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Synergistic interactions of 2,4-D, dichlorprop-p, dicamba, and halauxifen/fluroxypyr for controlling multiple herbicide-resistant kochia (*Bassia scoparia* L.)

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Abstract

Multiple herbicide-resistant (MHR) kochia is a serious concern in the U.S. Great Plains and warrants alternative herbicide mixtures for its control. Greenhouse and field experiments were conducted at Kansas State University research and extension centers near Hays and Garden City, KS, to investigate the interactions of 2,4-D, dichlorprop-p, dicamba, and halauxifen/fluroxypyr premix in various combinations for MHR kochia control. Two previously confirmed MHR (resistant to glyphosate, dicamba, and fluroxypyr) populations and a susceptible population were tested in a greenhouse study. Kochia at the Hays field site was resistant to glyphosate and chlorsulfuron, whereas the population at Garden City was resistant to glyphosate, dicamba, and fluroxypyr. Results from a greenhouse study indicated that 2,4-D, dicamba, dichlorprop-p, and a halauxifen/fluroxypyr premix provided 26% to 69% control of both MHR populations at 28 d after treatment (DAT). However, the control increased to 85% to 97% when these herbicides were applied in three-way mixtures. Synergistic interactions were observed when dicamba was mixed with dichlorprop-p, 2,4-D, dichlorprop-p + 2,4-D, and halauxifen/fluroxypyr + 2,4-D for shoot dry weight reductions (86% to 92%) of both MHR populations. Results from a field study also indicated synergistic interactions when dicamba was mixed with dichlorprop-p + 2,4-D, halauxifen/fluroxypyr + dichlorprop-p, and halauxifen/fluroxypyr + 2,4-D, resulting in 84% to 95% control of MHR kochia at 28 DAT across both sites. These results indicate that synergistic effects of mixing dicamba with other auxinic herbicides in two- or three-way mixtures can help control MHR kochia.

Introduction

Kochia [*Bassia scoparia* (L.) A. J. Scott] is among the most troublesome summer annual broadleaf weeds in agronomic crops across the North American Great Plains (Kumar et al. 2019a). Previous studies have reported a reduction of up to 95% in sorghum [*Sorghum bicolor* (L.) Moench ssp. *bicolor*] grain yield at a kochia density of 184 plants m^{-2} in Nebraska (Wicks et al. 1994) and 23% in soybean [*Glycine max* (L.) Merr.] at a kochia density of 135 plants m^{-2} in Montana (Yadav et al. 2020). Similarly, up to 60% sugarbeet (*Beta vulgaris* L.) root yield reduction has been reported at a kochia density of 268 plants m^{-2} in Montana (Kumar and Jha 2017). More recently, Geddes and Sharpe (2022) reported a grain yield reduction of 68% in corn (*Zea mays* L.), 62% in sorghum, 46% in sugarbeet, and 23% in sunflower (*Helianthus annuus* L.) with season-long interference of kochia. However, yield reduction due to kochia interference depends on several factors, including weed density, emergence timing relative to the crop, duration of interference, and environment.

Kochia possesses several unique biological attributes that make it challenging to manage. For instance, kochia exhibits an early and extended emergence (from mid-February through mid-June), robust growth habit, salt and drought tolerance, high seed production rate (>100,000 seeds per plant), low soil seedbank persistence (1 to 2 yr), and wind-mediated seed dispersal via a tumbling mechanism (Beckie et al. 2018; Dille et al. 2017; Kumar et al. 2019a). Furthermore, the evolution of herbicide resistance among kochia populations is a serious management challenge for producers. Currently, kochia populations with resistance to four herbicide sites of action (SOAs), including inhibitors of photosystem (PS) II (a Group 5 herbicide as categorized by the Weed Science Society of America), acetolactate synthase (ALS; Group 2), 5-enolpyruvyl-shikimate-3-phosphate synthase (Group 9), and synthetic auxins (Group 4), have been reported (Heap 2023). More recently, preliminary results from Saskatchewan and North Dakota reported poor kochia control with saflufenacil, a protoporphyrinogen oxidase inhibitor (Group 14) (Anonymous 2023; Jenks 2022). The widespread resistance to glyphosate and ALS inhibitors has been reported among kochia populations across the Great Plains region. In addition, resistance to auxinic herbicides (dicamba and fluroxypyr) has also become more prevalent with frequent use of these herbicides (Heap 2023; Kumar et al. 2019a). Dicamba and fluroxypyr resistance in kochia populations can evolve together simultaneously or independently without cross-resistance (Heap 2023). For example, results from a survey conducted in Canada indicated that 13% of the kochia populations were resistant to fluroxypyr, whereas only 4% of the populations were resistant to both fluroxypyr and dicamba (Geddes et al. 2021). Evidence of three- to four-way resistance (to glyphosate, ALS inhibitors, PS II inhibitors, and/or auxinic herbicides) in kochia populations further poses a serious management challenge (Beckie et al. 2018; Kumar et al. 2019a; Varanasi et al. 2015).

