufosinate + fluthiacet-methyl	89 (80–95) a	88 (79–94) ab	3,613 (2,963–4,327) a
Glufosinate + fomesafen	93 (86–97) a	91 (84–95) a	3,712 (3,052–4,436) a
Glufosinate + lactofen	93 (85–97) a	91 (84–95) a	3,469 (2,835–4,169) ab
No PRE (nontreated control)	0	0	2,221 (1,718–2,788) d
P-value	<0.0001	<0.0001	<0.0001

^aAbbreviation: PRE, preemergence.

^bMeans with the same letters are not statistically different from each other according to Tukey's honestly significant difference test ($\alpha = 0.05$).

^cInformation presented in parentheses refers to 95% confidence intervals.

^dThe data presented in the table are from experimental locations in Wisconsin during 2020 and 2021, and from experimental location in Illinois in 2021.

(MS TechnologiesTM, West Point, IA; BASF Corporation, Research Triangle Park, NC) soybean, confers additional tolerance to glyphosate and isoxaflutole, enabling preemergence isoxaflutole applications for enhanced waterhemp control (Craigmyle et al. 2013; Hay et al. 2019; Merchant et al. 2013; Smith et al. 2019). Annual rotation of herbicide SOAs and trait technologies provides optimal resistance mitigation.

Soybean Yield

The main effect of herbicide treatment was significant for soybean yield in the waterhemp response study ($P < 0.05$; Table 5). All herbicide treatments yielded more than the no-preemergence nontreated control (Table 5), with yield increases (yield-protection) of 31% to 67%. Postemergence-applied mixture treatments with glufosinate yielded 19% to 28% more than the preemergence-only check. Yield from plots treated individually with bentazon, flumiclorac-pentyl, and fluthiacet-methyl was not greater than preemergence-only plots (Table 5), presumably because competition from the surviving waterhemp was similar to the plot that received a preemergence-only application (Table 4). Both weed presence and herbicide injury may influence soybean yield. When glufosinate and 2,4-D were applied individually, yields were 26% and 23% greater, respectively, than yield from the preemergence-only treatment. However, when lactofen was applied alone, waterhemp control was equivalent to that of glufosinate and 2,4-D, but soybean yields were >18% lower (Table 5). In contrast, glufosinate + lactofen, which caused similar injury to lactofen applied alone (Table 4), did not reduce yield, and provided similar waterhemp control (Table 5). Fomesafen applied alone, which was less injurious to soybean than lactofen in the crop response study (Table 4), nor did it reduce yields in the waterhemp response study compared to glufosinate applications (Table 5). These data confirm that postemergence herbicide applications are critical to protect soybean yield, and that both weed control and crop safety may affect soybean yield.

Soybean yield loss from weeds is typically of greater importance than potential injury from herbicides (Young et al. 2003), and an

application of postemergence herbicides with multiple effective SOAs is likely beneficial to delaying the evolution of herbicide resistance (Norsworthy et al. 2012). Among the PPO inhibitor + glufosinate mixtures we tested, fomesafen presented an acceptable balance of crop safety and effective waterhemp control. Although fomesafen has been less injurious to soybean than lactofen, its weed control efficacy has not always exceeded 90% (Ellis and Griffin 2003; Hager et al. 2003; Harris et al. 1991; Higgins et al. 1988; Johnson et al. 2002). In our research, glufosinate + fomesafen provided 93% waterhemp control and reduced waterhemp biomass by 91%, while causing less crop injury (Table 4) and protecting yield potential (4,336 kg ha⁻¹ [crop response study], Table 4; 3,712 kg ha⁻¹ [waterhemp response study], Table 5). In addition, fomesafen can provide soil residual control of waterhemp for several weeks after its application (Oliveira et al. 2017).

