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Crop response to simulated diflufenican carryover and sprayer contamination with a diflufenican premixture

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Palmer amaranth [Amaranthus palmeri (S.) Wats] has evolved resistance to nine sites of action (SOAs) globally, leaving producers searching for effective herbicide options. Bayer CropScience has announced intentions to launch a series of Convintro[™] brand herbicides, one of which will be a premixture including diflufenican, metribuzin, and flufenacet for preemergence use in soybean [Glycine max (L.) Merr]. Diflufenican is a WSSA group 12 herbicide that would add a new SOA for soybean producers. With the anticipated launch of the premixture, research is needed to evaluate the potential for diflufenican carryover or tank contamination from a diflufenican premixture to injure corn, cotton, grain sorghum, rice, and soybean. Carryover experiments were conducted in 2022 and 2023 in Fayetteville, AR, and near Stuttgart, AR. Diflufenican was applied preplant at 0, 7.5, 15, 30, 60, and 120 g ai ha⁻¹, with crops being planted after a 1.3 cm irrigation or rainfall event occurred. Injury was <10% for rice and corn, <5% for grain sorghum, and no crop response from cotton at 14 days after emergence (DAE). No detrimental effect was observed on various crops by 28 DAE. Additionally, tank contamination experiments were conducted in Arkansas in Fayetteville, near Stuttgart, near Colt, and in Keiser. A 0.17:0.35:0.48 ratio of a diflufenican:metribuzin:flufenacet premixture, at rates up to 103 g ai ha⁻¹, was applied at the 3-leaf growth stage for each crop. Injury was <30% for cotton (Gossypium hirsutum L.), <20% for corn (Zea mays L.) and soybean, and <5% for grain sorghum (Sorghum bicolor L.) and rice (Oryza sativa L.) at 7 days after treatment (DAT). By 28 DAT, injury was <15% for cotton, <10% injury for soybean and rice, and no crop response for corn or grain sorghum. Overall, the potential for diflufenican carryover or risk of injury from sprayer contamination appears low, with no yield reductions occurring in any of the evaluated crops in either set of experiments.

KEYWORDS

soybean, crop injury, Convintro, group 12 herbicide, diflufenican

1 Introduction

Palmer amaranth is currently ranked the most problematic weed faced by soybean producers across the United States (Van Wychen, 2022). Research has shown that when competing with soybean, a density of 10 Palmer amaranth plants m^{-1} of row can reduce yields by 68% (Klingaman and Oliver, 1994). Herbicides remain the best control agent for weed species within a cropping system (Booth and Swanton, 2002); however, the weed has evolved resistance to 9 sites of action (SOA) globally (Heap, 2024). The need for new SOA remains high for producers to integrate into herbicide programs aimed at controlling Palmer amaranth.

In 2021, Bayer CropScience announced the launch of ConvintroTM brand herbicides, one of which will be a premixture for use in soybean (Anonymous, 2021a). The premixture will include diflufenican (WSSA Group 12), metribuzin (WSSA Group 5), and flufenacet (WSSA Group 15) for preemergence (PRE) use in soybean. Currently, WSSA groups 2, 3, 4, 5, 14, and 15 herbicides are recommended for use PRE in soybean (Barber et al., 2024). In addition, norflurazon, another Group 12 herbicide, is labeled for use in soybean (Anonymous, 2015). However, the herbicide is not used due to price, and the label restricts its use to the mid-southern United States. Therefore, if labeled, diflufenican would add a new SOA for soybean producers across the United States.

Diflufenican is a phytoene desaturase inhibitor discovered in 1979 as a potential herbicide for PRE and early postemergence (POST) use in winter wheat (*Triticum aestivum* L.) and barley (*Hordeum vulgare* L.) (Cramp et al., 1987). The typical symptomology is bleaching due to inhibiting the desaturation reaction of carotenoid biosynthesis, causing the accumulation of phytoene in place of the normal-colored carotenoids (Bartel and Watson, 1978). Research has shown that diflufenican is effective against several weed species, including catchweed bedstraw (*Galium aparine* L.), common chickweed (*Stellaria media* L. Vill.), and field pansy (*Viola arvensis* L.) (Haynes and Kirkwood, 1992). Additionally, the primary target weed of diflufenican in the United States is Amaranthus ssp (Anonymous, 2021a). The spectrum of diflufenican appears to be limited to broadleaf weed species and will need to be paired with other herbicides to achieve broad-spectrum weed control.

With the anticipation of the premixture being labeled in the coming years, research is needed to evaluate the potential for diflufenican to carryover and injure crops typically rotated with soybean. Herbicide persistence in the soil can be attributed to soil pH, soil composition, soil texture, soil microorganisms, water solubility, herbicide degradation, and application timing and methods (Curran, 2016). Acetolactate synthase inhibitors (WSSA Group 2) have been documented to have the ability to persist and carryover, causing injury to crops grown in the subsequent season. Up to 70% injury can occur in cotton during the subsequent growing season from an application of imazaquin during the previous growing season (Johnson et al., 1995).

