

# Impact of cereal rye cover crop on the fate of preemergence herbicides flumioxazin and pyroxasulfone and control of *Amaranthus* spp. in soybean

## Research Article

**Cite this article:** Nunes J, Arneson N.J, Wallace J, Gage K, Miller E, Lancaster S, Mueller T, Werle R (2023) Impact of cereal rye cover crop on the fate of preemergence herbicides flumioxazin and pyroxasulfone and control of *Amaranthus* spp. in soybean. *Weed Sci.* **71**: 493–505. doi: [10.1017/wsc.2023.46](https://doi.org/10.1017/wsc.2023.46)

Received: 13 June 2023

Revised: 4 August 2023

Accepted: 9 August 2023

First published online: 18 October 2023

### Associate Editor:

William Vencill, University of Georgia

### Keywords:

Herbicide interception; integrated weed management; *Secale cereale*; soil-applied herbicides; weed control

### Corresponding author:

Rodrigo Werle; Email: [rwerle@wisc.edu](mailto:rwerle@wisc.edu)

Jose Nunes<sup>1</sup> , Nicholas J. Arneson<sup>2</sup>, John Wallace<sup>3</sup>, Karla Gage<sup>4</sup>, Eric Miller<sup>5</sup>, Sarah Lancaster<sup>6</sup>, Thomas Mueller<sup>7</sup> and Rodrigo Werle<sup>8</sup> 

<sup>1</sup>Graduate Student, Department of Agronomy, University of Wisconsin, Madison, WI, USA; <sup>2</sup>Outreach Program Manager, Department of Agronomy, University of Wisconsin, Madison, WI, USA; <sup>3</sup>Assistant Professor, Department of Plant Science, Penn State University, University Park, PA, USA; <sup>4</sup>Associate Professor, School of Agricultural Sciences/School of Biological Sciences, Southern Illinois University Carbondale, Carbondale, IL, USA; <sup>5</sup>Assistant Scientist, School of Agricultural Sciences, Southern Illinois University Carbondale, Carbondale, IL, USA; <sup>6</sup>Assistant Professor and Extension Weed Specialist, Department of Agronomy, Kansas State University, Manhattan, KS, USA; <sup>7</sup>Professor Department of Plant Sciences, University of Tennessee, Knoxville, TN, USA and <sup>8</sup>Assistant Professor, Department of Agronomy, University of Wisconsin, Madison, WI, USA

### Abstract

Preemergence herbicides associated with cereal rye (*Secale cereale* L.) cover crop (hereafter “cereal rye”) can be an effective waterhemp [*Amaranthus tuberculatus* (Moq.) Sauer.] and Palmer amaranth (*Amaranthus palmeri* S. Watson) management strategy in soybean [*Glycine max* (L.) Merr.] production. Delaying cereal rye termination until soybean planting (planting green) optimizes biomass production and weed suppression but might further impact the fate of preemergence herbicides. Limited research is available on the fate of preemergence herbicides applied over living cereal rye in the planting green system. Field experiments were conducted in Illinois, Kansas, Pennsylvania, and Wisconsin to evaluate the fate of flumioxazin and pyroxasulfone and *Amaranthus* spp. residual control under different cover crop management practices in soybean in 2021 and 2022 (8 site-years). A flumioxazin + pyroxasulfone herbicide premix was applied preemergence at soybean planting under no-till without cereal rye, cereal rye early terminated before soybean planting, and cereal rye terminated at soybean planting. Flumioxazin and pyroxasulfone concentrations in the soil were quantified at 0, 7, and 21 d after treatment (DAT), and *Amaranthus* spp. density was determined at postemergence herbicide application. The presence of cereal rye biomass intercepted flumioxazin and pyroxasulfone at preemergence application and reduced concentration in the soil when compared with no-till, mainly at 0 DAT. Main differences in herbicide concentration were observed between no-till and cereal rye treatments rather than cereal rye termination times. Despite reducing herbicide concentration in the soil, the presence of the cereal rye biomass did not affect early-season residual *Amaranthus* spp. control. The adoption of effective preemergence herbicides associated with a properly managed cereal rye cover crop is an effective option for integrated *Amaranthus* spp. management programs in soybean production systems.

© University of Wisconsin-Madison and the Author(s), 2023. Published by Cambridge University Press on behalf of the Weed Science Society of America. This is an Open Access article, distributed under the terms of the Creative Commons Attribution-NonCommercial-NoDerivatives licence (<http://creativecommons.org/licenses/by-nc-nd/4.0/>), which permits non-commercial re-use, distribution, and reproduction in any medium, provided that no alterations are made and the original article is properly cited. The written permission of Cambridge University Press must be obtained prior to any commercial use and/or adaptation of the article.



### Introduction

The use of preemergence herbicides in combination with cereal rye (*Secale cereale* L.) cover crop (hereafter referred to as “cereal rye”) can be an effective weed management strategy in soybean [*Glycine max* (L.) Merr.] production systems (Bish et al. 2021; Cornelius and Bradley 2017; Perkins et al. 2021). Delaying cereal rye termination until soybean planting (i.e., planting green) is a practice that aims to increase cereal rye biomass production for weed suppression and ecosystem services in temperate regions where low growing degree-day accumulation is a limiting factor for cereal rye growth in the fall and spring (Fisher and Sprague 2022; Reed et al. 2019; Schramski et al. 2021). Applying preemergence herbicides over living cereal rye biomass in the planting green system is a new practice for which limited information is available regarding the fate of preemergence herbicides, whereas the most relevant research has been conducted using dead crop residue (Crutchfield et al. 1985; Erbach and Lovely 1975; Schmitz et al. 2001; Selim et al. 2003). While delaying cereal rye termination until soybean planting can increase biomass accumulation and improve weed suppression (Bish et al. 2021), greater biomass accumulation can also increase the interception of preemergence herbicides (Nunes et al. 2023) and potentially reduce preemergence weed control levels (Sperry et al. 2022).



Waterhemp [*Amaranthus tuberculatus* (Moq.) Sauer.] and Palmer amaranth (*Amaranthus palmeri* S. Watson), hereafter referred to as *Amaranthus* spp., are two of the most troublesome weed species in row-crop production systems in the U.S. Midwest (Steckel 2007; Van Wychen 2022). These dieocious *Amaranthus* species are highly competitive weeds (Steckel 2007) and prolific seed producers (Schwartz et al. 2016) and have rapidly evolved resistance to the main postemergence herbicides (i.e., glyphosate and imazethapyr) adopted for their management in soybean (Faleco et al. 2022; Heap 2023; Peterson et al. 2018). After the release of glyphosate-resistant crops in the late 1990s, farmers reduced the use of preemergence herbicides due to the excellent postemergence efficacy of glyphosate for controlling weeds such as *Amaranthus* spp. (Duke and Powles 2008; Green 2014). However, after years of reliance on glyphosate, herbicide-resistance cases have increased dramatically (Faleco et al. 2022; Heap 2023; Peterson et al. 2018), and the use of preemergence herbicides has again become an important tool for weed management programs in soybean (Beckie et al. 2019; Perkins et al. 2021).

Flumioxazin and pyroxasulfone are commonly used preemergence herbicides for weed control in soybean due to their efficacy in controlling *Amaranthus* spp. (Ferrier et al. 2022; Johnson et al. 2012; Mahoney et al. 2014; Perkins et al. 2021). Flumioxazin (Valor SX® herbicide, Valent U.S.A., Walnut Creek, CA) (Anonymous 2016) is a protoporphyrinogen oxidase (PPO)-inhibiting herbicide (WSSA Group 14) that was first registered in the United States in 2002 for preemergence control of broadleaf weed species in soybean and peanut (*Arachis hypogaea* L.) (Shaner 2014; USEPA 2023). Flumioxazin is considered a low environmental risk herbicide due to its short half-life and reduced soil persistence after application (Alister et al. 2008; Eason et al. 2022). Pyroxasulfone (i.e., Zidua® herbicide, BASF, Research Triangle Park, NC) (Anonymous 2022) is a very-long-chain fatty-acid (VLCFA)-inhibiting herbicide (WSSA Group 15) that was first registered in the United States in 2012 for preemergence control of grasses and small-seeded broadleaves in corn (*Zea mays* L.), wheat (*Triticum aestivum* L.), and soybeans (Shaner 2014; USEPA 2023). Pyroxasulfone can be used at lower field rates than other VLCFA-inhibiting herbicides, giving it a lower risk of environmental contamination due to its physicochemical properties (Nakatani et al. 2016; Westra et al. 2014; Yamaji et al. 2016). A premix of flumioxazin and pyroxasulfone (Fierce EZ® herbicide, Valent U.S.A.) (Anonymous 2019) is registered for residual weed control in soybean when applied early preplant or preemergence (within 3 d of soybean planting) at rates of 70.4 to 88.0 g ha<sup>-1</sup> of flumioxazin and 88.0 to 111.7 g ha<sup>-1</sup> of pyroxasulfone (Anonymous 2019).

The combination of cereal rye and preemergence herbicides is recommended for more diversified cropping and weed management systems (Bunchek et al. 2020; Perkins et al. 2021; Whalen et al. 2020). However, the main concern when applying preemergence herbicides to cereal rye is the interception of the spray solution, which can reduce herbicide concentration in the soil. Nunes et al. (2023) observed that the increase in cereal rye biomass at the time of preemergence application reduced S-metolachlor and sulfentrazone concentrations in the soil 28 d after application compared with tillage in the planting green system. Whalen et al. (2020) reported that delaying cover crop termination (21 vs. 7 d before soybean planting) increased cover crop biomass accumulation, which further reduced sulfentrazone concentration in the soil over time (0 to 84 d after application) compared with tillage. Despite investigating the impacts of cover

crops on the fate of preemergence herbicides, Nunes et al. (2023) did not quantify S-metolachlor and sulfentrazone concentrations in the soil over time, Whalen et al. (2020) terminated the cover crops and applied sulfentrazone before soybean planting, and neither Nunes et al. (2023) nor Whalen et al. (2020) compared early cereal rye termination versus termination at soybean planting (planting green) on preemergence herbicide fate. Hence, research is still needed to investigate whether spraying preemergence herbicides during cereal rye termination in the planting green system can have further impacts on herbicide fate compared with cereal rye early terminated. The objectives of this study were to investigate the impacts of cover crop management on the fate of flumioxazin and pyroxasulfone by quantifying their concentrations in the soil over time and to determine whether the potential impacts of cover crop management on herbicide concentration in the soil can affect early-season *Amaranthus* spp. control.