Soybean growers, particularly those who cultivate glufosinate-resistant Enlist E3 varieties, may prefer using herbicide mixtures with 2,4-D to reduce crop injury and ensure adequate weed control, and 2,4-D has long been considered a low-risk herbicide for resistance evolution (Torra et al. 2024). However, resistance to 2,4-D is increasing in waterhemp populations across the Midwest (Bernards et al. 2012; Evans et al. 2019; Faleco et al. 2024; Heap 2024; Shergill et al. 2018). Resistance to 2,4-D by weeds is typically a single-gene trait and confers elevated 2,4-D detoxification using cytochrome P450 monooxygenases or glycosyltransferases (Torra et al. 2024). Weeds metabolize 2,4-D more rapidly at higher temperatures, which may be problematic when it is mixed with glufosinate because glufosinate performs best under high temperature and humidity conditions (Coetzer et al. 2001). While PPO inhibitor-resistant waterhemp populations (Heap 2024) with target-site mutations (Barker et al. 2023; Lillie et al. 2020; Shoup et al. 2003) may still show some susceptibility to soil-applied PPO inhibitors, the duration and level of control are typically reduced compared to populations that have been confirmed to be susceptible (Lillie et al., 2020). Agrichemical and seed companies are developing new soybean stacked traits that will alleviate injury caused by PPO-inhibitor herbicides, and new PPO-inhibitor

herbicides are being developed that are expected to provide improved weed control (Prade 2022).

It is crucial to preserve the efficacy of glufosinate, 2,4-D, and PPO-inhibitor herbicides as essential tools for effective weed management in soybean production, especially given the rise of genetically modified crops that are resistant to multiple herbicides and the increasing prevalence of herbicide-resistant weed populations (Takano and Dayan 2020). Although resistance to glufosinate has not yet become widespread, implementing proactive and diverse management strategies now is essential to maintaining the herbicide's long-term effectiveness and mitigating the further evolution of multiple herbicide resistance (Takano and Dayan 2020). One step is by applying them only with effective mix partners in diversified preemergence-postemergence herbicide programs. A second step is to employ practices that enhance soybean competitiveness such as early planting, narrow row spacing, and well-timed termination of cover crops to aid in weed suppression. A third step is by integrating diversified management approaches, including conservation practices such as cover cropping for increased weed suppression, crop rotation and diversification, mechanical cultivation where feasible, and by implementing innovative technologies such as targeted herbicide application technologies and weed seed destruction. This multi-tactic approach could help eliminate viable weed seed return to the soil and interrupt the perpetuation of resistant alleles.

Practical Implications

Mixing glufosinate with PPO-inhibitor herbicides, 2,4-D, or bentazon is unlikely to cause injury that will result in yield loss when they are applied before the V6 soybean growth stage. However, caution is recommended when it comes to lactofen, which showed the highest potential for soybean injury in this study. Although these mixtures may not consistently enhance waterhemp control compared to glufosinate alone, they offer an important benefit for herbicide resistance management. By incorporating additional SOAs, such mixtures help reduce selection pressure, an important strategy for delaying the evolution of herbicide resistance in waterhemp and other challenging weed species. Bentazon, flumiclorac-pentyl, and fluthiacet-methyl do not provide commercially acceptable waterhemp control. Fomesafen, lactofen, and 2,4-D all provided good waterhemp control in individual applications (>88%) and are effective partners for glufosinate. Less soybean injury occurred with 2,4-D than any PPO-inhibitor herbicide, and mixtures with glufosinate provided effective waterhemp control. Mixing the herbicides evaluated in this study with glufosinate may help protect against yield loss from weed competition compared to applying those herbicides alone. Our findings also suggest that including glufosinate as part of a preemergence-postemergence herbicide program can improve waterhemp control under the conditions evaluated herein.

Acknowledgments. We thank Rachel Renz, Frank Lazar, and Matt Humbert, undergraduate students in the Wisconsin Cropping Systems Laboratory, for their assistance with data collection. We also thank Kinsey Tiemann, Laine Crawford, Nathan Stufflebeam, Luke Bergschneider, Christian Reiner, Stephanie Reiter, and Dustin Steinkamp, undergraduate students at Western Illinois University, for their assistance with herbicide mixing, application, and treatment data collection. We are especially grateful to Brent Heaton for his efforts in conducting the trials at the Illinois experimental locations.

Funding. This research was partially funded by the Wisconsin Soybean Marketing Board.