Research in Europe has evaluated the metabolism of diflufenican in the soil of wheat fields and the potential to injure rotational crops. Following a 5- or 9-month soil treatment with diflufenican, the residues of the herbicide in the soil were so low no hindrance could be observed for a sensitive sugar beet cultivar (*Beta vulgaris* subsp. Altissima group) (Rouchaud et al., 1991). The half-life of diflufenican is estimated to be 2 to 6 months when used in wheat (Cramp et al., 1987), with the rate of diflufenican soil biodegradation being greater during the warmer period correlating with increased microbial and enzymatic activity in the soil (Rouchaud et al., 1991). However, further research is needed to assess the use of diflufenican in climatic conditions conducive to soybean growth and the potential to carryover to subsequent rotational crops.

Another concern for producers is the contamination of sprayers from improper clean out and the potential for the diflufenican: metribuzin:flufenacet to injure other crops. Sprayer contamination with auxin herbicides (WSSA Group 4) such as florpyrauxifenbenzyl, dicamba, and 2,4-D have injured sensitive vegetation at low concentrations. Yield reductions of up to 52% have occurred from dicamba at a rate of 70 g ae ha⁻¹ (1/8X) when occurring at the V3/ V4 growth stage for non-dicamba tolerant soybean (Griffin et al., 2013). If labeled, Convintro, a premixture herbicide, carries three chemicals that can potentially injure crops from postemergence (POST) exposure from contaminated sprayers.

Currently, there is no published research on the sensitivity of different crops for POST exposure to diflufenican. Metribuzin has also been evaluated as a potential atrazine replacement in corn and grain sorghum with <10% injury for both crops (Richburg, 2019; Richburg et al., 2019). Additionally, research has evaluated low concentrations of metribuzin POST in cotton, soybean, and rice. Overall, no yield reductions occurred in cotton when applied at V5-8 (Hurst, 1982) and ≤15% injury occurred in rice from 2-3-leaf applications (Lawrence et al., 2021). Subsequently, up to 30% soybean injury can occur when exposed at the V4 growth stage (Stephenson et al., 2019). Finally, flufenacet is labeled for preplant to early POST use in corn and soybean (Anonymous, 2021b). Research has also been conducted to evaluate flufenacet for use in grain sorghum (Geier et al., 2009) with minimal concerns for injury early-postemergence. In addition, there is no published data on the sensitivity of POST exposure for cotton and rice to flufenacet.

Overall, this research aimed to understand the risk of soil-applied diflufenican rates to injure common rotational crops of soybean grown in the mid-southern United States. Additionally, research was conducted to evaluate the sensitivity of various crops to postemergence exposure to low rates of the diflufenican:metribuzin:flufenacet premixture.

2 Materials and methods

2.1 Crop response to simulated diflufenican carryover in the field

Field experiments evaluated the sensitivity to diflufenican carryover of crops grown in rotation with soybean in the Midsouth. In 2022 and 2023, experiments were conducted at two locations across Arkansas: the Milo J. Shult Agriculture Research and Extension Center in Fayetteville, AR, on a Leaf silt loam (fine, mixed, active, thermic Typic Albaquults) (USDA, 2024) with 18% sand, 69% silt, 13% clay, and 1.6% organic matter (Arkansas

Agricultural Diagnostic Laboratory, Fayetteville, AR), and the Rice Research and Extension Center near Stuttgart, AR, on a Dewitt silt loam (Fine, smectitic, thermic Typic Albaqualfs) (USDA, 2024) with 27% sand, 54% silt, 19% clay, 1.75% organic matter, and a pH of 6.2 (Arkansas Agricultural Diagnostic Laboratory, Fayetteville, AR). The seedbed was prepared using conventional tillage, including disking, field cultivation, and bedding of rows, except rice, before planting at both locations. The trials at Fayetteville were planted on raised beds using a four-row vacuum planter into plots measuring 6.1 m in length and 3.7 m in width. Additionally, experiments for rice were flat planted using a nine-row, small plot drill and direct seeded into plots measuring 5.2 m in length and 1.8 m in width at Stuttgart, AR.

The trials were designed as a randomized complete block with four replications and one factor (herbicide rate). Diflufenican was applied at 0, 7.5, 15, 30, 60, and 120 (1X) g ai ha⁻¹ preplant. Herbicide applications were made with a CO_2 -pressurized backpack sprayer and a 4-nozzle boom, using AIXR 110015 nozzles (TeeJet Technologies, Springfield, IL) calibrated to deliver 140 L ha⁻¹ at 4.8 km hr⁻¹. Following herbicide applications, four crops, rice (PVL03 Horizon Ag, LLC, Memphis, TN), corn (DKC 62-69, Bayer CropScience, St. Louis, MO), cotton (DP2020B3XF, Bayer CropScience, St. Louis, MO), and grain sorghum (DKS 62-69, Bayer CropScience, St. Louis, MO) (Table 1), were planted following a 1.3 cm rainfall or irrigation event. Plots were kept weed-free throughout the growing season using standard herbicide programs for each crop.

