Weed Technology

www.cambridge.org/wet

Pervasive weed management and soybean yield with 2,4-D and glufosinate applied alone, mixed, or sequentially

Eric A.L. Jones[1], Jill K. Alms[2] and David A. Vos[2]

[1]Assistant Professor, Agronomy, Horticulture, and Plant Science Department, South Dakota State University, Brookings, SD, USA and [2]Agricultural Research Manager, Agronomy, Horticulture, and Plant Science Department, South Dakota State University, Brookings, SD, USA

Research Article

Cite this article: Jones EAL, Alms JK, Vos DA (2025) Pervasive weed management and soybean yield with 2,4-D and glufosinate applied alone, mixed, or sequentially. Weed Technol. **39**(e100), 1–7. doi: 10.1017/wet.2025.10053

Received: 3 June 2025
Revised: 21 August 2025
Accepted: 4 October 2025

Associate Editor:
Rafael Pedroso, University of São Paulo (ESALQ/USP)

Nomenclature:
Glufosinate; 2; 4-D; common lambsquarters; *Chenopodium album* L.; common waterhemp; *Amaranthus tuberculatus* Moq. J.D. Sauer; redroot pigweed; *Amaranthus retroflexus* L.; velvetleaf; *Abutilon theophrasti* L.; yellow foxtail; *Setaria pumila* (Poir.) Roem. & Shult.

Keywords:
Herbicide interactions; herbicide resistance; weed management; sequential applications

Corresponding author:
Eric A.L. Jones; Email: eric.jones@sdstate.edu

Abstract

Field experiments were conducted near Beresford and South Shore, South Dakota, in 2023 and 2024, to determine weed control and soybean yield with 2,4-D and glufosinate applied individually, together, and sequentially. Herbicides were sequentially applied 12 d after an initial application. 2,4-D + glufosinate additively controlled and reduced the height of all tested weed species. Sequential applications resulted in greater control of common lambsquarters, waterhemp, redroot pigweed, and velvetleaf compared with 2,4-D or glufosinate applied alone, or 2,4-D + glufosinate applied in a mixture. The order in which herbicides were sequentially applied did not influence broadleaf weed control. Yellow foxtail control was greater with sequential applications of glufosinate. Soybean yield at Beresford was similar across all treatments. Yields were generally greater at South Shore with sequential herbicide applications when glufosinate was initially applied. The experiment results suggest that weed control and soybean yield are greater with 2,4-D + glufosinate or sequential application treatments that include 2,4-D and glufosinate.

Introduction

Herbicides are recommended to be applied in mixtures to increase their efficacy, application efficiency, control spectrum, and to reduce selection pressure on herbicide-resistant weed biotypes (Green 1989; Renton et al. 2024). Herbicide mixtures can be antagonistic, additive, or synergistic on select weeds (Colby 1967; Green 1989) depending on the chemical makeup or their effects on plant physiology (Barbieri et al. 2022; Green 1989). Physiological antagonism or synergism can occur when herbicides with different modes of action are mixed and act either negatively or positively in planta (Meyer et al. 2019; Ou et al. 2018). Because herbicide mixtures and sequential applications are often recommended to control unwanted weeds, understanding how they perform on various weed species under field conditions is critically necessary.

2,4-Dichlorophenoxyacetic acid (2,4-D), a Group 4 herbicide as categorized by the Weed Science Society of America (WSSA) and glufosinate (WSSA Group 10) applied either individually or mixed together and sequentially may become more common for weed management due to herbicide resistance and the commercial availability of tolerant soybean varieties (Shyam et al. 2021). 2,4-D is a slow-acting, phloem-mobile herbicide that asserts its activity predominantly on broadleaf weeds. It kills plants by derepressing transcription factors released by auxin receptors to cause an increase auxin concentrations, which leads to growth malformations, excessive production of reactive oxygen species, induction of stress responses, and ultimately, plant death due to chloroplast destruction (Grossman 2010; Peterson et al. 2016). Glufosinate is a fast-acting, contact herbicide with activity on some broadleaf and grass weeds (Corbett et al. 2004). Glufosinate inhibits glutamine synthetase (EC 6.3.1.2), which in turn inhibits the production of photosynthesis precursors, leading to the production of reactive oxygen species, and ultimately a disruption in cell membrane integrity (Takano et al. 2020). The 2,4-D and glufosinate mixture is a labeled for use on soybean (Anonymous 2023, 2024). Because 2,4-D and glufosinate have different modes of action, we performed research to determine whether a mixture of the two is antagonistic, additive, or synergistic on various weed species. Sequential applications of these herbicides may also be used on soybean. The 2,4-D and glufosinate labels indicate that sequential applications should be limited to occur 12 and 5 d, respectively, after an initial herbicide application (Anonymous 2023, 2024). Our investigations to assess the interaction of sequential applications of 2,4-D and glufosinate also included application timings to reflect the most restrictive application according to the product label.

Waterhemp is the most troublesome weed in the Midwest region of the United States due to the biology of the plant and widespread evolution of herbicide resistance (Butts et al. 2018; Jones et al. 2019). Waterhemp has evolved resistance to herbicides from WSSA Groups 2, 4, 5, 6, 9, 14,

© 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.