Competing interests. The authors declare they have no competing interests.

References

- Arneson NJ, Smith DH, DeWerff R, Oliveira M, Werle R (2020) Residual control of waterhemp with pre-emergence herbicides in soybean. [https://wicweeds.info/images/2018%202019%20waterhemp%20challenge/PreEmergence_waterhempFINAL.pdf](https://wic weeds.info/images/2018%202019%20waterhemp%20challenge/PreEmergence_waterhempFINAL.pdf). Accessed: October 4, 2024
- Arsenjevic N, de Avellar M, Butts L, Arneson NJ, Werle R (2021) Influence of sulfentrazone and metribuzin applied preemergence on soybean development and yield. *Weed Technol* 35:210–215. doi: [10.1017/wet.2020.99](https://doi.org/10.1017/wet.2020.99)
- Arsenjevic N, DeWerff R, Conley S, Ruark M, Werle R (2022) Influence of integrated agronomic and weed management practices on soybean canopy development and yield. *Weed Technol* 36:73–78. doi: [10.1017/wet.2021.92](https://doi.org/10.1017/wet.2021.92)
- Barker AL, Pawlak J, Duke SO, Beffa R, Tranel PJ, Wuerffel J, Young B, Porri A, Liebl R, Aponte R, Findley D, Betz M, Lerchl J, Culpepper S, Bradley K, Dayan FE (2023) Discovery, mode of action, resistance mechanisms, and plan of action for sustainable use of Group 14 herbicides. *Weed Sci* 71:173–188. doi: [10.1017/wsc.2023.15](https://doi.org/10.1017/wsc.2023.15)
- Barnes ER, Knezevic SZ, Lawrence NC, Irmak S, Rodriguez O, Jhala AJ (2020) Control of velvetleaf (*Abutilon theophrasti*) at two heights with POST herbicides in Nebraska popcorn. *Weed Technol* 34:560–567
- Beam SC, Flessner ML, Pittman KB (2018) Soybean flower and pod response to fomesafen, acifluorfen, and lactofen. *Weed Technol* 32:444–447. doi: [10.1017/wet.2018.37](https://doi.org/10.1017/wet.2018.37)
- Beckie HJ (2006) Herbicide-resistant weeds: management tactics and practices. *Weed Technol* 20:793–814
- Belfry KD, Shropshire C, Sikkema PH (2016) Identity-preserved soybean tolerance to protoporphyrinogen oxidase-inhibiting herbicides. *Weed Technol* 30:137–147
- Bernards ML, Crespo RJ, Kruger GR, Gaussoin R, Tranel PJ (2012). A waterhemp (*Amaranthus tuberculatus*) population resistant to 2,4-D. *Weed Sci* 60:379–384
- Besançon TE, Penner D, Everman WJ (2018) Reduced translocation is associated with antagonism of glyphosate by glufosinate in giant foxtail (*Setaria faberi*) and velvetleaf (*Abutilon theophrasti*). *Weed Sci* 66: 159–167
- Brooks ME, Kristensen K, Van Benthem KJ, Magnusson A, Berg CW, Nielsen A, Skaug HJ, Machler M, Bolker BM (2017) glmmTMB balances speed and flexibility among packages for zero-inflated generalized linear mixed modeling. *R J* 9:378–400. doi: [10.32614/RJ-2017-066](https://doi.org/10.32614/RJ-2017-066)
- Burke IC, Askew SD, Corbett JL, Wilcut JW (2005) Glufosinate antagonizes clethodim control of goosegrass (*Eleusine indica*). *Weed Technol* 19:664–668
- 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
- Correndo AA, MoroRosso LH, Ciampitti IA (2021) Agrometeorological data using R-software. Cambridge, MA: Harvard University Dataverse. doi: [10.7910/DVN/J9EUZU](https://doi.org/10.7910/DVN/J9EUZU)
- Craigmyle BD, Ellis JM, Bradley KW (2013) Influence of herbicide programs on weed management in soybean with resistance to glufosinate and 2,4-D. *Weed Technol* 27:78–84
- Davies LR, Hull R, Moss S, Neve P (2019) The first cases of evolving glyphosate resistance in UK poverty brome (*Bromus sterilis*) populations. *Weed Sci* 67:41–47
- Dayan FE, Barker AL, Bough R, Ortiz M, Takano HK, Duke SO (2019) Herbicide mechanisms of action and resistance, Pages 36–48 in Moo-Young M, ed. *Comprehensive Biotechnology* 3rd ed. Oxford, UK: Pergamon
- Edwards JT, Purcell LC (2005) Light interception and yield response of ultra-short-season soybean to diphenylether herbicides in the Midsouthern United States. *Weed Technol* 19:168–175
- Ellis JM, Griffiin JL (2003) Glyphosate and broadleaf herbicide mixtures for soybean (*Glycine max*). *Weed Technol* 17:21–27
- Etheridge RE, Hart WE, Hayes RM, Mueller TC (2001) Effect of venturi-type nozzles and application volume on postemergence herbicide efficacy. *Weed Technol* 15:75–80