## Materials and Methods

Field studies were conducted near Brooklyn, WI (hereafter “Wisconsin”; 42.87°N, 89.39°W), Carbondale, IL (hereafter “Illinois”; 37.70°N, 89.24°W), Rock Springs, PA (hereafter “Pennsylvania”; 40.72°N, 77.94°W), and Rossville, KS (hereafter “Kansas”; 39.12°N, 95.92°W) during the 2021 and 2022 growing seasons (8 site-years). The study was conducted following a randomized complete block design with a treatment arrangement adapted from a split-plot. The main plots were cover crop management (no-till without cover crop, hereafter “no-till”; no-till with cereal rye early-terminated, hereafter “cereal rye early-term”; and no-till with cereal rye planting green, hereafter “cereal rye plant-green”) applied as strips (two 37.6 by 3 m strips for each cover crop management) in the experimental area. The split-plot was the use of a preemergence herbicide treatment (yes and no preemergence) that was randomly applied to one of the two plots of each cover crop management within each block (Supplementary Figure S1). The cover crop management was applied as strips to facilitate cereal rye establishment in the fall. Instead of attributing cover crop management to adjacent strips, they were randomized within the experimental area to improve the distribution of treatments across the field. Thus, the reason for this design being adapted from a split-plot treatment arrangement (e.g., instead of having the two split-plots side by side within each block, they were randomized in the experimental area). Unlike a split-plot arrangement, where the split-plot is nested within the main plot, it was not assumed that the preemergence plots (split-plot) had any degree of dependence from each cover crop management (main plot). Experimental units consisted of 3 by 9.1 m plots (four soybean rows with 76-cm row spacing) replicated four times. Soil properties from each experimental site and year are described in Table 1.

## Study Establishment and Herbicide Applications

The study was initiated in the previous fall of each experimental year by no-till drilling the cereal rye after corn harvest at a seeding rate of 67 kg ha<sup>-1</sup>, 19-cm row spacing, and a seeding depth of 3.2 cm. The cover crop was chemically terminated in the following spring with glyphosate at 1,262 g ae ha<sup>-1</sup> (Roundup PowerMax®, Bayer CropScience, St Louis, MO) and ammonium sulfate at 2,200 g ha<sup>-1</sup> applied 5 to 15 d before soybean planting for the early termination and within 1 d of soybean planting for the planting green treatment (Table 2). To eliminate established glyphosate-resistant weeds present at the time of planting in the experimental area, glyphosate was tank mixed with glufosinate at 655 g ai ha<sup>-1</sup>

**Table 1.** Soil properties and texture for all sites in 2021 and 2022.

Site	Year	Soil properties					Soil texture
		OM <sup>a</sup>	Sand	Silt	Clay	pH	
			%			H <sub>2</sub> O	
Illinois	2021	2.5	8.0	77.0	15.0	6.5	Silt loam
	2022	1.9	3.0	78.0	19.0	6.1	Silt loam
Kansas	2021	1.2	76.0	16.0	8.0	6.4	Sandy loam
	2022	1.7	40.0	50.0	10.0	5.5	Silt loam
Pennsylvania	2021	2.0	9.2	56.7	34.1	7.0	Silty clay loam
	2022	2.0	23.2	44.5	32.3	7.0	Clay loam
Wisconsin	2021	1.7	40.0	42.0	18.0	7.0	Loam
	2022	1.6	48.0	37.0	15.0	7.1	Loam

<sup>a</sup>OM, organic matter.

**Table 2.** Cereal rye and soybean planting and cereal rye termination dates for all sites in 2021 and 2022.

Site	Year	Planting date		Cereal rye termination	
		Cereal rye <sup>a</sup>	Soybean <sup>b</sup>	Early termination	Planting green
Illinois	2021	October 2, 2020	May 17, 2021	May 7, 2021	May 17, 2021
	2022	October 7, 2021	May 13, 2022	April 28, 2022	May 12, 2022
Kansas	2021	September 24, 2020	May 25, 2021	May 10, 2021	May 25, 2022
	2022	October 18, 2021	May 20, 2022	May 10, 2022	May 20, 2022
Pennsylvania	2021	October 1, 2020	May 18, 2021	May 4, 2021	May 19, 2021
	2022	October 20, 2021	May 24, 2022	May 12, 2022	May 25, 2022
Wisconsin	2021	September 25, 2020	May 18, 2021	May 7, 2021	May 17, 2021
	2022	September 23, 2021	May 24, 2022	May 12, 2022	May 25, 2022

<sup>a</sup>Cereal rye: 'VNS' (Illinois, 2021), 'Guardian' (Illinois, 2022), 'Rymin' (Kansas, 2021 and 2022), and 'Aroostook' (Pennsylvania and Wisconsin, 2021 and 2022), all seeded at 67.3 kg ha<sup>-1</sup>.

<sup>b</sup>Soybean: 'NKS39-E3' (Illinois, 2021), 'XO3861E' (Illinois, 2022), 'P39T61SE' (Kansas, 2021), 'B392EE' (Kansas, 2022), 'IS234E3' (Pennsylvania, 2021), '7280E' (Pennsylvania, 2022), and 'S20-E3' (Wisconsin, 2021 and 2022), all seeded at 346,020 seeds ha<sup>-1</sup>, except for Illinois, seeded at 370,736 seeds ha<sup>-1</sup> in 2022, and Pennsylvania, seeded at 444,883 seeds ha<sup>-1</sup> in both years.

(Liberty<sup>®</sup>, BASF) as part of a standard burndown treatment applied to all treatments (no-till, cereal rye early-term, and cereal rye plant-green) at the planting green termination. For the preemergence treatments, flumioxazin at 70.4 g ai ha<sup>-1</sup> + pyroxasulfone at 89.3 g ai ha<sup>-1</sup> (Fierce EZ<sup>®</sup>, Valent U.S.A.) was included in the spray mix containing glyphosate and glufosinate and sprayed at the planting green termination time (Table 2).

Postemergence herbicide application within a treatment was triggered when 20% of *Amaranthus* spp. plants reached 10 cm in height, similar to the Perkins et al. (2021) study, which evaluated the number of days until *Amaranthus* spp. reached 10 cm in height as the plant height limit for postemergence control with most herbicides available for soybean. Because postemergence applications were triggered by *Amaranthus* spp. height, applications varied across treatments within each site-year with a minimum of 27 and maximum of 57 d after soybean planting (Supplementary Table S1). The postemergence herbicide treatment was composed of glufosinate at 655 g ai ha<sup>-1</sup> (Liberty<sup>®</sup>, BASF), 2,4-D at 1,095 g ae ha<sup>-1</sup> (Enlist One<sup>®</sup>, Corteva Agriscience, Indianapolis, IN), clethodim at 102 g ai ha<sup>-1</sup> (Select Max<sup>®</sup>, Valent U.S.A.), acetochlor at 1,261 g ai ha<sup>-1</sup> (Warrant<sup>®</sup>, Bayer CropScience, St Louis, MO), and ammonium sulfate at 1% v/v. All herbicide applications (cereal rye termination, preemergence application, and postemergence) were delivered with a CO<sub>2</sub>-pressurized backpack sprayer equipped with a 3-m handheld boom fit with six nozzles (TTI 110015 for cereal rye termination and preemergence application, and AIXR 110015 for postemergence application [TeeJet<sup>®</sup> Technologies, Wheaton, IL]) on 50-cm spacing calibrated to deliver 140 L ha<sup>-1</sup> of spray solution. Soybean was planted using a four-row no-till planter adjusted to

place seeds at 2.5-cm depth on 76-cm row spacing. The soybean variety and seeding rate varied across site-years (Table 2).

### Soil Sampling and Herbicide Analyses

Soil samples were collected from the treatments no-till without preemergence (standard check) and the treatments no-till, cereal rye early-term, and cereal rye plant-green with preemergence at 0, 7, and 21 d after treatment (DAT) to quantify flumioxazin and pyroxasulfone concentrations in the soil. Soil sampling, processing, and analysis practices were executed as recommended by Mueller and Senseman (2015). Three subsamples (soil cores 0- to 7.6-cm depth by 10-cm diameter) were collected from each plot with a golf cup cutter (Par Aide Products, St Paul, MN), placed in plastic bags that were then sealed, and immediately placed in a cooler before being stored in a freezer (-10 C). The samples collected in Illinois, Kansas, and Pennsylvania were frozen and shipped overnight to Wisconsin, where they were stored frozen until the beginning of the soil extraction process.

Soil samples were thawed at room temperature for 60 min and thoroughly homogenized manually while still inside each bag. A 15 ± 0.5 g subsample of homogenized soil from each sample was placed into a 50-ml conical polypropylene tube (Fisher Scientific, Pittsburgh, PA), and 30 ml of methanol (RPI Research Products, Mt Prospect, IL) was added to the soil. Tubes were sealed and shaken in a horizontal reciprocating shaker for 14 h. After shaking, the tubes were allowed to equilibrate statically for 30 min to separate the methanol solution from soil particles. Five milliliters of the solution were extracted from the tubes with a 10-ml syringe, and 2 ml were transferred to a liquid chromatography/mass

spectrometry vial passing through a 0.45- $\mu\text{m}$  filter (Fisher Scientific). Vials were kept in the freezer until overnight shipment in an insulated container with dry ice to the Department of Plant Sciences at the University of Tennessee in Knoxville, TN, for analysis. Tubes with soil and remaining solution were dried to constant weight at 65 C to determine the dry soil weight of each tube for herbicide quantification. Samples were analyzed using an Agilent 1260 Liquid Chromatograph coupled with a 6470 Mass Spectrometry detector. Analysis details included the use of a phenyl-hexyl analytical column and a gradient mobile phase of acetonitrile and water, both fortified with 0.1% formic acid. Parent and confirmatory ions for pyroxa-sulfone were 392.1 and 229/179.1 and for flumioxazin were 355.1 and 327.1/299.1, respectively. The limit of detection for both herbicides was 1.0 ng g<sup>-1</sup> dry weight soil, and recoveries were >85% for all soils and were not corrected for recovery.