2.2 Crop response to simulated diflufenican carryover in the greenhouse

Greenhouse studies were conducted in 2023 and 2024 at the Milo J. Shult Agriculture Research and Extension Center in

Fayetteville, Arkansas. A Captina silt loam soil (Fine-silty, siliceous, active, mesic Typic Fragiudults) (USDA, 2024) with 14% clay, 20% sand, 66% silt, and 2.3% organic matter (Agricultural Diagnostic Laboratory, Fayetteville, AR) was collected and sieved to remove large pieces of residue and reduce the size of large soil aggregates. The soil was then dried at 33 C for 14 days before the initiation of the experiment. Plastic tubs measuring 36 cm in length, 18 cm in width, and 15 cm in depth were filled with 5.5 kg of soil. The trials were initiated on November 13, 2023, and January 30, 2024.

The experiment was a complete randomized design with one factor (herbicide rate) and four replications. Diflufenican was applied at 0, 7.5, 15, 30, 60, and 120 (1X) g ai ha⁻¹ preplant. Herbicide applications were made in a spray chamber with two flatfan 1100067 nozzles (Teejet Technologies, Springfield, IL) calibrated to deliver 187 L ha⁻¹ at 1.61 km hr⁻¹. Following the herbicide application, the soil was mixed thoroughly in 19 L buckets and returned to its respective tub. Four different crops, including rice, grain sorghum, corn, and cotton, were hand-planted in two rows (18 cm) in each tub, with two crops per tub. Rice (PVL03) was planted at eleven seeds per row, grain sorghum (DKS 54-07) at seven seeds per row, corn (DKC 62-69) at five seeds per row, and cotton (DP2020B3XF) at five seeds per row.

2.3 Crop response to sprayer contamination of a diflufenican : metribuzin:flufenacet premixture

Field trials were conducted to evaluate the potential for crop injury caused by sprayer contamination from a diflufenican: metribuzin:flufenacet premixture (0.17:0.35:0.48 ratio). Two trials were conducted for each crop at various locations across Arkansas. Cotton (DP2127B3XF, Bayer CropScience, Saint Louis, MO,

TABLE 1 Application date, activating rainfall date, planting date, row spacing, and seeding rate for the simulated carryover experiments planted in Fayetteville and near Stuttgart, AR.

Location	Crop ^a	Application date	Activating rainfall a date	Rainfall amount ^b	Planting date	Row width	Seeding rate
				cm		cm	1,000 seed ha ⁻¹
Fayetteville	Corn	April 22, 2022	April 25, 2022	8.9	April 28, 2022	91	84
		April 18, 2023	April 22, 2023	1.3 ^c	April 26, 2023	91	84
	Cotton	May 13, 2022	May 16, 2022	1.3	May 17, 2022	91	99
		May 8, 2023	May 19, 2023	1.3	May 22, 2023	91	99
	Grain sorghum	May 13, 2022	May 16, 2022	1.3	May 17, 2022	91	210
		May 8, 2023	May 19, 2023	1.3	May 22, 2023	91	247
Stuttgart	Rice	April 28, 2022	May 6, 2022	5.2	April 29, 2022	19	4,200
		April 13, 2023	April 21, 2023	10.2	May 02, 2023	19	4,200

^aThe same crop varieties were planted at all locations.

^bRainfall amounts that occurred between herbicide application and planting.

^cActivated with overhead irrigation.

63167), grain sorghum (DKS 54-07), soybean (AG45XFO, Bayer CropScience, Saint Louis, MO, 63167), and corn (DKC 62-69) (Table 2) were planted at the Milo J. Shult Agriculture Research and Extension Center in Fayetteville, AR, on a Leaf silt loam (Fine, mixed, active, thermic Typic Albaquults) (USDA, 2024) with 18% sand, 69% silt, and 13% clay, with a pH of 6.6 and 1.6% organic matter (Arkansas Agricultural Diagnostic Laboratory, Fayetteville, AR). The same crops and cultivars were also grown at the Northeast Arkansas Research and Extension Center in Keiser, AR, on a Sharkey silty clay (Very-fine, smectitic, thermic Chromic Epiaquerts) (USDA, 2024) with 17% sand, 34% silt, and 49% clay, with a pH of 6.9 and 2.3% organic matter (Arkansas Agricultural Diagnostic Laboratory, Fayetteville, AR). Rice (PVL03) was planted at the Rice Research and Extension Center near Stuttgart, AR, on a Dewitt silt loam (Fine, smectic, thermic Typic Albaqualfs) (USDA, 2024) with 27% sand, 54% silt, and 19% clay, with a pH of 6.2 and 1.7% organic matter (Arkansas Agricultural Diagnostic Laboratory, Fayetteville, AR), and at the Pine Tree Research Station near Colt, AR, on a Calhoun silt loam (Fine-silty, mixed, active, thermic Typic Glossaqualfs) (USDA, 2024) with 17% sand, 68% silt, and 15% clay, with a pH of 6.7 and 1.4% organic matter (Arkansas Agricultural Diagnostic Laboratory, Fayetteville, AR). The seedbed was prepared using conventional tillage involving disking, field cultivation, and bedding of rows, except rice, at all locations. The trials were planted on raised beds using a four-row vacuum planter into plots measuring 7.6 m in length and 3.6 m in width at Fayetteville, 3.8 m in width, and 7.6 m in length at Keiser. Additionally, trials were flat planted using a nine-row, small-plot drill into plots measuring 5.2 m in length near Pine Tree and Stuttgart.