- Evans CM, Strom SA, Riechers DE, Davis AS, Tranel PJ, Hager AG (2019) Characterization of a waterhemp (*Amaranthus tuberculatus*) population from Illinois resistant to herbicides from five site-of-action groups. *Weed Technol* 33:400–410
- Faleco FA, Machado FM, Bobadilla LK, Tranel PJ, Stoltenberg D, Werle R (2024) Resistance to protoporphyrinogen oxidase inhibitors in giant ragweed (*Ambrosia trifida*). *Pest Manag Sci* 80:6211–6221
- Faleco FA, Oliveira MC, Arneson NJ, Renz M, Stoltenberg DE, Werle R (2022) Multiple herbicide resistance in waterhemp (*Amaranthus tuberculatus*) accessions from Wisconsin. *Weed Technol* 36:597–608. doi: [10.1017/wet.2022.81](https://doi.org/10.1017/wet.2022.81)
- Hager AG, Wax LM, Bollero GA, Stoller, EW (2003) Influence of diphenylether herbicide application rate and timing on common waterhemp (*Amaranthus rudis*) control in soybean (*Glycine max*) *Weed Technol* 17:14–20
- Harris JR, Gossett BJ, Murphy TR, Toler JE (1991) Response of broadleaf weeds and soybeans to the diphenylether herbicides. *J Prod Agric* 4:407–411
- Hay MM, Dille JA, Peterson DE (2019) Integrated pigweed (*Amaranthus* spp.) management in glufosinate-resistant soybean with a cover crop, narrow row widths, row-crop cultivation, and herbicide program. *Weed Technol* 33:710–719
- Heap I (2024) The International Herbicide-Resistant Weed Database. www.weedscience.org. Accessed: March 21, 2024
- Higgins JM, Whitwell T, Murdock EC, Toler JE (1988) Recovery of pitted morningglory (*Ipomoea lacunosa*) and ivyleaf morningglory (*Ipomoea hederacea*) following applications of acifluorfen, fomesafen, and lactofen. *Weed Sci* 36:345–353
- Jha P, Norsworthy JK (2009) Soybean canopy and tillage effects on emergence of Palmer amaranth (*Amaranthus palmeri*) from a natural seed bank. *Weed Sci* 57:644–651
- Johnson BF, Bailey WA, Wilson HP, Holshouser DL, Herbert DA Jr, Hines TE (2002) Herbicide effects on visible injury, leaf area, and yield of glyphosate-resistant soybean (*Glycine max*). *Weed Technol* 16:554–566
- Joseph DD, Marshall MW, Sanders CH (2018) Efficacy of 2,4-D, dicamba, glufosinate and glyphosate combinations on selected broadleaf weed heights. *Am J Plant Sci* 9:1321–1333
- Kapusta G, Jackson LA, Mason DS (1986) Yield response of weed-free soybeans (*Glycine max*) to injury from postemergence broadleaf herbicides. *Weed Sci* 34:304–307
- Kumaratilake AR, Preston C (2005) Low temperature reduces glufosinate activity and translocation in wild radish (*Raphanus raphanistrum*). *Weed Sci* 53:10–16
- Lanclos DY, Webster EP, Zhang W (2002) Glufosinate tank-mix combinations in glufosinate-resistant rice (*Oryza sativa*). *Weed Technol* 16:659–663
- Liang K, Ma Y, Xie Y, Zhou B, Wang R (2012) A new adaptive contrast enhancement algorithm for infrared images based on double plateaus histogram equalization. *Infrared Phys Technol* 55:309–315
- 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
- Martinson KB, Durgan BR, Gunsolus JL, Sothorn RB (2005) Time of day of application effect on glyphosate and glufosinate efficacy. *Crop Manag* 4:1–6
- Merchant RM, Sosnoskie LM, Culpepper AS, Steckel LE, York AC, Braxton LB, Ford JC (2013) Weed response to 2,4-D, 2,4-DB, and dicamba applied alone or with glufosinate. *J Cotton Sci* 17:212–218
- Nelson KA, Renner KA (2001) Soybean growth and development as affected by glyphosate and postemergence herbicide tank mixtures. *Agron J* 93:428–434
- Norsworthy JK, Ward SM, Shaw DR, Llewellyn RS, Nichols RL, Webster TM, Bradley KW, Frisvold G, Powles SB, Burgos NR, Witt WW (2012) Reducing the risks of herbicide resistance: best management practices and recommendations. *Weed Sci* 60(SP1):31–62
- Oliveira MC, Feist D, Eskelsen S, Scott JE, Knezevic SZ (2017) Weed control in soybean with preemergence- and postemergence-applied herbicides. *CFTM* 3:1–7
- Paruelo JM, Lauenroth WK, Roset PA (2000) Technical note: Estimating aboveground plant biomass using a photographic technique. *J Range Manag* 53:190–193
- Patrignani A, Ochsner TE (2015) Canopeo: A powerful new tool for measuring fractional green canopy cover. *Agron J* 107:2312–2320