Table 1. Average temperatures and precipitation during the experiments.[a]

		2023		2024	
		Average temperature	Precipitation	Average temperature	Precipitation
		C	mm	C	mm
Beresford	May	18.3	41	16.7	178.6
	June	22.8	74.4	21.7	119.1
	July	21.7	128.3	22.8	20.6
	August[b]	–	–	21.1	41.4
South Shore	May	16.7	36.3	18.3	98
	June	21.7	57.9	21.1	119.1
	July	20	39.9	21.1	126
	August[b]	–	–	19.4	55.9

[a]Experiments were conducted in Beresford and South Shore, South Dakota, in 2023 and 2024. Weather data are presented for the month when soybean was planted until the last evaluation period in each year.
[b]Evaluations were completed before August.

Table 2. Herbicides 2,4-D and glufosinate applied to pervasive weeds that compete with soybean.[a–d]

Initial	Sequential	Herbicide treatment
2,4-D	None	2,4-D
2,4-D	2,4-D	2,4-D fb 2,4-D
2,4-D	G	2,4-D fb G
2,4-D	2,4-D + G	2,4-D fb 2,4-D + G
G	None	G
G	2,4-D	G fb 2,4-D
G	G	G fb G
G	2,4-D + G	G fb 2,4-D + G
2,4-D + G	None	2,4-D + G
2,4-D + G	2,4-D	2,4-D + G fb D
2,4-D + G	G	2,4-D + G fb G
2,4-D + G	2,4-D + G	2,4-D + G fb 2,4-D + G
N	2,4-D	N fb 2,4-D
N	G	N fb G
N	2,4-D + G	N fb 2,4-D + G

[a]Abbreviations: fb, followed by; G, glufosinate, N, no initial treatment, 2,4-D, 2,4-dichlorophenoxyacetic acid.
[b]Each herbicide treatment was tested on common lambsquarters, common waterhemp, redroot pigweed, yellow foxtail, and velvetleaf.
[c]2,4-D was applied at 1,165 g ae ha^{-1}; glufosinate was applied at 655 g ai ha^{-1}.
[d]Experiments were carried out in Beresford and South Shore, South Dakota, in 2023 and 2024.

15, and 27, and reports of populations being resistant to multiple herbicides are common (Faleco et al. 2022; Heap 2025). Resistance to 2,4-D has been confirmed in Illinois, Minnesota, Missouri, and Nebraska (Bernards et al. 2012; Evans et al. 2019; Shergill et al. 2018; Singh et al. 2024). While glufosinate resistance has not yet been confirmed in waterhemp, control failures have been reported (Hamberg et al. 2023; Landau et al. 2025). Glufosinate and 2,4-D applied initially alone or in a mixture and then sequentially to waterhemp has been previously studied. The research determined that 2,4-D + glufosinate provided additive control, and sequential applications were more efficacious than single applications (Craigmyle et al. 2013a, 2013b; Haarman et al. 2020).

Although waterhemp is the focus of many weed management plans, other weed species are usually present in sufficient numbers to require management. Therefore, we sought to determine the effectiveness of various 2,4-D and glufosinate treatments on other common weed species. 2,4-D does not affect grass weed species and may increase grass weed population densities if applied too extensively (Hodgskiss et al. 2022). Currently, no published data exist on the effectiveness of 2,4-D and glufosinate mixed or applied sequentially against common lambsquarters, redroot pigweed, yellow foxtail, and velvetleaf, which are historically pervasive weeds of soybean (Shurtlee and Coble 1985; Staniforth 1965; Stoller and Woolley 1985). Thus the objectives of this research were 1) to determine whether 2,4-D + glufosinate had additive, antagonistic, or synergistic affects against these weeds; 2) to compare the effectiveness of sequential applications with singular herbicide applications; and 3) assess how the herbicide treatments affected soybean yield.

Materials and Methods

Field experiments were conducted in 2023 and 2024 at two locations in South Dakota: Beresford (43.050067°N, 96.896512°W) and South Shore (45.106553°N, 97.095680°W), for a total of 4 site-years. The soil at the Beresford location is an Egan-Trent silty clay loam (Fine-silty, mixed, superactive, mesic Pachic/Udic Haplustoll). At South Shore, the soil is a Kranzburg-Brookings silty clay loam (Fine-silty, mixed, superactive, frigid Calcic/Pachic Hapludoll). Common lambsquarters, waterhemp, and velvetleaf populations were identified at Beresford (2 site-years) and populations of redroot pigweed and yellow foxtail were identified at the South Shore site (2 site-years). Each site was tilled with a field cultivator before the experiments began. All fields had previously been cultivated with a corn-soybean rotation. Soybean seeds were planted at 395,000 seeds ha^{-1} with 76-cm row spacing for all experiments. The soybean varieties DSR-150SE (Dairyland Seed Company, Corteva Agriscience, Indianapolis, IN) and NK09-H7E3 (NK Seeds, Syngenta Crop Protection, Greensboro, NC) were planted on May 17 and May 23, 2023, at the Beresford and South Shore sites, respectively. Soybean varieties AE1900 (Mustang Seeds, M.S. Technologies, West Point, IA) and AE1030 (Mustang Seeds) were planted on May 16 and June 7, 2024, at the Beresford and South Shore sites, respectively. Weather data for each site year are provided in Table 1. Preemergence herbicides were omitted to ensure maximum weed emergence.