Applying two or more herbicide SOAs as a mixture is generally recommended to mitigate/delay herbicide resistance in weeds (Beckie and Reboud 2009; Green 1991). However, mixed applications of auxinic herbicides with other SOAs could be antagonistic or synergistic (Barbieri et al. 2022). Previous studies indicated a synergistic interaction with mixtures of auxinic herbicides to control herbicide-resistant weeds. Torbiak et al. (2021) reported ≥90% control of glyphosate-resistant kochia with a postemergence application of bromoxynil + fluroxypyr + 2,4-D or dichlorprop-p + MCPA + mecoprop-p or fluroxypyr + 2,4-D. Agbakoba and Goodin (1970) reported a synergistic effect of 2,4-D and picloram mixture to control field bindweed (*Convolvulus arvensis* L.). Zimmer et al. (2018) reported 71%, 90%, and 94% control of glyphosate-resistant horseweed (*Erigeron canadensis* L.) with individual applications of 2,4-D, halauxifen-methyl, and dicamba, respectively; however, control increased to 97% when these three herbicides were applied as a mixture. In the same study, mixing of 2,4-D or dicamba with halauxifen-methyl also increased the control of common ragweed (*Ambrosia artemisiifolia* L.), giant ragweed (*Ambrosia trifida* L.), and redroot pigweed (*Amaranthus retroflexus* L.) in preplant burndown applications.

Auxinic herbicides (Group 4) are hormone mimics with a complex mechanism of action as compared to enzyme inhibitors (i.e., most other groups) in which an altered target site can affect the entire group of herbicides. In addition, synergistic interactions among auxinic herbicides have previously been reported with turf herbicide combination products. To our knowledge, no previous research exists in the literature that has examined the interactions of auxinic herbicides in mixtures (two- or three-way mixtures) for controlling multiple herbicide-resistant (MHR) kochia in the Great Plains. Therefore, the main objective of this study was to determine the interactions of 2,4-D, dichlorprop-p, dicamba, and halauxifen/fluroxypyr applied in two- or three-way combinations for controlling MHR kochia.

Materials and Methods

Greenhouse Study

Greenhouse experiments were conducted at Kansas State University Agricultural Research Center (KSU-ARC) near Hays, KS, in fall 2021 and repeated in spring 2022. Two previously confirmed MHR kochia populations (10A and 4H) were used. These populations were originally collected from a site near Garden City, KS, and were confirmed to be resistant to glyphosate, dicamba, and fluroxypyr (Kumar et al. 2019b). A susceptible (SUS) kochia population collected from a pasture field at KSU-ARC was included. Information on tested auxinic herbicides, their rates, combinations, and manufacturers is summarized in Table 1.