The experiments were designed as a randomized complete block with four replications and one factor. The diflufenican: metribuzin:flufenacet premixture was applied at 0, 1.6 (1/640X), 6.4 (1/160X), 25.8 (1/40X), and 103.1 (1/10X) g ai ha⁻¹, with 1,031 g ai ha⁻¹ representing the max use rate. Herbicide applications were made at the V3 growth stage using a CO_2 -pressurized backpack sprayer and a 4-nozzle boom, calibrated to deliver 140 L ha⁻¹ at 4.8 km hr⁻¹. Plots were weed-free throughout the growing season using standard herbicides for each crop.

2.4 Fertility and irrigation

Preplant fertilizer was applied when needed based on soil test results and fertilizer recommendations from the University of Arkansas for soybean, corn, cotton, and grain sorghum (Ross et al., 2022; Kelley and Capps, 2022a; Robertson et al., 2022; Kelley and Capps, 2022b). Additionally, nitrogen (N) in the form of urea (46-0-0) was applied at a rate of 200 kg N ha⁻¹ for corn, 90 kg N ha⁻¹ for cotton, 112 kg N ha⁻¹ for grain sorghum, and 130 kg N ha⁻¹ for rice during the growing season (Hardke et al., 2022). Once rice reached the 5-leaf growth stage, a permanent flood was established near Stuttgart and Colt, AR. Finally, row crops were irrigated via furrow or overhead irrigation if 2.5 cm of rainfall did not occur within a seven-day period.

2.5 Field/greenhouse assessments

Visible injury ratings and SPAD (SPAD 502 Plus Chlorophyll Meter, Spectrum Technologies, Inc., Oak Ridge, TN) meter readings (excluding rice) of the uppermost fully expanded leaf of five plants were collected 14 and 28 days after emergence (DAE) for simulated diflufenican carryover in the field. Additionally, crop density in two 1-m sections of the row was collected 14 DAE, and groundcover

TABLE 2 Planting date, spray date, and row width of the five crops for the sprayer contamination experiments planted in Fayetteville, Keiser, near Stuttgart, and near Colt, AR.

Location	Crop ^a	Planting date	Application	Row width	Seeding rate	
			date	cm	1,000 seed ha ⁻¹	
Fayetteville	Cotton	May 8, 2023	June 12, 2023	91	112	
	Corn	April 12, 2023	May 16, 2023	91	84	
	Grain sorghum	May 3, 2023	May 23, 2023	91	247	
	Soybean	May 3, 2023	May 31, 2023	91	346	
Keiser	Cotton	May 17, 2023	June 7, 2023	97	99	
	Corn	April 13, 2023	May 11, 2023	97	84	
	Grain sorghum	May 17, 2023	May 30, 2023	97	222	
	Soybean	May 17, 2023	June 7, 2023	97	346	
Pine Tree	Rice	April 11, 2023	May 17, 2023	19	4,200	
Stuttgart ^b	Rice	May 8, 2023	June 5, 2023	19	4,200	

^aThe same crop varieties/cultivars were planted at all locations.

^bUnable to collect data due to herbicide carryover from the previous growing season killing the rice.

TABLE 3 Influence of simulated carryover from soil-applied diflufenican on crop injury, SPAD meter readings (greenness), crop density, crop groundcover, and relative yield.

		Injury	SP	AD	Density	Groundcover	
Crop	Rate	14 DAE ^a	14 DAE	28 DAE	14 DAE	28 DAE	Relative yield ^b
	g ai ha⁻¹	%			plants m ⁻²	%	<u> </u>
Cotton	0 ^c	-	39.3	40.9	13.0	8b	_d
	7.5	0	40.5	41.7	11.8	11a	118
	15	0	39.4	41.5	11.5	9ab	110
	30	0	40.3	42.3	11.9	8b	107
	60	0	40.0	41.0	12.1	9ab	105
	120	0	39.4	41.8	12.2	8b	114
	P-value	1.000	0.6182	0.6517	0.5942	0.0005	0.1847
Corn	0	_	33.2 ^f	40.6	9.2	45	-
	7.5	1b ^e	32.9	41.8	9.5	47	115
	15	2b	33.2	42.2	9.3	46	114
	30	2b	33.3	41.8	9.3	50	116
	60	3ab	34.1	41.6	9.6	50	107
	120	6a	33.7	42.6	9.7	50	99
	P-value	0.0005	0.5825	0.2832	0.7617	0.2460	0.2395
Grain sorghum	0	_	34.3	38.6	28.5	25	_g
	7.5	0c	33.8	38.2	29.2	25	104
	15	0c	35.7	39.4	28.8	22	103
	30	0c	34.8	39.0	30.5	22	97
	60	1b	34.7	38.1	28.6	24	103
	120	2a	34.5	38.6	27.8	22	99
	P-value	<0.0001	0.2565	0.2709	0.6977	0.2556	0.1983
Rice	0	-	NA ^h	NA	200a	62	-
	7.5	6	NA	NA	194ab	62	102
	15	6	NA	NA	202a	60	102
	30	9	NA	NA	185ab	58	105

(Continued)

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	Relative yield ^b	?	100	106	0.2783	
Groundcover	28 DAE	۵	57	61	0.6034	
Density	14 DAE	plants m ⁻²	190ab	174b	0.0030	
AD	28 DAE		NA	NA	NA	
SP	14 DAE		NA	NA	NA	
Injury	14 DAE ^a	%	8	6	0.3217	
	Rate	g ai ha ⁻¹	60	120	P-value	
	Crop					DAE, days after emergence.