### Cereal Rye Biomass, *Amaranthus* spp., and Weather Data Collection

Aboveground cereal rye biomass was determined at each termination time by clipping the plants at the soil surface in three 0.1-m<sup>-2</sup> quadrats randomly placed in each plot. Biomass samples were placed in paper bags and dried at 65 C for 7 d to determine cereal rye biomass in Mg ha<sup>-1</sup>. *Amaranthus* spp. density (plants m<sup>-2</sup>) data were collected at the time of postemergence herbicide application by counting the number of emerged *Amaranthus* spp. plants in two 0.25-m<sup>-2</sup> quadrats randomly placed in each plot. *Amaranthus tuberculatus* was evaluated in both years in Illinois, Pennsylvania, and Wisconsin, and *A. palmeri* in both years in Kansas. *Amaranthus tuberculatus* and *A. palmeri* density data were combined for analysis due to the intrinsic similarities between the two species as well as their similar response to preemergence herbicides and the adoption of cereal rye for weed suppression (Palhano et al. 2018; Perkins et al. 2021; Steckel 2007; Webster et al. 2016).

Daily precipitation (mm) and minimum, maximum, and average air temperatures C from cereal rye planting to soybean harvest of each experimental year were collected from weather stations adjacent to the experimental areas. The temperature data were used to estimate daily growing degree days (temperature base 10 C) from preemergence application until the last soil sampling at 21 DAT using the following equation (Mirsky et al. 2011):

$$\text{GDD} = \frac{T_{\max} + T_{\min}}{2} - T_{\text{base}} \quad [1]$$

where  $T_{\max}$  and  $T_{\min}$  are the maximum and minimum daily temperatures, respectively, and  $T_{\text{base}}$  is the base temperature at which physiological activity and growth occur (set at 10 C). For mean temperatures less than  $T_{\text{base}}$ , the GDD value was assumed to be zero.

### Statistical Analyses

All statistical analyses were performed in R statistical software v. 4.2.1 (R Core Team 2022). Data processing, manipulation, and visualization were performed with the TIDYVERSE collection of packages (Wickham et al. 2019); specific details about other packages are provided in each of the following sections.

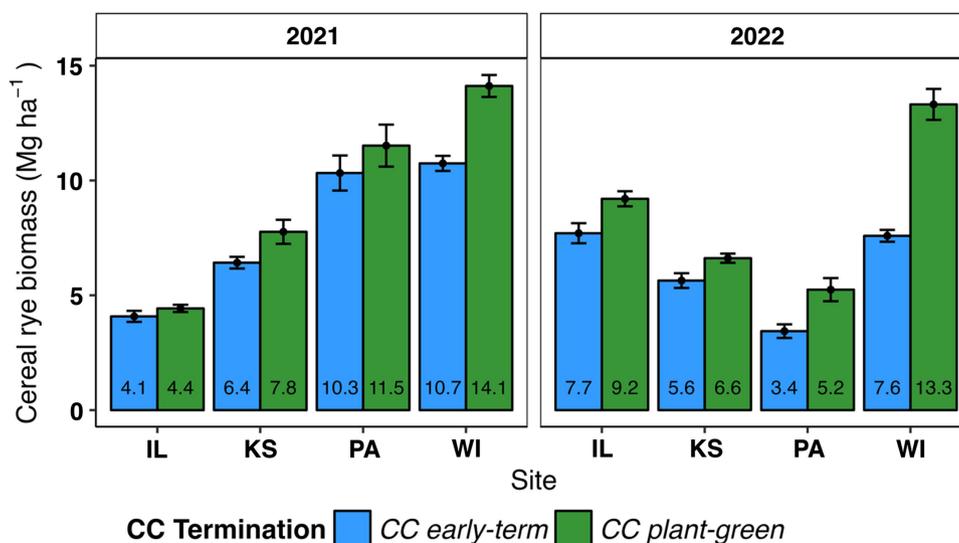
### Herbicide Concentration in the Soil

Flumioxazin and pyroxa-sulfone concentrations in the soil were analyzed independently by fitting separate generalized least-squares

models for each site and year using the *gls* function in R software (NLME package; Pinheiro et al. 2022). Generalized least-squares models are an alternative to deal with heteroscedasticity and/or correlation between errors in the data (Pinheiro and Bates 2000). In this study, because each plot was sampled three times, a continuous correlation structure was adopted to account for the serial correlation between sampling times. The continuous correlation structure is adapted from the first-order autoregressive model to accommodate unequal time points during events (Piepho and Edmondson 2018; Pinheiro and Bates 2000). Each model was composed of herbicide concentration in the soil as the dependent variable and cover crop management, sampling time, their interaction, and block as fixed effects. Model assumptions of normality and homoscedasticity were assessed by a visual inspection of residuals. To meet and improve such assumptions, herbicide concentration in soil was square-root transformed for all models. Fitted models were used to obtain estimated back-transformed marginal means using the *emmeans* function in R (EMMEANS package; Lenth 2022). When a significant interaction or main effect was observed (P-value  $\leq 0.05$ ), marginal means were separated using Fisher's LSD test at  $\alpha = 0.05$  (EMMEANS package; Lenth 2022).

Random forest models were performed to determine which variables and factors had the greatest influence on flumioxazin and pyroxa-sulfone concentrations in the soil at 0 and 21 DAT. Random forest is a machine learning algorithm that combines multiple decision trees using a subsample of bootstrapped observations from randomly selected explanatory variables to improve prediction accuracy and reduce overfitting. In random forest, multiple decision trees are trained on different subsets of the training data, with each tree making a prediction independently. The predictions of all the trees are then combined to produce the final prediction (Biau and Scornet 2016; Breiman 2001). Random forest is a powerful algorithm for the classification of feature importance in complex data sets. In weed science, Hartway et al. (2022) used random forest to determine which plant traits are associated with herbicide resistance, Striegel et al. (2021) classified which factors affected dicamba spray solution pH when mixed with other herbicides, and Baraibar et al. (2018) identified the most important variables explaining weed biomass in cover crops.

Separate models were fit to the flumioxazin and pyroxa-sulfone data using the herbicide concentration in the soil (ng g<sup>-1</sup> soil) at 0 DAT as the continuous dependent variable and site (Illinois, Kansas, Pennsylvania, and Wisconsin), year (2021 and 2022), cover crop management (no-till, cereal rye early-term, and cereal rye plant-green), and percent of soil organic matter, clay, and silt of each site as explanatory variables. As for the flumioxazin and pyroxa-sulfone models using the 21 DAT data as the continuous dependent variable, besides the aforementioned explanatory variables, accumulated precipitation (mm) and GDD ( $T_{\text{base}}$  10 C) from preemergence application until the last soil sampling time (21 DAT) were included as additional explanatory variables. For all models, *ntree* (the number of decision trees) was set to 1,000, and *mtry* (the number of different predictors sampled at each split) and *min\_n* (the minimum number of data points in a node required for additional splits) were tuned during model training and set according to the model with lowest root-mean-square error. Variable importance (VI) scores were determined by the permutation measure, which provides an estimate of the change in prediction accuracy should the variable be excluded from the model (Wright 2023). Higher VI values indicate the explanatory variable is important in the model and in explaining the variability of the dependent variable, whereas values near zero indicate the



**Figure 1.** Cereal rye cover crop dry biomass (Mg ha<sup>-1</sup>) at each termination time in all sites in 2021 and 2022. Error bars indicate the standard error of means. IL, Illinois; KS, Kansas; PA, Pennsylvania; and WI, Wisconsin. Cereal rye early terminated before soybean planting (early-term) and cereal rye terminated at soybean planting (plant-green).

explanatory variable is not important (Bourgoin et al. 2018; Louppe et al. 2013). All models were fit using the R meta package TIDYMODELS following the framework and workflow presented in Kuhn and Silge (2022). A scatter plot with the relationship between cereal rye biomass (Mg ha<sup>-1</sup>) at termination and flumioxazin and pyroxasulfone concentrations in the soil (ng g<sup>-1</sup> soil) at 0 DAT was also created to illustrate the effect of cereal rye biomass on herbicide in the soil at preemergence application. The data were plotted with the *ggplot* function, and a linear regression line was added using the *geom\_smooth* function in R (GGPLOT2 package; Wickham 2016).

Flumioxazin and pyroxasulfone concentrations in the soil (ng g<sup>-1</sup> soil) at 21 DAT were used to estimate the percent of herbicide dissipation based on the initial concentration in the no-till treatment at 0 DAT using the following equation:

$$\text{Dissipation (\%)} = 100 - \left( \frac{HC21 * 100}{NTavg0} \right) \quad [2]$$

where dissipation (%) is the percent of herbicide loss or reduction within 21 d, *HC21* is the herbicide concentration (ng g<sup>-1</sup> soil) of an individual plot at 21 DAT, and *NTavg0* is the average of the four no-till observations at 0 DAT. Because no-till without cereal rye was treated as a check in this study, its herbicide concentration at 0 DAT was utilized as the standard to estimate herbicide dissipation based on the assumption that it had the least herbicide interception during preemergence application. Individual generalized linear mixed models (*glmmTMB* function) with a beta distribution were fit to the herbicide dissipation data of each site, treating cover crop management as a fixed effect and block nested within year as random effects in the model (GLMMTMB package; Brooks et al. 2017). Fitted models were used to obtain estimated marginal means using the *emmeans* function in R (EMMEANS package; Lenth 2022). When a significant main effect was observed (P-value ≤ 0.05), marginal means were separated using Fisher's LSD test at α = 0.05 (EMMEANS package; Lenth 2022).

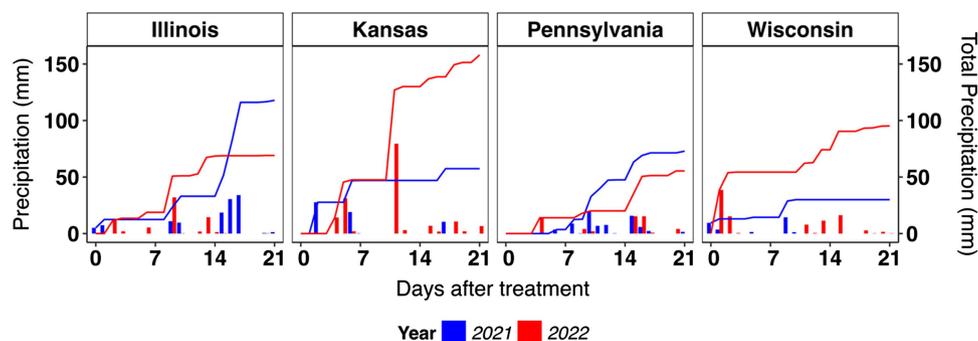
#### Amaranthus spp. Density

To analyze *Amaranthus* spp. density, the data set was first split into two groups based on the pressure in each site-year. A "low"

*Amaranthus* spp. pressure group was composed of Illinois 2022, Kansas 2021 and 2022, and Pennsylvania 2021 and 2022. These 5 site-years presented the lowest mean of *Amaranthus* spp. density (≤10 plants m<sup>-2</sup>) in the no-till without preemergence treatment across sites and years. Conversely, Illinois 2021 and Wisconsin 2021 and 2022 presented the highest *Amaranthus* spp. density (≥56 plants m<sup>-2</sup>) and were grouped as having "high" *Amaranthus* spp. pressure. The no-till without preemergence was selected as the treatment to evaluate *Amaranthus* spp. pressure, because it best represented the natural *Amaranthus* spp. infestation of each site-year, as it did not have the presence of a preemergence herbicide or cover crop weed control or suppression. Generalized linear mixed models (*glmmTMB* function) with a negative binomial distribution were fit to the *Amaranthus* spp. density data of each group treating cover crop management and preemergence and their interaction as fixed effects and block nested within site-year as a random effect in the model (GLMMTMB package; Brooks et al. 2017). The negative binomial distribution was adopted as recommended by Stroup (2015) to deal with count data that often do not meet the normality and equal variance assumptions of ANOVA. Fitted models were used to obtain estimated marginal means using the *emmeans* function in R (EMMEANS package; Lenth 2022). When a significant interaction or main effect was observed (P-value ≤ 0.05), marginal means were separated using Fisher's LSD test at α = 0.05 (EMMEANS package; Lenth 2022).