Treatments were arranged in a randomized complete block design with four replications. Individual plots were 3 m wide by 12 m long. Herbicide treatments are listed in Table 2. Herbicide treatments were applied to plots with a CO_2-pressurized backpack sprayer calibrated to deliver 140 L ha^{-1} at 165 kPa while traveling at 4.8 km h^{-1} and 46 cm above the target weed height. Weeds were approximately 15 cm high (7.6 to 56 cm in 2023; 5 to 38 cm in 2024) at the time of treatment. The target height of 15 cm was selected based on the 2,4-D label despite the glufosinate label recommending treating weeds at a target height of 7.6 cm

https://doi.org/10.1017/wet.2025.10053 Published online by Cambridge University Press

Table 3. Visual estimates of weed control 28 d after initial applications of 2,4-D and glufosinate to soybean.[a–c]

Herbicide treatment	ABUTH	AMARE	AMATA	CHEAL	SETPU
	% (SE)				
2,4-D	92 (4) b	84 (4) e	89 (3) cd	94 (3) ab	–[d]
2,4-D fb 2,4-D	99 (0.3) a	88 (3) de	96 (1) ab	97 (1) a	–[d]
2,4-D fb G	99 (0.125) a	99 (0) a	97 (1) a	99 (0.1) a	79 (3) c
2,4-D fb 2,4-D + G	99 (0) a	99 (0.25) a	97 (1.5) ab	99 (0) a	77 (5) c
G	97 (1) a	92 (2) cd	66 (4) f	82 (4) c	58 (8) de
G fb 2,4-D	99 (0.25) a	99 (0.25) ab	94 (2) abc	97 (1) a	63 (7) d
G fb G	99 (0) a	96 (2.5) ab	98 (0.5) a	99 (0.3) a	92 (3) ab
G fb 2,4-D + G	99 (0) a	99 (0.125) a	99 (0.125) a	99 (0) a	93 (2) ab
2,4-D + G	99 (0.3) a	96 (0.9) abc	87 (4) d	96 (3) a	62 (9) d
2,4-D + G fb 2,4-D	99 (0) a	99 (0.125) a	96 (2) ab	98 (1) a	49 (3) e
2,4-D + G fb G	99 (0) a	99 (0.125) a	99 (0.4) a	99 (0) a	95 (2) a
2,4-D + G fb 2,4-D + G	99 (0) a	99 (0.125) a	99 (0) a	99 (0) a	94 (2) a
N fb 2,4-D	93 (2) b	64 (5) f	69 (3) f	88 (5) bc	–[d]
N fb G	99 (0.25) a	94 (2) bcd	80 (3) e	87 (3) c	83 (2) bc
N fb 2,4-D + G	98 (0.4) a	94 (3) abcd	92 (2) bcd	96 (1) a	77 (3) c

[a]Abbreviations: ABUTH, velvetleaf; AMARE, redroot pigweed; AMATA, common waterhemp; CHEAL, common lambsquarters; fb, followed by; G, glufosinate; N, no initial treatment; SETPU, yellow foxtail; 2,4-D, 2,4-dichlorophenoxyacetic acid.
[b]Means that share the same letter within columns are not statistically different based on Fisher's least significant difference test ($P < 0.05$).
[c]Treatments that violated the constant variance assumption were not included in the analysis, but 95% confidence intervals were used to determine whether values were similar.
[d]Clethodim was applied with 2,4-D-only treatments; therefore, no data were collected for yellow foxtail.

(Anonymous 2023, 2024). Herbicides were applied sequentially 12 d after the initial herbicide application (DAIT). The weeds were approximately 50 cm tall at the sequential application (20 to 91 cm in 2023; 35 to 66 cm in 2024). All treatments were applied with 8003 AIXR spray nozzles (TeeJet Technologies, Glendale Heights, IL). These nozzles were selected to achieve a consistent herbicide application based on the 2,4-D label (Anonymous 2023). The 2,4-D product was Enlist One (Corteva Agriscience, Indianapolis, IN), applied at 1,165 g ae ha^{-1} in all 2,4-D treatments. The glufosinate product was Liberty (BASF, Raleigh, NC), applied at 655 g ai ha^{-1}, along with 10 g L^{-1} ammonium sulfate whenever glufosinate was applied. Clethodim (560 g ai ha^{-1}) was applied 21 DAIT with the spray parameters as described above to 2,4-D-only treatments to control grass weeds that could confound the control of broadleaf weeds. No response variable data were recorded for grass species in these plots.