- Prade J (2022). BASF and Corteva Agriscience collaborate to deliver the future of weed control to soybean farmers. Joint News Release - BASF and Corteva Agriscience collaborate to deliver the future of weed control to soybean farmers. Accessed: December, 2024
- Priess GL, Norsworthy JK, Roberts TL, Gbur EE (2020) Impact of postemergence herbicides on soybean injury and canopy formation. *Weed Technol* 34:727–734. doi: [10.1017/wet.2020.55](https://doi.org/10.1017/wet.2020.55)
- R Core Team (2021) R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing. <https://www.r-project.org/>
- Riley EB, Bradley KW (2014) Influence of application timing and glyphosate tank-mix combinations on the survival of glyphosate-resistant giant ragweed (*Ambrosia trifida*) in soybean. *Weed Technol* 28:1–9
- Sarangi D, Jhala AJ (2015) Tips for identifying postemergence herbicide injury symptoms in soybean. Circular 497(8). Lincoln: University of Nebraska Extension
- Shergill LS, Barlow BR, Bish MD, Bradley KW (2018) Investigations of 2,4-D and multiple herbicide resistance in a Missouri waterhemp (*Amaranthus tuberculatus*) population. *Weed Sci* 66:386–394. doi: [10.1017/wsc.2017.82](https://doi.org/10.1017/wsc.2017.82)
- Shoup DE, Al-Khatib K, Peterson DE (2003) Common waterhemp (*Amaranthus rudis*) resistance to protoporphyrinogen oxidase-inhibiting herbicides. *Weed Sci* 51:145–150
- Smith A, Soltani N, Kaastra AJ, Hooker DC, Robinson DE, Sikkema PH (2019) Annual weed management in isoxaflutole-resistant soybean using a two-pass weed control strategy. *Weed Technol* 33:411–425
- Steckel GJ, Wax LM, Simmons FW, Phillips WH (1997) Glufosinate efficacy on annual weeds is influenced by rate and growth stage. *Weed Technol* 11:484–488
- Striegel A, Eskridge KM, Lawrence NC, Knezevic SZ, Kruger GR, Proctor CA, Hein GL, Jhala AJ (2020) Economics of herbicide programs for weed control in conventional, glufosinate-, and dicamba/glyphosate-resistant soybean across Nebraska. *Agron J* 112:5158–5179
- Takano HK, Beffa R, Preston C, Westra P, Dayan FE (2019) Reactive oxygen species trigger the fast action of glufosinate. *Planta* 249:1837–1849. <https://doi.org/10.1007/s00425-019-03124-3>
- Takano HK, Beffa R, Preston C, Westra P, Dayan FE (2020a) A novel insight into the mode of action of glufosinate: how reactive oxygen species are formed. *Photosynth Res* 144:361–372. <https://doi.org/10.1007/s11220-020-00749-4>
- Takano HK, Beffa R, Preston C, Westra P, Dayan FE (2020b) Glufosinate enhances the activity of protoporphyrinogen oxidase inhibitors. *Weed Sci* 68:324–332
- Takano HK, Dayan FE (2020) Glufosinate-ammonium: a review of the current state of knowledge. *Pest Manag Sci* 76:3911–3925
- Tharp BE, Schabenberger O, Kells JJ (1999). Response of annual weed species to glufosinate and glyphosate. *Weed Technol* 13:542–547
- Thornton PE, Thornton MM, Mayer BW, Wei Y, Devarakonda R, Vose RS, Cook RB (2016) Daymet: Daily Surface Weather Data on a 1-km Grid for North America, Version 3. Oak Ridge, TN: National Aeronautics and Space Administration, Oak Ridge National Laboratory ORNL DAAC. https://daac.ornl.gov/cgi-bin/dsvviewer.pl?ds_id=1328
- Torra J, Alcántara-de la Cruz R, de Figueiredo MRA, Gaines TA, Jugulam M, Merotto A Jr, Palma-Bautista C, Rojano-Delgado AM, Riechers DE (2024) Metabolism of 2,4-D in plants: comparative analysis of metabolic detoxification pathways in tolerant crops and resistant weeds. *Pest Manag Sci* 80:6041–6052. <https://doi.org/10.1002/ps.8373>
- Tranel PJ, Riggins CW, Bell MS, Hager AG (2011) Herbicide resistances in *Amaranthus tuberculatus*: a call for new options. *J Agric Food Chem* 59:5808–5812
- [USGS] U.S. Geological Survey (2018) Estimated Annual Agricultural Pesticide Use. https://water.usgs.gov/nawqa/pnsp/usage/maps/show_map.php?year=2018&hilo=L&map=GLUFOSINATE. Accessed: April 1, 2024
- Van Wychen L (2022) 2022 Survey of the most common and troublesome weeds in broadleaf crops, fruits & vegetables in the United States and Canada.