#### Results and Discussion

Delaying cereal rye termination until soybean planting led to greater cereal rye biomass accumulation compared with the early termination across all site-years. Overall, Wisconsin was the site with the greatest cereal rye biomass levels in both years, while Illinois 2021 and Pennsylvania 2022 were the 2 site-years with the least biomass production (Figure 1). Accumulated precipitation from preemergence application to the last soil sampling time at 21 DAT also varied across site-years. Wisconsin 2021 had the least total precipitation within the 21-d period (30 mm), whereas Kansas 2022 had the greatest (158 mm; Figure 2). Given such differences in cereal rye biomass accumulation, precipitation, and



**Figure 2.** Daily (bars) and total (lines) precipitation from preemergence herbicide application to 21 d after treatment (last soil sampling time) for all sites in 2021 and 2022. Total precipitation in 2021 and 2022 for each site, respectively: Illinois, 118 and 69 mm; Kansas, 57 and 158 mm; Pennsylvania, 73 and 55 mm; and Wisconsin, 30 and 95 mm. Refer to Supplementary Figure S4 for air temperature data of each site-year.

other site-specific factors that might have impacted herbicide concentration in the soil, flumioxazin and pyroxasulfone data were analyzed independently, and results are presented individually by site-year.

Soil samples from the no-till without preemergence were collected to ensure that the residual concentration of flumioxazin and pyroxasulfone was negligible in all areas where the study was conducted. The concentrations of flumioxazin and pyroxasulfone was  $\leq 1.1$  and  $2.7 \text{ ng g}^{-1}$  soil, respectively, in all 8 site-years of data (Supplementary Tables S2 and S3). Thus, this treatment was not part of the statistical analyses of the study. Flumioxazin and pyroxasulfone results are presented in Tables 3 and 4, respectively. For visualization of trends across sites and years, refer to Supplementary Figures S2 (flumioxazin) and S3 (pyroxasulfone) for a graphical presentation of the data.

### Illinois 2021 and 2022

Illinois presented the smallest concentration of flumioxazin and pyroxasulfone in the soil across site-years (Tables 3 and 4). In 2021, there was only an effect of sampling time ( $P$ -value  $< 0.001$ ) for both herbicides, given that their concentrations was highest at 0 DAT and decreased over time at a similar rate in all treatments. In 2022, there was an interaction between cover crop management and sampling time ( $P$ -value = 0.017 [flumioxazin] and = 0.001 [pyroxasulfone]) for both herbicides. The cereal rye treatments reduced flumioxazin and pyroxasulfone concentrations in the soil compared with no-till at 0 DAT. However, there were no differences between the two cereal rye termination times. Additionally, there were also no differences in the concentration of either herbicide in the soil at 7 and 21 DAT across cover crop management, only a decrease over time in their concentrations (Tables 3 and 4). The lack of difference between no-till and the cereal rye treatments in 2021 was likely due to the relatively low cereal rye biomass accumulated ( $\leq 4.4 \text{ Mg ha}^{-1}$ ) in both termination times in this year (Figure 1). Conversely, greater levels of cereal rye biomass ( $\geq 7.7 \text{ Mg ha}^{-1}$ ) were observed in 2022 for both termination times (Figure 1) and likely intercepted sufficient spray solution to reduce the concentration of both herbicides in both cover crop management treatments at 0 DAT (Nunes et al. 2023; Tables 3 and 4). In previous research, Nunes et al. (2023) and Whalen et al. (2020) observed an inverse relationship between cover crop biomass accumulation at termination and herbicide concentration in the soil after preemergence application. Such findings corroborate that cereal rye biomass accumulation at the time of preemergence application can have a direct impact on herbicide concentration in the soil over time.

### Kansas 2021 and 2022

Kansas was the only site where there were no interactions between cover crop management and sampling time for either flumioxazin or pyroxasulfone in both years ( $P$ -value  $> 0.05$ ; Tables 3 and 4). The main effects of cover crop management and sampling time were significant for both herbicides in 2021 and for flumioxazin in 2022, whereas only cover crop management ( $P$ -value  $< 0.001$ ) was significant for pyroxasulfone in 2022. Flumioxazin concentration was greatest at 0 DAT and decreased over time at similar rates in all cover crop management practices, with the smallest concentration at 21 DAT in both years. No-till resulted in the greatest flumioxazin concentration at all sampling times, and except for cereal rye early-term in 2021, the cereal rye treatments reduced flumioxazin concentration compared with no-till at all sampling times (Tables 3 and 4).

Pyroxasulfone behavior was different within and between years when compared with flumioxazin. In 2021, although the concentration was smallest at 21 DAT, the greatest concentration was observed at 7 DAT. As for 2022, pyroxasulfone concentration in the soil was similar for all sampling times. Thus, unlike flumioxazin, which showed a decreasing trend over time in both years, pyroxasulfone concentration increased from 0 to 7 DAT in 2021 and did not change over time in 2022 (Tables 3 and 4). This behavior suggests that pyroxasulfone was released from the crop residue (no-till and cereal rye biomass) to the soil with rainfall in 2021 and was likely more stable in the environment than flumioxazin when intercepted by crop residue. As for the effect of cover crop management, both cereal rye treatments reduced pyroxasulfone concentration compared with no-till at all sampling times in both years. Yet differences between early termination and planting green were only observed in 2021, when delaying cereal rye termination until soybean planting likely resulted in greater herbicide interception and, thus, lower pyroxasulfone concentration in the soil (Tables 3 and 4). The differences between cereal rye treatments in 2021 for pyroxasulfone are likely due to the greater cereal rye biomass accumulated and lower overall precipitation in 2021 compared with 2022 (Figures 1 and 2). Such factors can contribute to higher herbicide interception during application and lower herbicide wash-off from the cereal rye biomass (Nunes et al. 2023).

### Pennsylvania 2021 and 2022

Similar responses between flumioxazin and pyroxasulfone were observed in both years that the study was conducted in

**Table 3.** Flumioxazin concentration in the soil ( $\text{ng g}^{-1}$  soil) as a function of cover crop management at 0, 7, and 21 d after treatment (DAT) for all sites in 2021 and 2022.<sup>a</sup>

Cover crop management	Illinois											
	2021						2022					
	0 DAT		7 DAT		21 DAT		0 DAT		7 DAT		21 DAT	
	$\text{ng g}^{-1}$ soil											
No-till	19.7 a	A	5.4 a	B	1.8 a	C	20.3 a	A	6.0 a	B	1.2 a	C
Early-term	21.7 a	A	4.2 a	B	0.5 a	C	7.2 b	A	3.6 a	AB	1.0 a	B
Plant-green	18.2 a	A	3.6 a	B	1.6 a		7.6 b	A	3.5 a	AB	0.9 a	B
	P-values											
Cover crop							0.826					
DAT							<0.001					
Cover crop:DAT							0.017					
Cover crop management	Kansas											
	2021						2022					
	0 DAT		7 DAT		21 DAT		0 DAT		7 DAT		21 DAT	
	$\text{ng g}^{-1}$ soil											
No-till	36.2 a	A	16.1 a	A	6.8 a	B	33.2 a	A	18.3 a	B	6.2 a	C
Early-term	15.9 a	A	15.8 a	A	4.1 a	B	17.3 b	A	9.2 b	B	4.2 b	C
Plant-green	5.2 b	A	5.0 b	A	0.7 b	B	11.5 b	A	6.7 b	B	2.4 b	C
	P-values											
Cover crop							<0.001					
DAT							<0.001					
Cover crop:DAT							0.183					
Cover crop management	Pennsylvania											
	2021						2022					
	0 DAT		7 DAT		21 DAT		0 DAT		7 DAT		21 DAT	
	$\text{ng g}^{-1}$ soil											
No-till	66.4 a	A	33.8 a	B	11.2 a	C	20.1 a	A	17.3 a	B	5.8 a	C
Early-term	4.1 b	B	13.8 b	A	2.5 b	B	17.1 a	A	12.3 a	B	3.2 a	C
Plant-green	2.3 b	A	5.2 c	A	3.5 b	A	22.9 a	A	12.6 a	B	5.9 a	C
	P-values											
Cover crop							<0.001					
DAT							<0.001					
Cover crop:DAT							0.333					
Cover crop management	Wisconsin											
	2021						2022					
	0 DAT		7 DAT		21 DAT		0 DAT		7 DAT		21 DAT	
	$\text{ng g}^{-1}$ soil											
No-till	30.5 a	A	17.9 a	B	14.7 a	B	28.7 a	A	15.7 a	B	6.4 a	C
Early-term	10.7 b	A	10.1 a	A	7.8 a	A	11.0 b	A	11.6 ab	A	3.8 a	B
Plant-green	1.2 c	B	10.4 a	A	8.3 a	A	4.9 c	A	6.1 b	A	3.0 a	A
	P-values											
Cover crop							<0.001					
DAT							0.189					
Cover crop:DAT							<0.001					

<sup>a</sup>Means followed by common lowercase letters indicate no statistical difference between cover crop management within DAT, and common uppercase letters indicate no statistical difference between DAT within cover crop management by the LSD test ( $\alpha = 0.05$ ).