Weed control was evaluated using visual estimates based on a scale ranging from 0% to 100%, where 0% equals no control (i.e., no injury symptoms on any tissue) and 100% equals complete control (i.e., total necrosis). Weed height reduction was determined by measuring from the soil surface to the apical growing point of three representative plants of each species arbitrarily selected in the central region of each plot. Percentage height reduction was calculated by dividing the heights of the plants in the treated plots by the heights of the plants in the nontreated plots. Control and height reduction evaluations were conducted 28 DAIT. Soybeans were harvested after reaching physiological maturity using a combine, and the yield was adjusted to 13% moisture.

Statistical Analysis

Control, height reduction, and soybean yield data were subjected to ANOVA using the Glimmix procedure with SAS software (v.9.4; Statistical Analysis Systems, Cary, NC) ($\alpha = 0.05$). Herbicide treatment was considered a fixed effect, whereas block and year and their interactions were considered random effects. Year was considered random to allow inferences to be made across broader conditions and locations (Blouin et al. 2011; Moore and Dixon 2015). Treatment means were separated using Fisher's least significant difference test ($P \leq 0.05$).

2,4-D + glufosinate mixtures were evaluated at 28 DAIT to determine whether the resultant activity was additive, antagonistic, or synergistic using the Colby method (Colby 1967). The Colby method calculates an expected control value for an herbicide mixture based on the control of the individual herbicides, and the expected control value is compared with the control of the tested mixture. Equation 1 shows how the Colby method was applied to analyze 2,4-D + glufosinate treatments:

$$E = (XplusY) - \left(\frac{xy}{100}\right) \quad [1]$$

where E is the expected percent control of two herbicides applied in a mixture, X is the percent control of X herbicide when applied alone, and Y is the percent control of Y herbicide when applied alone. The expected control was compared with the observed control using a two-sided t-test ($\alpha = 0.05$). If the control was greater than the expected percent control, the mixture was considered to be synergistic, whereas if the percent control was lower than the expected percent control, the mixture was antagonistic (Colby 1967). If the observed and expected percent controls were equal, the mixture was considered additive (Colby 1967). Since 2,4-D does not control yellow foxtail, statistical deviations from the single and mixed treatments of 2,4-D and glufosinate can provide evidence of either antagonism or synergism (Flint and Barrett 1989; Meyer and Norsworthy 2019). Height reduction data of the broadleaf weeds were also subjected to the Colby method. The control of sequential herbicide treatments was compared to the control of the 2,4-D + glufosinate mixture to determine whether activity was antagonistic or synergistic (Burke et al. 2005).

Results

Velvetleaf Control

An initial application of 2,4-D and no initial treatment followed by (fb) a secondary application of 2,4-D provided the least control (92% and 93% control, respectively) of velvetleaf compared with the other herbicide treatments (Table 3). Every treatment provided more than 90% velvetleaf control, suggesting that all herbicide

https://doi.org/10.1017/wet.2025.10053 Published online by Cambridge University Press

Table 4. Weed height reduction evaluated 28 d after initial applications of 2,4-D and glufosinate to soybean.[a–c]

Herbicide treatment	ABUTH	AMARE	AMATA	CHEAL	SETPU
	% (±SE)				
2,4-D	83 (6) cd	79 (1) bc	68 (6) cd	89 (4) abc	–[d]
2,4-D fb 2,4-D	95 (3) abc	77 (1.5) cd	86 (3) ab	91 (3) ab	–[d]
2,4-D fb G	98 (2) ab	100 (0) a	88 (4) ab	95 (3) a	67 (6) bc
2,4-D fb 2,4-D + G	100 (0) a	96 (1.5) a	93 (4) a	100 (0) a	65 (4) bc
G	75 (10) d	62 (2) f	47 (5) e	50 (1) f	62 (5) bc
G fb 2,4-D	93 (4) abc	90 (2) ab	88 (5) ab	90 (4) abc	71 (3) b
G fb G	100 (0) a	98 (0.6) a	91 (4) a	93 (4) a	90 (4) a
G fb 2,4-D + G	99 (1) a	96 (1) a	94 (2) a	97 (2) a	91 (3) a
2,4-D + G	96 (3) abc	74 (1.5) cde	77 (6) bc	95 (3) a	67 (3) bc
2,4-D + G fb 2,4-D	96 (3) a	99 (0.5) a	88 (4) ab	94 (3) a	63 (2) bc
2,4-D + G fb G	97 (3) ab	99 (0.5) a	96 (2) a	97 (2) a	90 (4) a
2,4-D + G fb 2,4-D + G	98 (2) ab	99 (0.25) a	95 (2) a	100 (0) a	92 (3) a
N fb 2,4-D	72 (8) d	74 (1) cde	60 (3) de	81 (5) bcd	–[d]
N fb G	75 (8) d	67 (3) def	70 (5) cd	80 (6) cd	59 (3) c
N fb 2,4-D + G	83 (10) bcd	65 (2) ef	67 (4) cd	76 (5) e	64 (7) bc