Seeds of all three kochia populations (10A, 4H, and SUS) were separately sown in plastic trays (54 by 28 by 10 cm) containing a commercial potting mixture (Miracle-Gro® Moisture Control® Potting Mix; Miracle-Gro Lawn Products, Marysville, OH). Greenhouse conditions were maintained at 25/23 ± 3 C day/night temperatures and 16/8 h (day/night) photoperiod supplemented with metal-halide lamps (560 μmol m^{-2} s^{-1}). Kochia seedlings (2 to 3 cm tall) from each population were transplanted in 10- by 10-cm square plastic pots (one seedling per pot) containing the same potting mixture as mentioned above. Seedlings were watered as needed for optimum growth of the plants. Experiments were conducted in a randomized complete block design with 12 replications (one replication = one plant per pot). Actively growing kochia seedlings (7 to 9 cm tall) from each population were treated with various herbicides (Table 1) using a stationery spray chamber (Research Track Sprayer; De Vries Manufacturing, Hollandale, MN) equipped with an even flat-fan nozzle tip (TeeJet XR8001E; Spraying Systems, Wheaton, IL) calibrated to deliver 132 L ha^{-1} of spray solution at 241 kPa. To avoid any vapor drift among treatments, all treated plants within each population were kept at 3.3 m distance apart on greenhouse benches. Kochia control (%) was visually assessed at 7, 14, and 28 d after treatment (DAT) on a scale of 0% to 100%, where 0% = no control and 100% = complete control. Visual assessments were based on herbicide injury symptoms such as epinasty (curling, twisting, and cupping), chlorosis, and necrosis of kochia seedlings. For each herbicide treatment and kochia population, the aboveground biomass of each plant was determined at 28 DAT by clipping plants at the soil surface, placing them in paper bags, and drying them at 65 C for 4 d to obtain shoot dry weights. The aboveground shoot dry weight data were converted to percent reduction of shoot dry weight using Equation 1:

$$\text{Shoot dry weight reduction (\%)} = \left[\frac{C - T}{C}\right] \times 100 \qquad [1]$$

where C is the shoot dry weight from the nontreated check treatment (average of 12 replications), and T is the shoot dry weight from a treated pot.

Field Study

Field experiments were conducted at KSU-ARC in 2021 and 2022, and Kansas State University Southwest Research and Extension Center (KSU-SWREC) near Garden City, KS, in 2022. Experiments at each site/year were conducted in a fallow field (soybean stubble in Hays and corn stubble in Garden City) to determine the interactions of auxinic herbicides applied in two- or three-way mixtures for controlling MHR kochia. The soil type at

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Table 1. Auxinic herbicides applied alone or in various mixtures tested for controlling multiple herbicide-resistant kochia under greenhouse and field experiments.

Herbicide combination[a]	Trade Name	Rate	Manufacturer
		g ai or ae ha^{-1}	
Dicamba	Clarity	560	BASF
Halauxifen/fluroxypyr	Pixxaro	5/123	Corteva Agriscience
Dichlorprop-p	Duplosan	560	Nufarm
2,4-D	Weedone LV4	538	Nufarm
Dicamba + halauxifen/fluroxypyr	Clarity + Pixxaro	560 + 5/123	BASF and Corteva Agriscience
Dicamba + dichlorprop-p	Clarity + Duplosan	560 + 560	BASF and Nufarm
Dicamba + 2,4-D	Clarity + Weedone LV4	560 + 538	BASF and Nufarm
Halauxifen/fluroxypyr + dichlorprop-p	Pixxaro + Duplosan	5/123 + 560	Corteva Agriscience and Nufarm
Halauxifen/fluroxypyr + 2,4-D	Pixxaro + Weedone LV4	5/123 + 538	Corteva Agriscience and Nufarm
Dichlorprop-p + 2,4-D	Duplosan + Weedone LV4	560 + 538	Nufarm
Dicamba + halauxifen/fluroxypyr + dichlorprop-p	Clarity + Pixxaro + Duplosan	560 + 5/123 + 560	BASF, Corteva Agriscience and Nufarm
Dicamba + halauxifen/fluroxypyr + 2,4-D	Clarity + Pixxaro + Weedone LV4	560 + 5/123 + 538	BASF, Corteva Agriscience and Nufarm
Dicamba + dichlorprop-p + 2,4-D	Clarity + Duplosan + Weedone LV4	560 + 560 + 538	BASF and Nufarm
Halauxifen/fluroxypyr + dichlorprop-p + 2,4-D	Pixxaro + Duplosan + Weedone LV4	5/123 + 560 + 538	Corteva Agriscience and Nufarm

[a]Treatments containing dichlorprop-p had nonionic surfactant (NIS) at 0.5% v/v.