Pennsylvania. An interaction ( $P$ -value  $< 0.001$ ) between cover crop management and sampling time was detected for both herbicides in 2021 (Tables 3 and 4). The large amount of cereal rye biomass accumulated in 2021 ( $\geq 10.3 \text{ Mg ha}^{-1}$ ) likely led to a substantial interception of both herbicides during application and a reduction in the concentration of flumioxazin and pyroxasulfone at all sampling times compared with no-till in 2021. Moreover, the concentration of both herbicides was greatest at 0 DAT and least at 21 DAT in the no-till. However, in the cereal rye early-term treatment, the highest flumioxazin and pyroxasulfone concentrations were observed at 7 DAT, and no differences across

sampling times were observed for cereal rye plant-green in 2021 (Tables 3 and 4). This indicates that despite being intercepted by the cereal rye biomass, the soil-applied herbicides were likely released from the crop residue to the soil after application in the early termination at a greater extent than in the termination at soybean planting. In fact, 7 DAT was the only sampling time that a difference between cereal rye early-term and cereal rye plant-green was observed for both herbicides in 2021 (Tables 3 and 4). Because cereal rye biomass accumulation was high for both cereal rye termination times ( $\geq 10.3 \text{ Mg ha}^{-1}$ ) in 2021, we believe that at this level of biomass, there were no differences in the interception of the

**Table 4.** Pyroxasulfone concentration in the soil ( $\text{ng g}^{-1}$  soil) as a function of cover crop management at 0, 7, and 21 d after treatment (DAT) for all sites in 2021 and 2022.<sup>a</sup>

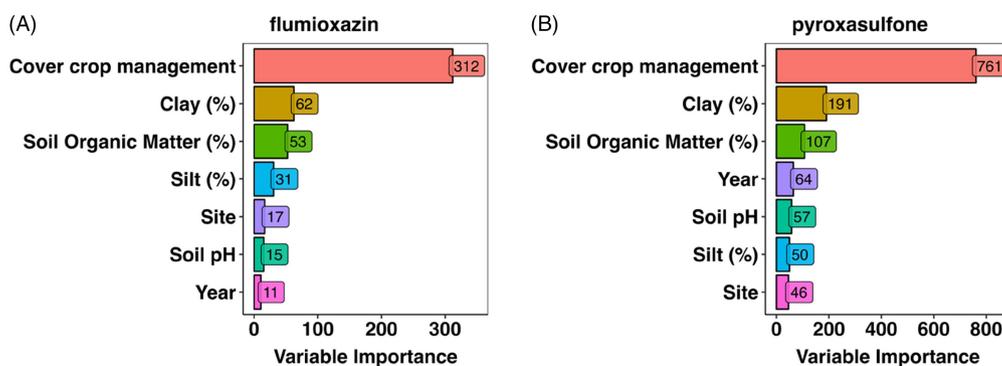
Cover crop management	Illinois											
	2021						2022					
	0 DAT		7 DAT		21 DAT		0 DAT		7 DAT		21 DAT	
	$\text{ng g}^{-1}$ soil											
No-till	25.7 a	A	8.0 a	B	2.4 a	C	31.0 a	A	11.6 a	B	5.2 a	C
Early-term	30.3 a	A	7.3 a	B	2.0 a	C	11.0 b	A	9.0 a	AB	4.4 a	B
Plant-green	28.3 a	A	9.0 a	B	2.2 a	C	12.2 b	A	12.8 a	A	4.1 a	B
	P-values											
Cover crop							0.813					
DAT							<0.001					
Cover crop:DAT							0.968					
	Kansas											
	2021						2022					
	0 DAT		7 DAT		21 DAT		0 DAT		7 DAT		21 DAT	
	$\text{ng g}^{-1}$ soil											
No-till	53.0 a	B	61.5 a	A	30.4 a	C	50.8 a	A	42.4 a	A	25.1 a	A
Early-term	17.1 b	B	40.4 b	A	12.0 b	C	24.9 b	A	36.7 b	A	15.0 b	A
Plant-green	9.3 c	B	11.5 c	A	5.0 c	C	16.1 b	A	21.4 b	A	10.9 b	A
	P-values											
Cover crop							<0.001					
DAT							0.011					
Cover crop:DAT							0.406					
	Pennsylvania											
	2021						2022					
	0 DAT		7 DAT		21 DAT		0 DAT		7 DAT		21 DAT	
	$\text{ng g}^{-1}$ soil											
No-till	96.5 a	A	72.8 a	A	45.5 a	B	27.0 a	A	34.8 a	A	22.6 a	A
Early-term	6.0 b	B	23.4 b	A	8.8 b	B	21.1 a	A	26.2 a	A	15.9 a	A
Plant-green	4.8 b	A	12.2 c	A	7.9 b	A	28.5 a	A	22.1 a	A	14.3 a	A
	P-values											
Cover crop							<0.001					
DAT							0.004					
Cover crop:DAT							<0.001					
	Wisconsin											
	2021						2022					
	0 DAT		7 DAT		21 DAT		0 DAT		7 DAT		21 DAT	
	$\text{ng g}^{-1}$ soil											
No-till	64.2 a	A	48.1 a	B	49.4 a	AB	31.8 a	A	26.2 a	A	12.1 a	B
Early-term	15.7 b	B	30.4 b	A	20.1 b	B	12.9 b	B	19.1 a	A	9.0 a	B
Plant-green	3.6 c	B	18.9 b	A	22.1 b	A	5.9 c	AB	9.3 b	A	3.1 b	B
	P-values											
Cover crop							<0.001					
DAT							<0.001					
Cover crop:DAT							<0.001					

<sup>a</sup>Means followed by common lowercase letters indicate no statistical difference between cover crop management within DAT, and common uppercase letters indicate no statistical difference between DAT within cover crop management by the LSD test ( $\alpha = 0.05$ ).

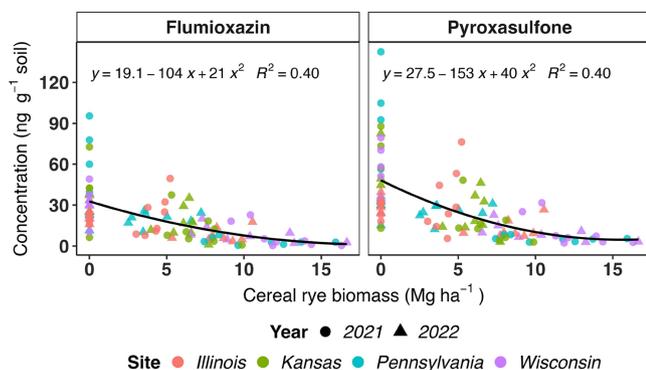
preemergence herbicides during application. Hence, no differences between cereal rye early-term and cereal rye plant-green at 0 DAT. Nevertheless, the additional  $1.2 \text{ Mg ha}^{-1}$  of cereal rye biomass accumulated when terminated at soybean planting likely retained flumioxazin and pyroxasulfone to a greater extent than the early termination at 7 DAT. Moreover, it is unclear whether cereal rye can foliar absorb and metabolize part of the preemergence herbicides applied in the planting green system. If absorbed by the

living cereal rye, it can also contribute to lower flumioxazin and pyroxasulfone concentrations in the soil at 7 DAT (Tables 3 and 4).

Lower levels of cereal rye biomass ( $\leq 5.2 \text{ Mg ha}^{-1}$ ) were recorded for both termination times in 2022 compared with 2021 in Pennsylvania (Figure 1). Similar to Illinois 2021, when low cereal rye biomass was observed, there were no differences between cover crop management practices on the concentration of flumioxazin and pyroxasulfone at all sampling times. Only the effect of time was



**Figure 3.** Random forest variable importance for flumioxazin (A) and pyroxasulfone (B) concentration in the soil ( $\text{ng g}^{-1}$  soil) at 0 d after treatment. Root-mean-square error from each model: flumioxazin, 13.3; and pyroxasulfone, 18.7.



**Figure 4.** Response of flumioxazin and pyroxasulfone concentration in the soil ( $\text{ng g}^{-1}$  soil) at 0 d after application as a function of cereal rye cover crop biomass at termination.

significant for flumioxazin, given that its concentration was greatest at 0 DAT and lowest at 21 DAT. For pyroxasulfone, there were no differences across sampling times ( $P$ -value  $> 0.05$ ; Tables 3 and 4). The lack of differences between treatments is likely due to the low cereal rye biomass accumulation, which possibly resulted in minor herbicide interception during application.

#### Wisconsin 2021 and 2022

Wisconsin was the only site in which the cover crop management and sampling time interaction for flumioxazin and pyroxasulfone in both years was significant ( $P$ -value  $< 0.001$ ; Tables 3 and 4). Due to the high cereal rye biomass accumulated at this site ( $\geq 7.6 \text{ Mg ha}^{-1}$ ; Figure 1), the cereal rye treatments intercepted and reduced the concentration of both herbicides compared with no-till at 0 DAT in 2021 and 2022. Moreover, the higher cereal rye biomass accumulation by the cereal rye terminated at planting increased the interception of both herbicides and reduced their concentrations in the soil at 0 DAT in both years. For flumioxazin, despite the initial reduction due to spray solution interception by the cereal rye biomass, there were no differences between cover crop management practices in either year at 21 DAT. Conversely, for pyroxasulfone, only the cereal rye early-term in 2022 was similar to no-till at 21 DAT. As for the effect of sampling time, the overall concentration of both herbicides was highest at 0 DAT and decreased over time in the no-till. In contrast, for the cereal rye treatments, due to the initial herbicide interception, flumioxazin and pyroxasulfone concentrations were either constant or

increased with time, with the highest peaks observed at 7 DAT in most instances (Tables 3 and 4).