[a]Abbreviations: ABUTH, velvetleaf; AMARE, redroot pigweed; AMATA, common waterhemp; CHEAL, common lambsquarters; SETPU, yellow foxtail; fb, followed by; G, glufosinate; N, no initial treatment; 2,4-dichlorophenoxyacetic acid.
[b]Means that share the same letter within columns are not statistically different based on Fisher's least significant difference test ($P < 0.05$).
[c]Treatments that violated the constant variance assumption were not included in the analysis, but 95% confidence intervals were used to determine whether values were similar.
[d]Clethodim was applied to 2,4-D-only treatments; therefore, no data were collected for yellow foxtail.

treatments were effective. Velvetleaf height was reduced by 20% or more with sequential herbicide applications (Table 4). reduced Velvetleaf height was reduced with 2,4-D + glufosinate and no initial treatment fb 2,4-D + glufosinate, results that are similar to those of some sequential applications (Table 4). 2,4-D + glufosinate and no initial treatment fb 2,4-D + glufosinate were determined to provide additive control and height reduction (Table 5).

Redroot Pigweed Control

Glufosinate and 2,4-D + glufosinate provided approximately 10% more control of redroot pigweed than 2,4-D applied alone (Table 3). Similarly, no initial treatment fb an application of glufosinate and no initial treatment fb an application of 2,4-D + glufosinate provided 30% more control than no initial treatment fb an application of 2,4-D (Table 3). All sequential applications provided more redroot pigweed control than single applications except for 2,4-D fb 2,4-D for which control was approximately 8% less than a single treatment (Table 3).

Applications of 2,4-D and 2,4-D + glufosinate resulted in a reduction in redroot pigweed height by approximately 15% more than a single application of glufosinate (Table 4). Redroot pigweed height was similarly reduced when plots received no initial treatments fb all other herbicide treatments (Table 4). Height reduction with sequential herbicide applications followed a similar trend compared with nontreated control plants (Tables 3 and 4). An application of 2,4-D + glufosinate and no initial treatment fb 2,4-D + glufosinate were additive for redroot pigweed control and height reduction (Table 5).

Waterhemp Control

The 2,4-D and 2,4-D + glufosinate treatments provided more waterhemp control than glufosinate applied alone. Less waterhemp control occurred with no initial treatment fb an application of 2,4-D compared with 2,4-D applied alone (Table 3). Glufosinate was not as effective at controlling waterhemp as no initial treatment fb glufosinate and 2,4-D + glufosinate, and no initial treatment fb 2,4-D + glufosinate provided similar waterhemp control (Table 3). Common waterhemp control was improved by 7% to 30% with most sequential herbicide applications compared with a single herbicide (Table 3).

Waterhemp height reductions and waterhemp control were similar (Table 4). Waterhemp control with 2,4-D + glufosinate and no initial treatment fb 2,4-D + glufosinate were determined to be additive (Table 4). The effect of 2,4-D + glufosinate and no initial treatment fb 2,4-D + glufosinate on waterhemp height was determined to be additive (Table 5).

Common Lambsquarters Control

The treatments 2,4-D and 2,4-D + glufosinate provided 12% to 16% more control of common lambsquarters than glufosinate alone (Table 3). Common lambsquarters control was improved by approximately 8% more when no initial herbicide was fb 2,4-D + glufosinate than no initial treatment fb 2,4-D and no initial treatment fb glufosinate (Table 3). Sequential herbicide applications provided similar common lambsquarters control to that of 2,4-D + glufosinate and no initial treatment fb 2,4-D + glufosinate. However, the sequential applications provided 12% greater common lambsquarters control than glufosinate, no initial treatment fb 2,4-D, and no initial treatment fb glufosinate (Table 3).

Glufosinate and no initial treatment fb 2,4-D + glufosinate resulted in the least height reduction of common lambsquarters (Table 4). Common lambsquarters height was similarly reduced with the treatments 2,4-D, 2,4-D + glufosinate, no initial treatment fb 2,4-D, and no initial treatment fb glufosinate. Height reductions were greatest with sequential applications (Table 3). Common lambsquarters control and height reduction were determined to be additive for 2,4-D + glufosinate and no initial treatment fb 2,4-D + glufosinate (Table 5).

Yellow Foxtail Control

Two glufosinate applications provided greater control of yellow foxtail than one glufosinate application (92% and 58%, respectively) (Table 3). Glufosinate applied once provided 58% control, glufosinate fb 2,4-D provided 63% control, and 2,4-D fb

https://doi.org/10.1017/wet.2025.10053 Published online by Cambridge University Press

Table 5. Weed control and height reduction 28 d after initial applications of 2,4-D and glufosinate to soybean.[a]

Species	Herbicide treatment	Expected	Observed	P-value
Control		---------%---------		
ABUTH	2,4-D + G	100	99	0.80
	N fb 2,4-D + G	100	98	0.13
AMARE	2,4-D + G	98	96	0.26
	N fb 2,4-D + G	94	94	0.41
AMATA	2,4-D + G	96	87	0.13
	N fb 2,4-D + G	94	92	0.50
CHEAL	2,4-D + G	99	96	0.50
	N fb 2,4-D + G	98	96	0.30
Height reduction				
ABUTH	2,4-D + G	96	96	_[b]
	N fb 2,4-D + G	93	83	0.50
AMARE	2,4-D + G	92	74	0.053
	N fb 2,4-D + G	88	65	0.055
AMATA	2,4-D + G	83	77	0.50
	N fb 2,4-D + G	90	67	0.11
CHEAL	2,4-D + G	94	95	0.80
	N fb 2,4-D + G	96	76	0.16

[a]Abbreviations: ABUTH, velvetleaf; AMARE, redroot pigweed; AMATA, common waterhemp; CHEAL, common lambsquarters; SETPU, yellow foxtail; fb, followed by; G, glufosinate; N, no initial treatment; 2,4-D, 2,4-dichlorophenoxyacetic acid.
[b]Not calculated.