Nontreated yields for seed cotton were 2,200 kg ha $^{-1}$, 8,550 kg ha $^{-1}$ for corn, 6,720 kg ha $^{-1}$ for grain sorghum, and 9,970 kg ha $^{-1}$ for rice.

⁷The 1X rate of diflufenican is 120 g ai ha⁻¹

used in analysis due to late season herbicide misapplication ¹Only yield data from 2022

letter are different according to Sidak method (α =0.05) same the s timing for each crop not containing used in analysis. 2023 Means within each evaluation Only SPAD readings from

Only yield data from 2023

SPAD meter readings

used in analysis due to bird damage were not collected for images were captured using an unmanned aerial system [DJI Mavic Air 2S (DJI Technology Co., LTD., Nanshan, Shenzhen, China)] 28 DAE. Overhead images were then analyzed using Field Analyzer (Green Research Services, LLC., Fayetteville, AR) to determine the percentage of crop groundcover. Additionally, visible injury ratings and SPAD meter readings were collected 7 and 14 DAE for simulated carryover in the greenhouse. Crop density in two 18cm sections of the row was collected 7 DAE. Visible injury ratings were collected weekly until 28 days after treatment (DAT) for the simulated sprayer contamination experiments. SPAD meter readings of five plants per plot were collected 7 and 21 DAT for all crops excluding rice. All injury ratings were conducted on a scale of 0 to 100%, with 0% representing no crop injury and 100% representing complete crop death (Frans et al., 1977).

Grain yields were collected at maturity with a small-plot combine (Kincaid 8XP, Haven, KS) adjusted to the proper moisture for each crop. The center two rows at Fayetteville and Keiser and the whole plot near Colt and Stuttgart were harvested. Seedcotton yields were determined by hand-picking the center two 3-m sections of row in Fayetteville. Additionally, aboveground biomass was collected 14 DAE in the greenhouse and air dried for three days at 66 C to a constant mass and then weighed.

2.6 Data analysis

Statistical analysis was performed using R Studio version 4.3.2 (R Core Team, 2022) using the 'glmmTMB' package (function glmmTMB; Brooks et al., 2017). Injury, SPAD, crop density, groundcover, relative yield, and biomass were fit to a generalized linear mixed-effect model (GLMM) (Stroup, 2015). Herbicide rate was considered a fixed effect, and block nested within site or year were considered random effects for the carryover experiments in the field. Subsequently, the fixed effects remained the same; however, run was considered a random effect for greenhouse experiments. Injury was evaluated by rating timing in the simulated carryover experiments. However, injury evaluations in the sprayer contamination experiments occurred seven days apart for four weeks; therefore, the data were analyzed as a repeated measure (Gbur et al., 2012). Soybean were analyzed by rating evaluation because the injury evaluations (repeated measures) were not correlated, so the least parsimonious model was selected. All injury data were bound between 0 and 1 and analyzed using a beta distribution. After the residuals failed to violate the Shapiro-Wilks normality test, SPAD, crop density, groundcover, relative yields, and crop biomass were analyzed by evaluation timing as a Gaussian or normal distribution. Analysis of variance was performed on each fitted model using the car package (Fox and Weisberg, 2019) with Type III Wald chi-square test. Estimated marginal means (Searle et al., 1980) for herbicide rates were obtained using the emmeans package (Lenth, 2022). The Sidak method was used to adjust for multiple comparisons (Midway et al., 2020) and a compact letter display was generated using the multcomp package (Hothorn et al., 2008) to represent the significantly different groups visually.

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3 Results and discussion

3.1 Crop response to simulated diflufenican carryover

Overall, results from the field trial indicated that there was minimal risk for diflufenican to carryover from soybean and injure common rotational crops. When corn was planted into various rates of diflufenican, crop injury was less than 10% by 14 DAE (Table 3). The greatest injury to corn was observed at the V3 growth stage by the highest rate of diflufenican evaluated, with an overall trend for less injury as the diflufenican rate decreased. A similar trend occurred for grain sorghum 14 DAE, with injury not exceeding 5%. Injury to grain sorghum only occurred following diflufenican at 60 and 120 g ai ha⁻¹. Injury to rice ranged from 6% to 9%, with no difference among diflufenican rates at 14 DAE. Likewise, cotton exhibited a high degree of tolerance to the diflufenican rates evaluated, with no injury 14 DAE. For the crops for which injury was observed, it existed as

TABLE 4 Influence of simulated carryover from soil-applied diflufenican on crop injury, SPAD meter readings (greenness), crop density, and biomass in the greenhouse.