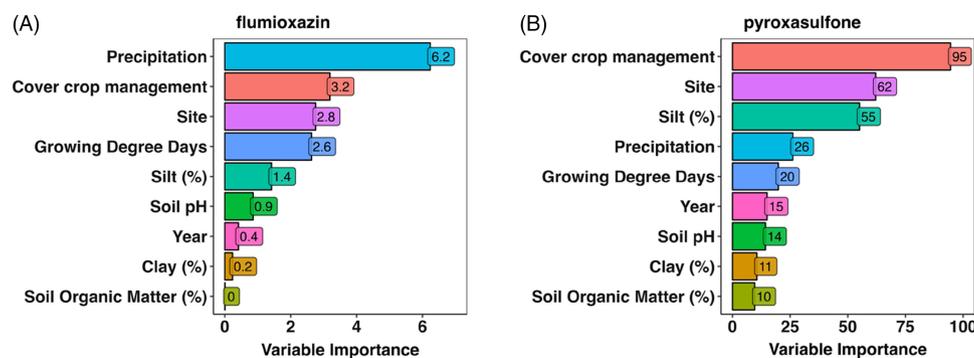
#### Random Forest for Herbicide Concentration in the Soil

Random forest models were fit to flumioxazin and pyroxasulfone data at 0 and 21 DAT to better understand which explanatory variables had the largest influence on herbicide concentration in the soil. Cover crop management was the most important factor explaining herbicide concentration at 0 DAT for both herbicides (Figure 3). This outcome was expected, given that herbicide concentration in the soil at 0 DAT had a negative linear relationship with the increase in cereal rye biomass at termination (Figure 4). Thus, although cereal rye biomass was not part of the random forest models, its importance was indirectly related to cover crop management. At 21 DAT, cover crop management remained the most important factor for pyroxasulfone response, followed by site and silt content (Figure 5). However, for flumioxazin, precipitation was the most important variable explaining its response, followed by cover crop management and site (Figure 5). The fact that cover crop management was replaced by precipitation as the most important variable explaining flumioxazin concentration in the soil at 21 DAT is an intriguing observation, given the importance of cover crop management in this study. One consideration about the two herbicides is that flumioxazin has a slightly lower water solubility ( $1.79 \text{ mg L}^{-1}$  at  $25 \text{ C}$ ) than pyroxasulfone ( $3.45 \text{ mg L}^{-1}$  at  $20 \text{ C}$ ; Shaner 2014). Thus, precipitation might be of more importance for flumioxazin movement from the cereal rye biomass to the soil as compared with pyroxasulfone.

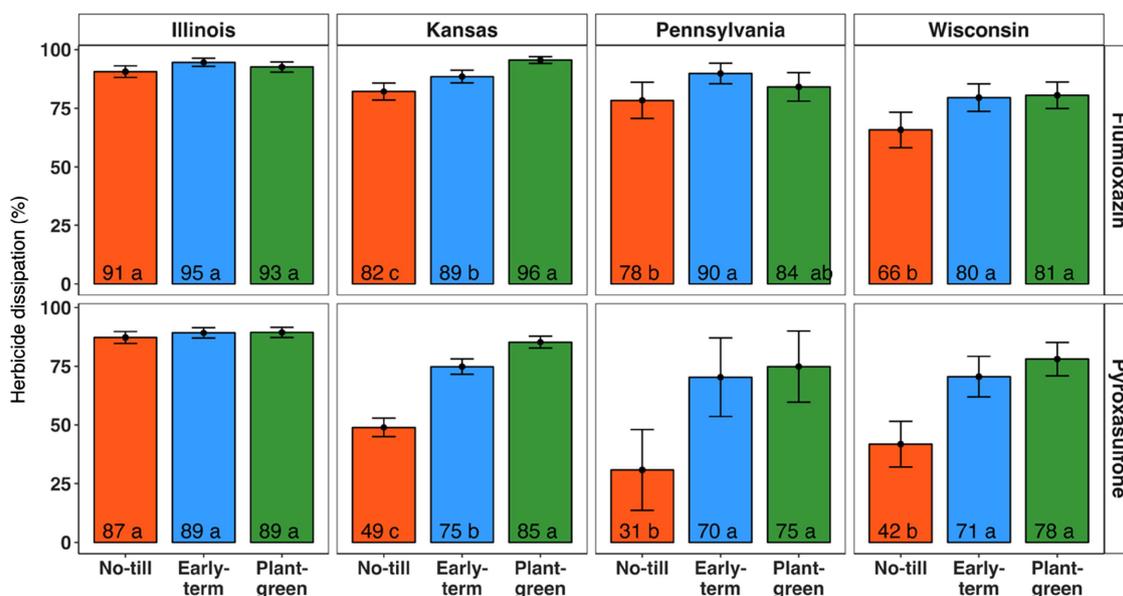
#### Herbicide Dissipation

The aim of calculating the degree of flumioxazin and pyroxasulfone dissipation was to estimate the effect of cover crop management practices on herbicide dissipation and which herbicide was subject to more dissipation within 21 DAT. To address these questions, the data were pooled across years for all sites to objectively focus on the effects of cover crop management practices and sites on herbicide dissipation.

Illinois presented the highest herbicide dissipation across all sites, with about 90% of flumioxazin and pyroxasulfone dissipated at 21 DAT, and was the only site with no differences in herbicide dissipation across cover crop management practices (Figure 6). The lack of differences between cover crop management practices in herbicide dissipation is likely due to low herbicide interception



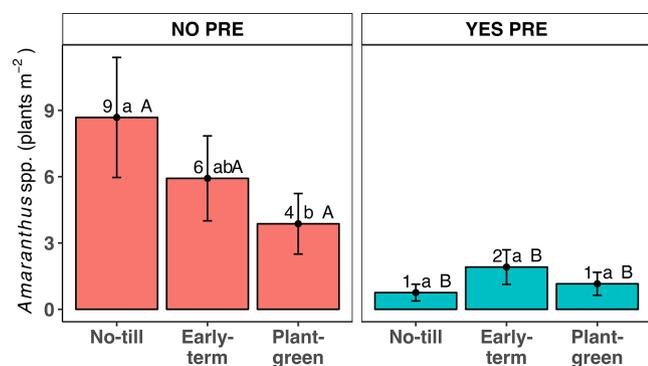
**Figure 5.** Random forest variable importance for flumioxazin (A) and pyroxasulfone (B) concentration in the soil ( $\text{ng g}^{-1}$  soil) at 21 d after treatment. Root-mean-square error from each model: flumioxazin, 3.2; and pyroxasulfone, 7.4.



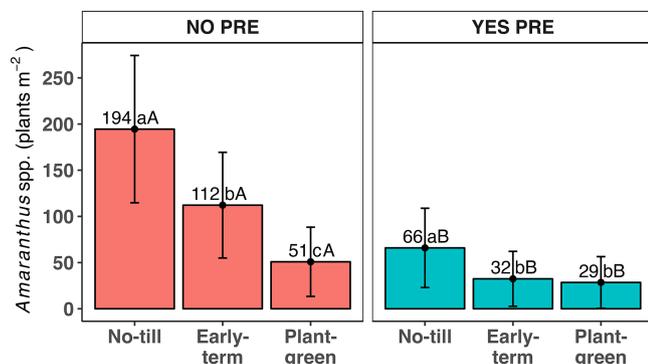
**Figure 6.** Flumioxazin and pyroxasulfone dissipation (%) as a function of cover crop management in 21 d based on the average of each herbicide concentration in the soil ( $\text{ng g}^{-1}$  soil) in the no-till treatment at 0 d after treatment. Data pooled across 2021 and 2022 for each site. Error bars indicate the standard errors of means. Means followed by a common letter are not statistically different by the LSD test ( $\alpha = 0.05$ ). Illinois P-values: flumioxazin, 0.373; and pyroxasulfone, 0.250; Kansas P-values: flumioxazin, <0.001; and pyroxasulfone, <0.001; Pennsylvania P-values: flumioxazin, 0.025; and pyroxasulfone, <0.001; and Wisconsin P-values: flumioxazin, 0.032; and pyroxasulfone, <0.001.

during preemergence application at this site. As for the higher herbicide dissipation compared with other sites, soil microbes are the main source of herbicide degradation in the environment for most herbicides (Ferrell and Vencill 2003). No information about the soil microbial community of each site is available to confirm whether microbial degradation was the cause of such high herbicide dissipation in Illinois. Another hypothesis is that the herbicides might have leached below the sampled soil depth (Mueller et al. 2014). However, precipitation, clay content, soil pH, and soil organic matter, the main factors influencing herbicide adsorption and leaching (Weber et al. 2004; Westra et al. 2014), were similar to those for the other site-years (Figure 2; Table 1). The only observed difference was the higher silt content in Illinois (Table 1). Yet it is unlikely that the high silt content favored herbicide leaching below the sampled soil depth, especially for pyroxasulfone, which has been noted for low leaching potential in the soil (Nakatani et al. 2016; Westra et al. 2014; Yamaji et al. 2016). Thus, it is unclear why Illinois presented the highest flumioxazin and pyroxasulfone degradation compared with other sites.

Flumioxazin and pyroxasulfone dissipation were impacted by cover crop management at the three other sites (Kansas, Pennsylvania, and Wisconsin; Figure 6). Overall, no-till resulted in the least herbicide dissipation over time across herbicides and sites, whereas cereal rye plant-green was the treatment with the most dissipation, and no differences in cereal rye early-term were detected in most instances (Figure 6). Herbicide fate in the environment is a complex combination of processes (Alletto et al. 2010; Locke and Bryson 1997). With the cereal rye system, we hypothesize that the difference in dissipation and final herbicide concentration or dissipation compared with no-till is due to the portion of the applied herbicides that remained adsorbed to the cereal rye biomass and was not washed off to the soil within 21 d. Moreover, we also believe that once intercepted by the cereal rye, the herbicides were more susceptible to degradation from environmental processes, such as photodegradation and volatilization, which may have contributed to herbicide dissipation. Several studies support that once intercepted by crop residue, a portion of the total applied herbicide is not released from the



**Figure 7.** *Amaranthus* spp. density (plants m<sup>-2</sup>) as a function of cover crop management and preemergence herbicide (PRE) at the time of postemergence herbicide application in the low *Amaranthus* spp. pressure group. Data pooled from Illinois 2022, Kansas 2021 and 2022, and Pennsylvania 2021 and 2022. Cover crop management and preemergence interaction P-value = 0.043. Error bars indicate the standard errors of means. Equal lowercase letters indicate no statistical difference between treatments (no-till, cereal rye early-term, and cereal rye plant-green) within the use of preemergence herbicides (YES PRE and NO PRE), and equal uppercase letters indicate no statistical difference between the use of preemergence herbicides within a treatment by the LSD test ( $\alpha = 0.05$ ).



**Figure 8.** *Amaranthus* spp. density (plants m<sup>-2</sup>) as a function of cover crop management and preemergence herbicide (PRE) at the time of postemergence herbicide application in the high *Amaranthus* spp. pressure group. Data pooled from Illinois 2021 and Wisconsin 2021 and 2022. Cover crop management and preemergence interaction P-value = 0.003. Error bars indicate the standard errors of means. Equal lowercase letters indicate no statistical difference between treatments (no-till, cereal rye early-term, and cereal rye plant-green) within the use of preemergence herbicides (YES PRE and NO PRE), and equal uppercase letters indicate no statistical difference between the use of preemergence herbicides within a treatment by the LSD test ( $\alpha = 0.05$ ).

plant material, regardless of rainfall accumulation (Banks and Robinson 1982, 1986; Carbonari et al. 2016). One limitation of this study is that herbicide concentrations were measured only in the soil and not in the cereal rye biomass. Although the overall degree of herbicide dissipation based on the soil concentration in the no-till treatment at 0 DAT was estimated, it is not possible to determine how much of the herbicides was still adsorbed to the cereal rye residue. Future studies quantifying both sources (soil and residue) for herbicide concentration can bring great value to further understanding the impacts of cereal rye cover crop on herbicide fate in the environment.