Table 6. Soybean yield 28 d after initial treatments with 2,4-D and glufosinate applied to soybean.[a,b]

Treatment	Beresford	South Shore
	kg ha^{-1} (±SE)	
Nontreated control	1,064 (161) c	766 (284) h
2,4-D	2,251 (293) ab	2,272 (280) cdef
2,4-D fb 2,4-D	2,423 (293) ab	2,130 (143) cdef
2,4-D fb G	2,431 (224) ab	2,040 (85) efg
2,4-D fb 2,4-D + G	2,645 (228) ab	2,031 (143) efg
G	2,402 (168) ab	2,353 (195) cde
G fb 2,4-D	2,622 (221) ab	2,601 (160) bc
G fb G	2,573 (287) ab	2,918 (66) ab
G fb 2,4-D + G	2,636 (227) ab	3,050 (77) a
2,4-D + G	2,689 (218) ab	2,460 (192) cde
2,4-D + G fb 2,4-D	2,844 (200) a	2,539 (154) bcd
2,4-D + G fb 2,4-D + G	2,497 (192) ab	2,907 (70) ab
2,4-D + G fb G	2,302 (136) ab	2,954 (81) ab
N fb 2,4-D	2,109 (187) b	2,155 (294) defg
N fb G	2,460 (222) ab	1,890 (103) fg
N fb 2,4-D + G	2,426 (208) ab	1,766 (88) g

[a]Abbreviations: fb, followed by; G, glufosinate; N, no initial treatment; 2,4-D, 2,4-dichlorophenoxyacetic acid.
[b]Means that share the same letter within columns are not statistically different based on Fisher's least significant difference test ($P < 0.05$).

glufosinate provided 79% control. Similarly, yellow foxtail height reduction was approximately 26% greater with two applications of glufosinate than treatments that included only one application of glufosinate (92% vs. 62%, respectively) (Table 4). Treatments of 2,4-D + glufosinate and glufosinate alone controlled yellow foxtail and reduced its height in similar ways, which suggests that the tank mixture has an additive activity (Tables 3 and 4). The same trend was observed with the no initial treatment fb 2,4-D+glufosinate and no initial treatment fb glufosinate treatments, further suggesting additive activity (Tables 3 and 4).

Soybean Yield

Due to a significant interaction between location and treatment ($P = 0.008$) and differences in the composition of weed species at both locations, soybean yield data were analyzed by location. Yields were similar with all treatments. At Beresford, the highest yield was measured after the 2,4-D + glufosinate fb 2,4-D treatment, and the lowest yield, other than from the nontreated control, was measured after no initial treatment fb an application of 2,4-D (Table 6). Yields from all other treatments were similar (Table 6).

Soybean yield at the South Shore location was higher with two sequential applications of glufosinate (Table 6), and these results correspond with increased yellow foxtail control (Tables 4, 5, and 6). Yields were similar after treatments with one application of glufosinate (Table 6). The yield from 2,4-D-only treatments at South Shore are likely not true representations of yield because clethodim was applied to control yellow foxtail. Therefore, yield would likely be lower if only 2,4-D was applied to a field with yellow foxtail or other grass weed species.

Discussion

Waterhemp control in our research was comparable to that reported previously using similar treatments (Craigmyle et al. 2013a, 2013b; Duenk et al. 2023). Waterhemp control estimates for the glufosinate and no initial treatment fb glufosinate were counterintuitive because no initial treatment fb glufosinate provided greater control; glufosinate effectiveness decreases as weed size increases (Steckel et al. 1997). The differences in control with these two treatments is likely a function of plant growth after treatment. Waterhemp plants treated with glufosinate had approximately 2 wk longer to exhibit regrowth compared with plants that received no initial treatment fb an application of glufosinate (Haarmann et al. 2020; Jones et al. 2024). Despite the control separation, neither of these treatments was effective against waterhemp, further bolstering the product label's recommendation that plants be treated when they are 7.6 cm tall by using nozzles that create fine droplets to increase coverage (Anonymous 2023).

While common lambsquarters, redroot pigweed, and velvetleaf control have not previously been reported with mixtures of 2,4-D and glufosinate or sequential applications of them, the control reported in previous research with single herbicide applications is similar to our results (Coetzer et al. 2002; Fawcett and Slife 1978; Robinson et al. 2012; Steckel et al. 1997). Yellow foxtail control in our research was much less than reported in previous research that used similar glufosinate rates applied to similar sized weeds (Corbett et al. 2004; Hamill et al. 2000). Additive control and height reduction occurred in all tested weed species, which is concordant with results from previous research on similar weed species (Craigmyle et al. 2013b; Merchant et al. 2013; Meyer and Norsworthy 2019).