		Injury	SPAD		Density	
Crop	Rate	7 DAE ^a	7 DAE	14 DAE	7 DAE	Biomass
	g ai ha ⁻¹	%	Greenness	Greenness	plants tray ⁻¹	g
Cotton	0^{b}	_	34.5	33.7	9.5	1.68
	7.5	0	34.7	34.4	9.1	1.58
	15	0	35.6	34.4	9.8	1.74
	30	0	33.6	33.8	8.8	1.57
	60	0	33.4	34.3	9.1	1.69
	120	0	33.9	32.9	9.4	1.73
	P-value	1.000	0.4548	0.6571	0.1397	0.8738
Corn	0	_	36.5	31.3	9.5	4.3
	7.5	0	39.4	30.9	9.4	4.4
	15	0	36.9	28.8	9.9	4.8
	30	0	36.8	31.4	9.5	4.7
	60	1	37.5	29.4	8.5	4.5
	120	6	35.6	29.7	9.6	4.8
	P-value	0.1345	0.1903	0.1342	0.4863	0.9553
Grain sorghum	0	_	33.3	28.1	12.8	1.13
	7.5	0	32.0	30.0	12.9	1.23
	15	0	32.5	29.6	13.0	1.22
	30	0	32.0	29.8	13.0	1.19
	60	0	32.0	28.8	12.6	1.19
	120	4	30.5	29.7	13.1	1.23
	P-value	0.1526	0.2105	0.4169	0.9407	0.9587
Rice	0	-	22.6	24.9	15.8	0.47
	7.5	0	22.1	24.2	15.8	0.51
	15	0	23.9	24.2	14.9	0.49
	30	0	24.6	23.3	16.4	0.45
	60	1	24.7	23.3	15.1	0.48
	120	3	21.9	23.4	16.3	0.48
	P-value	0.3582	0.0667	0.6563	0.8861	0.9552

^aDAE, days after emergence.

^bThe 1X rate of diflufenican is 120 g ai ha⁻¹

bleaching on the older lower leaves and a slight reduction of crop vigor, which is consistent with the typical symptomology of phytoene desaturase inhibitors (Bartel and Watson, 1978).

By 28 DAE, all crops displayed adequate tolerance with no crop injury occurring (data not shown). Additionally, no differences in leaf chlorophyll across diflufenican rates occurred at 14 or 28 DAE for any crop based on SPAD readings (Table 3). Rice density 14 DAE ranged from 174 to 200 plants m⁻², with the highest rate of diflufenican reducing the number of plants relative to the

nontreated check, with no differences among the other evaluated rates. Additionally, no differences in crop density occurred for the diflufenican rates evaluated among the other crops at 14 DAE. Likewise, groundcover for all crops at 28 DAE was comparable or statically greater for the rates of diflufenican relative to the nontreated check. Finally, no differences in relative yield arose for the diflufenican rates tested within the crops evaluated.

Like the field results, less than 10% injury was observed for all crops in the greenhouse at 7 DAE, with no differences among the

TABLE 5 Influence of simulated sprayer contamination with diflufenican:metribuzin:flufenacet premixture and evaluation timing on injury to different crops.

		Injury					
Crop	Rate	7 DAT ^a	14 DAT	21 DAT	28 DAT		
	g ai ha⁻¹		%	<i>{</i>			
Cotton	1.6 ^b	0 d ^c	0 d	0 d	0 d		
	6.4	1 d	1 d	0 d	0 d		
	25.8	15 bc	16 bc	12 c	2 d		
	103.1	30 a	32 a	24 ab	12 c		
	RM P-value ^d		0.02	295—————	· 		
Corn	1.6	0 c	0 c	0 c	0 c		
	6.4	2 c	2 c	0 c	0 c		
	25.8	8 b	2 c	0 c	0 c		
	103.1	19 a	15 ab	7 b	0 c		
	RM P-value	<0.0001					
Grain sorghum	1.6	0 b	0 b	0 b	0 b		
	6.4	0 b	0 b	0 b	0 b		
	25.8	1 a	0 b	0 b	0 b		
	103.1	4 a	2 a	0 b	0 b		
	RM P-value	<0.0001					
Soybean	1.6	0 c	1 b	0 b	0		
	6.4	2 b	2 b	0 b	0		
	25.8	4 b	3 b	3 b	1		
	103.1	18 a	13 a	13 a	5		
	P-value ^e	<0.0001	<0.0001	<0.0001	0.08		
	RM P-value						
Rice ^f	1.6	2 ab	0 b	0 b	0 b		
	6.4	0 b	0 b	0 b	0 b		
	25.8	0 b	0 b	0 b	0 b		
	103.1	3 ab	8 a	7 a	6 a		
	RM P-value						

^aDAT, days after treatment; RM, repeated measures.

^bThe diflufenican:metribuzin:flufenacet ratio is 0.17:0.35:0.48.

 c Means within herbicide rate by evaluation timing for each crop not containing the same letter are different according to Sidak method (α =0.05).