#### *Amaranthus* spp. Density at Postemergence Application

Splitting the 8 site-years of *Amaranthus* spp. density data into two pressure groups (low:  $\leq 10$  plants m<sup>-2</sup>; high:  $\geq 56$  plants m<sup>-2</sup>)

highlighted the benefits of *Amaranthus* spp. suppression by cereal rye and the importance of adjusting cereal rye termination to increase biomass accumulation in scenarios with and without the use of a preemergence herbicide (Figures 7 and 8). An interaction between cover crop management and preemergence in *Amaranthus* spp. density was detected for the low- (P-value = 0.043) and the high-pressure groups (P-value = 0.003). For both groups, *Amaranthus* spp. density was 55% (low *Amaranthus* spp. pressure) and 42% (high *Amaranthus* spp. pressure) lower in the cereal rye treatments compared with no-till when the preemergence herbicides were not sprayed (Figures 7 and 8), corroborating previous reports that cereal rye can effectively suppress *Amaranthus* spp. emergence (Bish et al. 2021; Cornelius and Bradley 2017). The effective *Amaranthus* spp. suppression observed in this study is likely due to adequate levels of cereal rye biomass accumulation across site-years. Nichols et al. (2020) indicated 5 Mg ha<sup>-1</sup> as the cover crop biomass threshold for effective weed suppression (75% weed biomass reduction). In our study, 6 site-years had cereal rye biomass levels above this threshold in both termination times (Figure 1).

Differences between cereal rye termination times (early termination versus planting green) were only observed under high *Amaranthus* spp. pressure (Figure 8). In this group, cereal rye plant-green without preemergence provided an additional 54% reduction in *Amaranthus* spp. density compared with cereal rye early-term without preemergence and had a lower mean density (15 plants m<sup>-2</sup>) than the no-till with preemergence treatment. Hence, besides improving *Amaranthus* spp. suppression compared with the early termination, delaying cereal rye termination until soybean planting was as effective as the preemergence herbicides adopted in this study for *Amaranthus* spp. control (Figure 8). Cornelius and Bradley (2017) also reported that cereal rye provided early-season *A. tuberculatus* emergence suppression at a comparable level to a preemergence residual herbicide program. Thus, our findings corroborate those of other studies on the benefits of cereal rye for *Amaranthus* spp. management. Nevertheless, it is important to point out that replacing a preemergence herbicide program with the adoption of cereal rye as the sole approach for early-season *Amaranthus* spp. control should be considered with care. Weed suppression by cover crops in general is directly correlated with the increase in cover crop biomass and ground coverage (Nichols et al. 2020). Thus, if not properly established and managed, cereal rye might not uniformly accumulate adequate levels of biomass for effective *Amaranthus* spp. suppression. Moreover, weed species respond differently to cover crop suppression, and some species might not be effectively suppressed as *Amaranthus* spp. is by cereal rye (Cornelius and Bradley 2017). Therefore, as an approach to diversify and adopt as many tools as possible, both cereal rye and preemergence herbicides should be considered as part of integrated weed management programs by farmers.

The fact that cereal rye plant-green only showed a benefit under high *Amaranthus* spp. pressure (Figure 8) raises the question of whether *Amaranthus* spp. suppression is correlated not only with cereal rye biomass accumulation but also with its pressure in the field. In the low- pressure group (Figure 7), there was no benefit in delaying cereal rye termination to increase biomass production for *Amaranthus* spp. suppression. Bish et al. (2021) also hypothesized that *A. tuberculatus* suppression might be linked to its soil seedbank infestation level and that there may be a threshold of *A. tuberculatus* seed density for which cereal rye biomass cannot compensate. The authors observed that even a

high level of cereal rye biomass (14 Mg ha<sup>-1</sup>) was not enough to provide effective *A. tuberculatus* suppression when its density was recorded at  $\geq 927$  plants m<sup>-2</sup> in the absence of cereal rye at 28 d after soybean planting. Conversely, a significant reduction in *A. tuberculatus* density was observed in two other years when *A. tuberculatus* density was  $\leq 385$  plants m<sup>-2</sup> without cereal rye at 28 d after planting. In our study, the no-till without preemergence had an average of 194 *Amaranthus* spp. plants m<sup>-2</sup> in the high-pressure group. Considering the findings from Bish et al. (2021), at this level, cereal rye can effectively reduce *Amaranthus* spp. emergence, which was indeed confirmed in both cereal rye treatments. Nevertheless, the hypothesis of a threshold in *Amaranthus* spp. pressure at which cereal rye can no longer effectively suppress it should be investigated under field conditions.

The application of the preemergence herbicides flumioxazin and pyroxasulfone provided effective residual *Amaranthus* spp. control in all treatments and both pressure groups (Figures 7 and 8). The effectiveness of flumioxazin and pyroxasulfone on *Amaranthus* spp. control has already been described in the literature. Perkins et al. (2021) reported that flumioxazin and pyroxasulfone, applied alone or as a premix, were some of the most effective residual herbicides for *A. tuberculatus* and *A. palmeri* control in soybeans. Also, Ferrier et al. (2022) described the combination of flumioxazin and pyroxasulfone as having an additive effect in the control of multiple herbicide-resistant *A. tuberculatus*. Thus, our findings support that flumioxazin and pyroxasulfone are effective preemergence herbicide options for *Amaranthus* spp. control.

Although the application of flumioxazin and pyroxasulfone resulted in lower *Amaranthus* spp. density for all cover crop management practices, their interaction with the cereal rye treatments was different depending on the *Amaranthus* spp. pressure group (Figures 7 and 8). In low *Amaranthus* spp. pressure, there was no difference between cover crop management when the preemergence herbicides were sprayed (Figure 7). However, under the high *Amaranthus* spp. pressure, both cereal rye treatments resulted in an additional 46% (average of cereal rye early-term and cereal rye plant-green) reduction in *Amaranthus* spp. density compared with no-till with preemergence (Figure 8). Therefore, despite reducing flumioxazin and pyroxasulfone concentrations in the soil compared with no-till in most instances, the presence of cereal rye biomass did not impact overall early-season *Amaranthus* spp. control in this study. Instead, the presence of cereal rye biomass improved *Amaranthus* spp. control compared with no-till both in the presence and absence of the preemergence residual treatment. Thus, the combination of preemergence herbicides and cereal rye under the planting green system can be an effective integrated *Amaranthus* spp. management strategy in soybean.

Cereal rye cover crop biomass impacted the fate of the preemergence herbicides flumioxazin and pyroxasulfone by reducing their concentrations in the soil compared with no-till. Delaying cereal rye termination until soybean planting resulted in further reduction in the concentration of both herbicides in some instances. Yet main differences were observed between no-till and cereal rye treatments rather than cereal rye termination times (early vs. planting green). Despite reducing flumioxazin and pyroxasulfone concentrations in the soil, the presence of cereal rye biomass did not affect early-season residual *Amaranthus* spp. control compared with no-till. Therefore, the adoption of effective preemergence herbicides associated with a properly managed cereal rye cover crop is an effective option for integrated *Amaranthus* spp. management in soybean production systems.

Additional research is warranted to further our understanding regarding environmental fate of preemergence herbicides with different physicochemical properties in cropping systems where cover crops are included.

**Supplementary material.** To view supplementary material for this article, please visit <https://doi.org/10.1017/wsc.2023.46>

**Acknowledgments.** We thank the members of the Kansas State University, Pennsylvania State University, Southern Illinois University, University of Tennessee, and University of Wisconsin–Madison Weed Science laboratories for their technical support for this project. This research was funded by the United Soybean Board. No conflicts of interest have been declared.

## References

- Alister C, Rojas S, Gomes P, Kogan M (2008) Dissipation and movement of flumioxazin in soil at four field sites in Chile. *Pest Manag Sci* 64:579–583
- Alletto L, Coquet Y, Benoit P, Hedddadj D, Barriuso E (2010) Tillage management effects on pesticide fate in soils. A review. *Agron Sustain Dev* 30:367–400
- Anonymous (2016) Valor SX® herbicide product label. Form 1501-I. Walnut Creek, CA: Valent. 31 p
- Anonymous (2019) Fierce EZ® herbicide product label. Form 2212-C. Walnut Creek, CA: Valent. 12 p
- Anonymous (2022) Zidua® herbicide product label. NVA 2022-04-0489-0019. Research Triangle Park, NC: BASF. 26 p
- Banks PA, Robinson EL (1982) The influence of straw mulch on the soil reception and persistence of metribuzin. *Weed Sci* 30:164–168
- Banks PA, Robinson EL (1986) Soil reception and activity of acetochlor, alachlor, and metolachlor as affected by wheat (*Triticum aestivum*) straw and irrigation. *Weed Sci* 34:607–611
- Baraibar B, Mortensen DA, Hunter MC, Barbercheck ME, Kaye JP, Finney DM, Curran WS, Bunchek J, White CM (2018) Growing degree days and cover crop type explain weed biomass in winter cover crops. *Agron Sustain Dev* 38:1–9
- Beckie HJ, Ashworth MB, Flower KC (2019) Herbicide resistance management: recent developments and trends. *Plants* 8:161
- Biau G, Scornet E (2016) A random forest guided tour. *TEST* 25:197–227
- Bish M, Dintelmann B, Oseland E, Vaughn J, Bradley K (2021) Effects of cereal rye seeding rate on waterhemp (*Amaranthus tuberculatus*) emergence and soybean growth and yield. *Weed Technol*. 35:838–844
- Bourgoin C, Blanc L, Bailly J-S, Cornu G, Berenguer E, Oszwald J, Tritsch I, Laurent F, Hasan AF, Sist P, Gond V (2018) The potential of multisource remote sensing for mapping the biomass of a degraded Amazonian forest. *Forests* 9:1–21
- Breiman L (2001) Random forests. *Mach Learn* 25:5–32
- Brooks ME, Kristensen K, van Benthem KJ, Magnusson A, Berg CW, Nielsen A, Skaug HJ, Maechler M, Bolker BM (2017) glmmTMB balances speed and flexibility among packages for zero-inflated generalized linear mixed modeling. *R J* 9:378–400
- Bunchek JM, Wallace JM, Curran WS, Mortensen DA, VanGessel MJ, Scott BA (2020) Alternative performance targets for integrating cover crops as a proactive herbicide-resistance management tool. *Weed Sci* 68:534–544
- Carbonari C, Gomes G, Trindade M, Silva J, Velini E (2016) Dynamics of sulfentrazone applied to sugarcane crop residues. *Weed Sci* 64:201–206
- Cornelius CD, Bradley KW (2017) Influence of various cover crop species on winter and summer annual weed emergence in soybean. *Weed Technol* 31:503–513
- Crutchfield DA, Wicks GA, Burnside OC (1985) Effect of winter wheat (*Triticum aestivum*) straw mulch level on weed control. *Weed Sci* 34: 110–114
- Duke SO, Powles SB (2008) Glyphosate: a once-in-a-century herbicide. *Pest Manag Sci* 64:319–325
- Eason K, Grey T, Cabrera M, Basinger N, Hurdle N (2022) Assessment of flumioxazin soil behavior and thermal stability in aqueous solutions. *Chemosphere* 288(Pt 2):132477