The overall control and height reductions of broadleaf weed species were greater with sequential herbicide applications than with 2,4-D and glufosinate applied alone, but the order in which the herbicides were applied did not influence their efficacy. The order of sequential applications was important in controlling yellow foxtail and reducing its height. Both control and height reductions were greater with the glufosinate fb glufosinate regimen, which is similar to results reported in previous research (Jones et al. 2022). Control of common lambsquarters, redroot pigweed, and velvetleaf was similar between applications of 2,4-D + glufosinate and sequential applications. Height reductions of these

https://doi.org/10.1017/wet.2025.10053 Published online by Cambridge University Press

species followed a similar trend, excluding redroot pigweed, for which height reduction was greater with sequential applications. Waterhemp and yellow foxtail control and height reduction were greater with sequential herbicide applications compared with a single application of 2,4-D + glufosinate. Therefore, applying the 2,4-D + glufosinate mixture sequentially is recommended based on the weed species to be controlled. This conclusion is further reinforced by reduced soybean yields with herbicide treatments at South Shore that provided poor yellow foxtail control.

Practical Implications

The herbicides 2,4-D and glufosinate applied sequentially or together were more effective in managing the weed community as a whole. Since the herbicide application order did not influence the effectiveness for sequential applications to manage broadleaf species, both 2,4-D and glufosinate should be applied to reduce selection pressure on resistant biotypes. The mixture of 2,4-D + glufosinate is likely the most utilitarian because sequential applications rely on ideal conditions between the applications. This recommendation is further bolstered by soybean yields being generally higher when the mixture was applied or when both herbicides were applied sequentially. While yellow foxtail management was more effective with two glufosinate applications, this species should be managed with other effective herbicides or nonchemical tactics. These results further reinforce that weeds should be treated at a smaller size (<10 cm) with these herbicides because no initial treatments fb herbicide treatments were less effective against the weed species we tested and resulted in low soybean yield. Recommendations for these herbicides should focus on timely application (i.e., weeds should be 7.6 cm vs. >15 cm tall) using the most appropriate nozzle (i.e., fine vs. coarse droplet) and output for coverage (i.e., 140 vs. 187 L ha^{-1}).

Acknowledgments. We thank Allen Heuer, Bradley Rops, and Josalyn Fousert for their technical support. We also thank Micheal D.K. Owen for reviewing the manuscript before it was submitted.

Funding. Funding was provided by the South Dakota Soybean Research and Promotion Council.

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

References

Anonymous (2023) Enlist One® herbicide label. Indianapolis, IN: Corteva Agriscience

Anonymous (2024) Liberty 280SL® herbicide label. Research Triangle Park, NC: BASF Agricultural Solutions

Barbieri GF, Young BG, Dayan FE, Streibig JC, Takano H, Merotto A Jr, Avila LA (2022) Herbicide mixtures: interactions and modeling. Adv Weed Sci 40: e020220051

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

Blouin DC, Webster EP, Bond JA (2011) On the analysis of combined experiments. Weed Technol 25:165–169

Burke IC, Askew SD, Corbett JL, Wilcut JW (2005) Glufosinate antagonizes clethodim control of goosegrass (*Eleusine indica*). Weed Technol 19:664–668

Butts TR, Vieira BC, Latorre DO, Werle R, Kruger GR (2018) Competitiveness of herbicide-resistant waterhemp (*Amaranthus tuberculatus*) with soybean. Weed Sci 66:729–738

Coetzer E, Al-Khatib K, Peterson DE (2002) Glufosinate efficacy on *Amaranthus* species in glufosinate-resistant soybean (*Glycine max*). Weed Technol 16:326–331

Colby SR (1967) Calculating synergistic and antagonistic response of herbicide combinations. Weeds 15:20–22

Corbett JL, Askew SD, Thomas WE, Wilcut JW (2004) Weed efficacy evaluations for bromoxynil, glufosinate, glyphosate, pyrithiobac, and sulfosate. Weed Technol 18:443–453

Craigmyle BD, Ellis JM, Bradley KW (2013a) Influence of herbicide program on weed management in soybean with resistance to glufosinate and 2,4-D. Weed Technol 27:78–84

Craigmyle BD, Ellis JM, Bradley KW (2013b) Influence of weed height and glufosinate + 2,4-D combinations on weed control in soybean with resistance to 2,4-D. Weed Technol 27:271–280

Duenk E, Soltani N, Miller RT, Hooker DC, Robinson DE, Sikkema PH (2023) Multiple-herbicide-resistant waterhemp control in glyphosate/glufosinate/2,4-D-resistant soybean with one- and two-pass weed control programs. Weed Technol 37:34–39

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 sites-of-action. Weed Technol 33:400–410

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

Fawcett RS, Slife FW (1978) Effects of 2,4-D and dalapon on weed seed production and dormancy. Weed Sci 26:543–547