^dRM P-values were generated using the glmmTMB procedure with repeated measures in R Studio 4.3.2 using a beta distribution.

^cP-values were generated using the glmmTMB procedure without repeated measures in R Studio 4.3.2 using a beta distribution.

^fData from one site used in the analysis due to herbicide carryover from the previous growing season.

diflufenican rates tested with any crop (Table 4). By 14 DAE, no injury was elicited for the crops evaluated (data not shown), indicating the risk for diflufenican carryover is low. There were no differences in leaf chlorophyll content for the rates tested within a crop at 7 or 14 DAE. Additionally, no differences in crop density at 7 DAE or crop biomass were found for the diflufenican rates evaluated. Based on these results, more than 1.4 parts per billion of diflufenican in the soil, the highest concentration evaluated, would be required to cause concern for >10% crop response.

Previous research has evaluated the persistence of diflufenican in soil planted to wheat outside of the United States (Rouchaud et al., 1991). Typically, diflufenican was found only in the top 10 cm of soil, indicating that no leaching occurs even under periods of heavy rainfall which is likely contributed to the low solubility of diflufenican in water (Rouchaud et al., 1991) and its high lipophilicity (log P 4.9) (Knight and Kirkwood, 1991) Additionally, 40 to 60% of the soil-applied diflufenican was broken down over a 4-month period, with soil microbial activity being one form of diflufenican degradation (Rouchard et al., 1991). The estimated half-life of diflufenican in soil ranges from two to six months in winter wheat (Cramp et al., 1987) and would likely be shorter under warmer conditions typical of summer-grown crops. A shorter half-life in warmer climates is supported by Rouchaud et al. (1991) in which soil biodegradation of diflufenican was always greater during warmer periods due to the increased microbial and enzymatic soil activities. Overall, producers should have minimal concern for injury to rotational crops when using diflufenican PRE in soybean; albeit, the metribuzin and flufenacet components of Convintro may be more restrictive.

3.2 Crop response to tank contamination of a diflufenican : metribuzin:flufenacet premixture

Overall, crops do not appear highly sensitive to postemergence exposure at the V3 growth stage to sprayer contamination of the diflufenican:metribuzin:flufenacet premixture evaluated, up to a 1/ 10X rate. Cotton injury 7 DAT was no more than 30% at 7 DAT (Table 5), with injury decreasing as the rate of the diflufenican: metribuzin:flufenacet premixture decreased. A similar trend occurred in corn, with injury not exceeding 20% and decreasing as the rate of the premixture decreased. Injury to soybean was no more than 18%, increasing with the rate of the premixture increasing. Injury to rice and grain sorghum never exceeded 5% and decreased with the rate of the premixture for grain sorghum, and no differences among rates for rice. Leaf greenness for cotton was lowest for the 1/10X rate of the premixture, increasing as the herbicide rate decreased (Table 6). A similar trend was found for corn, with leaf greenness increasing as the rate of the premixture decreased. No differences in greenness were detected for grain sorghum or soybean over the rates of the premixture evaluated. Any reduction in the greenness of leaves, as measured using the SPAD meter, could be mainly attributed to the bleaching TABLE 6 Influence of simulated sprayer contamination with diflufenican:metribuzin:flufenacet premixture on SPAD meter readings and grain or seed cotton.

		SP		
Crop	Rate	7 DAT ^a Greenness	21 DAT Greenness	Relative yield ^b
	g ai ha⁻¹			%
Cotton	0 ^c	42.5ab ^d	41	-
	1.6	43.0ab	40.8	115
	6.4	44.2a	41.6	112
	25.8	38.5bc	39.9	101
	103.1	34.3c	40.7	97
	P-value	< 0.0001	0.2265	0.2505
Corn	0	35.6a	41.0	-
	1.6	35.5a	41.8	103
	6.4	35.3a	42.8	102
	25.8	29.7b	41.4	106
	103.1	27.6b	41.3	109
	P-value	< 0.0001	0.2248	0.6347
Grain sorghum	0	35.4	41.8ab	-
	1.6	36.5	43.0a	108
	6.4	36.1	41.6ab	103
	25.8	34.4	42.0ab	104
	103.1	36.0	40.8b	88
	P-value	0.0796	0.0117	0.1847
Soybean	0	38.3	39.4	-
	1.6	37.4	40.9	96
	6.4	37.8	39.3	94
	25.8	35.0	40.7	95
	103.1	38.5	40.0	94
	P-value	0.1543	0.0794	0.9751
Rice ^e	0	NA^{f}	NA	-
	1.6	NA	NA	124
	6.4	NA	NA	121
	25.8	NA	NA	125
	103.1	NA	NA	107
	P-value	NA	NA	0.2856

^aDAT, days after treatment.

^bNontreated yields for cotton was 2,720 kg ha⁻¹, 10,320 kg ha⁻¹ for corn, 5,340 kg ha⁻¹ for grain sorghum, 3,500 kg ha⁻¹ for soybean, and 9,280 kg ha⁻¹ for rice.