- Erbach DC, Lovely WG (1975) Effect of plant residue on herbicide performance in no-tillage corn. *Weed Sci* 23:512–515
- Faleco F, Oliveira M, Arneson N, Renz M, Stoltenberg D, Werle R (2022) Multiple herbicide resistance in waterhemp (*Amaranthus tuberculatus*) accessions from Wisconsin. *Weed Technol* 36:597–608
- Ferrell JA, Vencill WK (2003) Flumioxazin soil persistence and mineralization in laboratory experiments. *J Agric Food Chem* 51:4719–4721
- Ferrier J, Soltani N, Hooker DC, Robinson DE, Sikkema PH (2022) The interaction of pyroxasulfone and flumioxazin applied preemergence for the control of multiple-herbicide-resistant waterhemp (*Amaranthus tuberculatus*) in soybean. *Weed Technol* 36:318–323
- Fisher JL, Sprague CL (2022) Narrow-row soybean and a cereal rye cover crop suppress glyphosate-resistant horseweed (*Conyza canadensis*). *Weed Technol* 36:781–788
- Green JM (2014) Current state of herbicides in herbicide-resistant crops. *Pest Manag Sci* 70:1351–1357
- Hartway CR, Ott JP, Grulke NE (2022) Plant life span and persistence of soil seedbanks predict the emergence of herbicide resistance in noxious weeds. *Weed Sci* 70:448–454
- Heap I (2023) The International Herbicide-Resistant Weed Database. [www.weedscience.org](http://www.weedscience.org) Accessed: March 16, 2023
- Johnson G, Breitenbach F, Behnken L, Miller R, Hoverstad T, Gunsolus J (2012) Comparison of herbicide tactics to minimize species shifts and selection pressure in glyphosate-resistant soybean. *Weed Technol* 26:189–194
- Kuhn M, Silge J (2022) Tidy Modeling with R: A Framework for Modeling in the Tidyverse. 1st ed. Sebastopol, CA: O'Reilly Media. 381 p
- Lenth R (2022) emmeans: Estimated Marginal Means, aka Least-Squares Means. R package v. 1.8.2. <https://CRAN.R-project.org/package=emmeans> Accessed: March 16, 2023
- Locke MA, Bryson CT (1997) Herbicide-soil interactions in reduced tillage and plant residue management systems. *Weed Sci* 45:307–320
- Loupe G, Wehenkel L, Sutura A, Geurts P (2013) Understanding variable importances in forests of randomized trees. Pages 431–439 in 2013 Proceedings of Neural Information Processing Systems 26
- Mahoney K, Shropshire C, Sikkema P (2014) Weed management in conventional- and no-till soybean using flumioxazin/pyroxasulfone. *Weed Technol* 28:298–306
- Mirsky S, Curran W, Mortensen D, Ryany M, Shumway D (2011) Timing of cover-crop management effects on weed suppression in no-till planted soybean using a roller-crimper. *Weed Sci* 59:380–389
- Mueller TC, Boswell BW, Mueller SS, Steckel LE (2014) Dissipation of fomesafen, saflufenacil, sulfentrazone, and flumioxazin from a Tennessee soil under field conditions. *Weed Sci* 62:664–671
- Mueller TC, Senseman SA (2015) Methods related to herbicide dissipation or degradation under field or laboratory conditions. *Weed Sci* 63:133–139
- Nakatani M, Yamaji Y, Honda H, Uchida Y (2016) Development of the novel pre-emergence herbicide pyroxasulfone. *J Pestic Sci* 41:107–112
- Nichols V, Martinez-Feria R, Weisberger D, Carlson S, Basso B, Basche A (2020) Cover crops and weed suppression in the U.S. Midwest: a meta-analysis and modeling study. *Agric Environ Lett* 5:e20022
- Nunes J, Arneson N, DeWerff R, Ruark M, Conley S, Smith D, Werle R (2023) Planting into a living cover crop alters preemergence herbicide dynamics and can reduce soybean yield. *Weed Technol* 37:226–235
- Palhano MG, Norsworthy JK, Barber T (2018) Cover crops suppression of Palmer amaranth (*Amaranthus palmeri*) in cotton. *Weed Technol* 32:60–65
- Perkins C, Gage K, Norsworthy J, Young B, Bradley K, Bish M, Hager A, Steckel L (2021) Efficacy of residual herbicides influenced by cover-crop residue for control of *Amaranthus palmeri* and *A. tuberculatus* in soybean. *Weed Technol* 35:77–81
- Peterson MA, Collavo A, Ovejero R, Shivrain V, Walsh MJ (2018) The challenge of herbicide resistance around the world: a current summary. *Pest Manag Sci* 74:2246–2259
- Piepho HP, Edmondson RN (2018) A tutorial on the statistical analysis of factorial experiments with qualitative and quantitative treatment factor levels. *J Agron Crop Sci* 204:429–455
- Pinheiro J, Bates D (2000) Mixed-effects Models in S and S-PLUS. New York: Springer. 528 p
- Pinheiro J, Bates D, R Core Team (2022) nlme: Linear and Nonlinear Mixed Effects Models. R package v. 3.1-157. <https://CRAN.R-project.org/package=nlme>. Accessed: March 16, 2023
- R Core Team (2022) R: A Language and Environment for Statistical Computing. Vienna, Austria: R Foundation for Statistical Computing
- Reed HK, Karsten HD, Curran WS, Tooker JF, Duiker SW (2019) Planting green effects on corn and soybean production. *Agron J* 111:2314–2325
- Schmitz GL, Witt WW, Mueller TC (2001) The effect of wheat (*Triticum aestivum*) straw levels on chlorimuron, imazaquin, and imazethapyr dissipation and interception. *Weed Technol* 15:129–136
- Schramski JA, Sprague CL, Renner KA (2021) Effects of fall-planted cereal cover-crop termination time on glyphosate-resistant horseweed (*Conyza canadensis*) suppression. *Weed Technol* 35:223–233
- Schwartz L, Norsworthy JK, Young BG, Bradley KW, Kruger GR, Davis VM, Steckel LE, Walsh MJ (2016) Tall waterhemp (*Amaranthus tuberculatus*) and Palmer amaranth (*Amaranthus palmeri*) seed production and retention at soybean maturity. *Weed Technol* 30:284–290
- Selim HM, Zhou L, Zhu H (2003) Herbicide retention in soil as affected by sugarcane mulch residue. *J Environ Qual* 32:1445–54
- Shaner DL, ed (2014) Herbicide Handbook. 10th ed. Lawrence, KS: Weed Science Society of America. 513 p
- Sperry BP, Ferguson JC, Bond JA, Kruger GR, Johnson AB, Reynolds DB (2022) Effect of differential levels of simulated overhead irrigation on residual herbicides applied to wheat straw-covered soil for barnyardgrass control. *Weed Technol* 36:648–654
- Steckel LE (2007) The dioecious *Amaranthus* spp.: here to stay. *Weed Technol* 21:567–570
- Striegel S, Oliveira MC, Arneson N, Conley SP, Stoltenberg DE, Werle R (2021) Spray solution pH and soybean injury as influenced by synthetic auxin formulation and spray additives. *Weed Technol* 35:113–127
- Stroup WW (2015) Rethinking the analysis of non-normal data in plant and soil science. *Agron J* 107:811–827
- [USEPA] U.S. Environmental Protection Agency (2023) Pesticide Registration. <https://www.epa.gov/pesticide-registration>. Accessed: March 10, 2023
- Van Wychen L (2022) 2022 Survey of the Most Common and Troublesome Weeds in Broadleaf Crops, Fruits & Vegetables in the United States and Canada. Weed Science Society of America National Weed Survey Dataset. [http://wssa.net/wp-content/uploads/2022\\_weed\\_survey\\_broadleaf\\_crops.xlsx](http://wssa.net/wp-content/uploads/2022_weed_survey_broadleaf_crops.xlsx). Accessed: July 31, 2023
- Weber JB, Wilkerson GG, Reinhardt CF (2004) Calculating pesticide sorption coefficients (*K<sub>d</sub>*) using selected soil properties. *Chemosphere* 55:157–166
- Webster TM, Simmons DB, Culpepper AS, Grey TL, Bridges DC, Scully BT (2016) Factors affecting potential for Palmer amaranth (*Amaranthus palmeri*) suppression by winter rye in Georgia, USA. *Field Crops Res* 192:103–109
- Westra E, Shaner D, Westra P, Chapman P (2014) Dissipation and leaching of pyroxasulfone and S-metolachlor. *Weed Technol* 28:72–81
- Whalen DM, Shergil LS, Kinne LP, Bish MD, Bradley KW (2020) Integration of residual herbicides with cover crop termination in soybean. *Weed Technol* 34:11–18
- Wickham H (2016) ggplot2: Elegant Graphics for Data Analysis. New York: Springer-Verlag. 213 p
- Wickham H, Averick M, Bryan J, Chang W, McGowan LD, François R, Grolemund G, Hayes A, Henry L, Hester J, Kuhn M, Pedersen TL, Miller E, Bache SM, Müller K, et al. (2019) Welcome to the tidyverse. *J Open Source Softw* 4(43):1686
- Wright M (2023) Ranger: A Fast Implementation of Random Forests. <https://cran.r-project.org/web/packages/ranger/ranger.pdf>. Accessed: March 17, 2023
- Yamaji Y, Honda H, Hanai R, Inoue J (2016) Soil and environmental factors affecting the efficacy of pyroxasulfone for weed control. *J Pestic Sci* 41: 1–5