Flint JL, Barrett M (1989) Antagonism of glyphosate toxicity to johnsongrass (*Sorghum halepense*) by 2,4-D and dicamba. Weed Sci 37:700–705

Green JM (1989) Herbicide antagonism at the whole plant level. Weed Technol 3:217–226

Grossmann K (2010) Auxin herbicides: current status of mechanism and mode of action. Pest Manag Sci 66:113–120

Haarman JA, Young BG, Johnson WG (2020) Control of waterhemp (*Amaranthus tuberculatus*) regrowth after failed applications of glufosinate or fomesafen. Weed Technol 34:794–800

Hamberg RC, Yadav R, Owen MDK, Licht MA (2023) Differential susceptibility of Iowa waterhemp (*Amaranthus tuberculatus*) populations to 2,4-D, dicamba, and glufosinate. Can J Plant Sci 103:595–599

Hamill AS, Knezevic SZ, Chandler K, Sikkema PH, Tardif FJ, Shrestha A, Swanton CJ (2000) Weed control in glufosinate-resistant corn (*Zea mays*). Weed Technol 14:578–585

Heap I (2025) The International Herbicide-Resistant Weed Database. www.weedscience.org. Accessed: April 15, 2025

Hodgskiss CL, Legleiter TR, Young BG, Johnson WG (2022) Effects of herbicide management practices on the weed density and richness in 2,4-D-resistant cropping systems in Indiana. Weed Technol 36:130–136

Jones EAL, Bradshaw CL, Contreras DJ, Cahoon CW, Jennings KM, Leon RG, Everman WJ (2024) Growth and fecundity of Palmer amaranth escaping glufosinate in soybean. Weed Technol 38:e43

Jones EAL, Leon RG, Everman WJ (2022) Control of pervasive row crop weeds with dicamba and glufosinate applied alone, mixed, or sequentially. Weed Technol 36:733–739

Jones EAL, Owen MDK, Leon RG (2019) Influence of multiple herbicide resistance on growth in *Amaranthus tuberculatus*. Weed Res 59:235–244

Landau C, Bradley K, Burns E, DeWerff R, Dobbels A, Essman A, Flessner M, Gage K, Hager A, Jhala A, Johnson PO, Johnson W, Lancaster S, Lingenfelter D, Loux M, Miller E, Owen MDK, Sarangi D, Sikkema P, Sprague C, VanGessel M, Werle R, Young B, Williams M (2025) Weather and glufosinate efficacy; a retrospective analysis looking forward to the changing climate. Weed Sci 73:e32

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

Meyer CJ, Norsworthy JK (2019) Influence of weed size on herbicide interactions for Enlist™ and Roundup Ready® Xtend® technologies. Weed Technol 33:569–577

Meyer CJ, Peter F, Norsworthy JK, Beffa R (2019) Uptake, translocation, and metabolism of glyphosate, glufosinate, and dicamba mixtures in *Echinochloa crus-galli* and *Amaranthus palmeri*. Pest Manag Sci 76:3078–3087

Moore KJ, Dixon PM (2015) Analysis of combined experiments revisited. Agron J 107:763–771

Ou J, Thompson CR, Stahlman PW, Bloedow N, Jungulam M (2018) Reduced translocation of glyphosate and dicamba in combination contributes to poor control of *Kochia scoparia*: Evidence of antagonism. Sci Rep 8:5330

Peterson MA, McMaster SA, Riechers DE, Skelton J, Stahlman PW (2016) 2,4-D past, present, and future: A review. Weed Technol 30:303–345

Renton M, Willse A, Aradhya C, Tyre A, Head G (2024) Simulated herbicide mixtures delay both specialist monogenic and generalist polygenic resistance evolution in weeds. Pest Manag Sci 80:5983–5994

Robinson AP, Simpson DM, Johnson WG (2012) Summer annual weed control with 2,4-D and glyphosate. Weed Technol 26:657–660

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

Shurtlee JL, Coble HD (1985) Interference of certain broadleaf weed species in soybean (*Glycine max*). Weed Sci 33:654–657

Shyam C, Chahal PS, Jhala AJ, Jugulam M (2021) Management of glyphosate-resistant Palmer amaranth (*Amaranthus palmeri*) in 2,4-D-, glufosinate-, and glyphosate-resistant soybean. Weed Technol 35:136–143

Singh N, Peters TJ, Miller RP, Naeve SL, Sarangi D (2024) Profile and extent of herbicide-resistant waterhemp (*Amaranthus tuberculatus*) in Minnesota. Weed Sci 72:673–682

Staniforth DW (1965) Competitive effects of three foxtail species on soybeans. Weeds 13:191–193

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

Stoller EW, Woolley JT (1985) Competition for light by broadleaf weeds in soybeans (*Glycine max*). Weed Sci 33:199–202

Takano HK, Beffa R, Preston C, Westra P, Dayan FE (2020) A novel insight into the mechanism of action of glufosinate: how reactive oxygen species are formed. Photosynth Res 144:361–372

https://doi.org/10.1017/wet.2025.10053 Published online by Cambridge University Press