^cThe diflufenican:metribuzin:flufenacet ratio is 0.17:0.35:0.48.

fSPAD meter readings were not collected for rice.

^dMeans within each evaluation timing for each crop not containing the same letter are different according to Sidak method (α =0.05).

^eData from one site used in analysis due to herbicide carryover from the previous growing season.

symptomology caused by diflufenican or chlorosis from metribuzin. It is unlikely that the residual herbicide flufenacet would cause a decrease in leaf greenness when applied over the top of the crops evaluated since the main symptomology is stunting and malformation of the plant (Rowe and Penner, 1990).

Trends remained the same for cotton at 14 and 21 DAT, with injury decreasing as the rate of the premixture decreased (Table 5). The injury was transient, as evidenced by less than 15% for the 1/10X rate of the diflufenican:metribuzin:flufenacet premixture by 28 DAT. No crop response occurred from the 1/640X rate of the premixture at any evaluation timings for cotton. Injury from the 1/10X rate of the premixture was similar to the previous evaluation for corn 14 DAT. Overall, crop injury was transient, with no crop response occurring for any rates of the premixture evaluated by 28 DAT. Injury to grain sorghum remained consistent with the previous evaluation 14 DAT. Furthermore, the injury was transient, with no crop response occurring for any rates of the premixture at 21 or 28 DAT. Soybean injury was less than 15% by 21 DAT, with only the 1/40X and the 1/10X rates of the premixture eliciting a crop response. By 28 DAT, injury was ≤5% for soybean from the rates of the premixture evaluated. For rice, injury was <10% for the 1/10X rate of the premixture 14, 21, and 28 DAT, while no crop response was elicited from the additional rates evaluated.

Grain yield or seedcotton was collected at maturity for each crop evaluated and reported relative to the nontreated check (Table 6). No differences in yield occurred within a crop for the rates of the premixture evaluated. At the highest rate of the premixture evaluated (1/10X), cotton, corn, and soybean appear to have the highest risk of displaying a negative crop response, but the potential of causing high injury levels appears low. While the 1/10X rate of the premixture did elicit some crop response, the injury was transient and did not result in yield reductions for any of the evaluated crops. Overall, the sensitivity of the crops evaluated appears to be low, with injury from a 1/160X rate of the premixture never exceeding 5% at any evaluation timing.

Previous research has examined the sensitivity of crops to herbicides with high potential to injury nearby crops, such as florpyrauxifen-benzyl, dicamba, and 2,4-D. Florpyrauxifen-benzyl caused up to 96% injury to soybean at a 1/10X rate at the V3 growth stage, and up to 20% injury at a 1/500X rate 14 DAT (Miller and Norsworthy, 2018). Soybean injury from dicamba rates of less than 1/1,000X have been reported (Miller and Norsworthy, 2018; Griffin et al., 2013; Solomon and Bradley, 2014). In cotton, yield reductions of \geq 19% have occurred from a 1/1000X rate of 2,4-D when applied during vegetative development (Egan et al., 2014). Each of these published findings leads to the conclusion that there is a much lower risk of crop injury from sprayer contamination with the dilfufenican:metribuzin:flufenacet premixture.

4 Conclusions

With the anticipation of a diflufenican:metribuzin:flufenacet premixture being labeled, diflufenican will add a new SOA for soybean producers across the United States. Diflufenican will be paired with two additional SOAs, allowing producers to practice best management strategies to help slow the evolution of weed resistance (Norsworthy et al., 2012). Diflufenican will add another chemical option to integrate with other strategies aimed at the control of problematic weed species.

With the wide array of crops grown in the mid-southern U.S., injury from herbicide carryover and sprayer contamination can occur. Results from these trials indicate that the potential for diflufenican to carryover from soybean and injure rotational crops appears low. Less than 10% injury occurred for all crops from the 120 g ai ha⁻¹ (1X) rate of diflufenican, with no injury observed by the final evaluation. Greenhouse studies show that it will take more than 1.4 parts per billion of diflufenican in the soil to cause concern for crop injury. Crop response to postemergence exposure to 1/10X rate of the premixture was noted, but crops did recover from the early season injury with no yield loss. Overall, the sensitivity of the different crops to postemergence exposure to the premixture appears low, with a 1/640X rate failing to elicit a crop response. Hence, producers should be cautiously optimistic that there will be minimal risks from herbicide carryover or sprayer contamination, causing injury to subsequent or adjacent crops to soybean when utilizing the diflufenican:metribuzin:flufenacet premixture PRE.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Author contributions

MW: Conceptualization, Data curation, Formal Analysis, Investigation, Methodology, Project administration, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing. JN: Conceptualization, Data curation, Funding acquisition, Investigation, Methodology, Project administration, Resources, Software, Supervision, Validation, Writing – review & editing. LB: Conceptualization, Methodology, Writing – review & editing. TR: Conceptualization, Methodology, Writing – review & editing. BT: Conceptualization, Methodology, Writing – review & editing. LP: Conceptualization, Methodology, Visualization, Writing – review & editing. AG: Data curation, Formal Analysis, Writing – review & editing.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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