- Westminster, CO: Weed Science Society of America National Weed Survey Dataset. <https://wssa.net/wp-content/uploads/2022-Weed-Survey-Broadleaf-crops.xlsx>. Accessed: October 4, 2024
- Van Wychen L (2023) 2023 Survey of the most common and troublesome weeds in grass crops, pasture and turf in the United States and Canada. Westminster, CO: Weed Science Society of America National Weed Survey Dataset. <http://wssa.net/weed/surveys/2023> Weed Survey_Grass crops.xlsx. Accessed: October 4, 2024
- Werle R, DeWerff RP, Mobli A, Arneson NJ (2023) Evaluation of foliar-applied post-emergence corn-soybean herbicides on giant ragweed and waterhemp control in Wisconsin. *Agrosyst Geosci Environ* 6:e20338 <https://doi.org/10.1002/agg2.20338>
- Whitaker JR, York AC, Jordan DL, Culpepper S (2010) Palmer amaranth (*Amaranthus palmeri*) control in soybean with glyphosate and conventional herbicide systems. *Weed Technol* 24:403–410
- Wichert RA, Talbert RE (1993). Soybean [*Glycine max* (L.)] response to lactofen. *Weed Sci* 41:23–27
- Young BG, Young JM, Matthews JL, Owen MDK, Zelaya IA, Hartzler RG, Wax LM, Rorem KW, Bollero GA (2003) Soybean development and yield as affected by three postemergence herbicides. *Agron J* 95:1152–1156