Table 2. Monthly mean air temperature and total precipitation during the growing seasons at the study sites.

	Temperature			Total precipitation		
	Hays		Garden City	Hays		Garden City
	2021	2022	2022	2021	2022	2022
	C			mm		
May	16	17	18	194	86	55
June	26	24	25	20	36	32
July	27	27	28	61	45	54
August	26	26	26	84	35	10

the Hays site (38.51°N, 99.20°W) was Roxbury silt loam, pH 7.6, with 2.1% organic matter. The soil type at the Garden City site (38.00°N, 100.48°W) was Ulysses silt loam, pH 8.0, with 1.4% organic matter. The field site at the Hays location had a natural infestation of glyphosate- and chlorsulfuron-resistant kochia, whereas the field site at the Garden City location had a natural kochia infestation with confirmed multiple resistance to glyphosate, dicamba, and fluroxypyr (Kumar et al. 2019b). The same treatments as listed in Table 1 were tested across both sites. Data on mean monthly air temperature (C) and total monthly precipitation (millimeters) during each growing season at both sites were collected from nearby Kansas Mesonet (https://mesonet.k-state.edu/) weather stations and are presented in Table 2. At each site, 15 treatments were laid out in a randomized complete block design with four replications and a plot size of 3 m by 6 m. All treatments (Table 1) were applied on young actively growing kochia seedlings (8 to 10 cm tall) using a CO_2-pressurized backpack sprayer equipped with four TeeJet XR8001 flat-fan nozzles (Spraying Systems Co., Wheaton, IL), calibrated to deliver 132 L ha^{-1} of spray solution at 276 kPa. Percent visual control of kochia was recorded 7, 14, and 28 DAT on a scale of 0% to 100%, where 0% = no control and 100% = complete control. These control ratings were based on the typical auxinic herbicide injury symptoms such as epinasty (curling, twisting, and cupping), chlorosis, and necrosis of kochia seedlings in treated plots in comparison to a nontreated weedy check. At 28 DAT, kochia plants were manually clipped at the soil level from two randomly placed 1-m^2 quadrats in each plot and shoot dry weights were determined after oven-drying the samples at 65 C for 4 d. Similar to the greenhouse study, the aboveground shoot dry weight data were converted to percent reduction of shoot dry weight using Equation 1.

Statistical Analyses

All data collected from greenhouse and field studies (percent visual control and shoot dry weight reductions) were subjected to ANOVA using the MIXED procedure with SAS software (version 9.3; SAS Institute, Inc., Cary, NC). The experimental run-by-herbicide treatment and year-by-herbicide treatment interactions were not significant with $P > 0.05$ for both the greenhouse and field studies at the Hays location; therefore, data were pooled across experimental runs for the greenhouse study and across years for the Hays field study. The herbicide treatment-by-site interaction was significant ($P < 0.0001$), therefore, data for Hays and Garden City were analyzed separately. The fixed effects in the ANOVA model for greenhouse experiments included experimental runs, herbicide treatments, selected kochia populations, and their interactions. The fixed effects in the ANOVA model for field experiments included years, sites, herbicide treatments, and their interactions. The random effects in the ANOVA model included replication and all interactions involving replication for both greenhouse and field experiments. Data on percent visual control and shoot dry weight reductions were arcsine square root transformed before analysis to improve the homogeneity of variance and normality of the residuals; however, back-transformed data were presented with mean separation based on the transformed data. Treatment means were separated using Fisher's protected LSD test ($P < 0.05$). The data from nontreated plots were not included in the analyses.

For both greenhouse and field studies, the Colby equation (Equation 2) was used to calculate the expected values for two-way mixtures to determine the interaction of tested auxinic herbicides (Colby 1967):

$$E = (X + Y) - \frac{(XY)}{100} \quad [2]$$

where E is the expected kochia control or shoot dry weight reduction with the application of auxinic herbicide A + B in a mixture, and X and Y are the observed kochia control or shoot dry weight reduction with individual application of auxinic herbicide A

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and B, respectively. Similarly, the expected values for three-way mixtures were calculated using Equation 3 (Colby 1967):

$$E = (X + Y + Z) - \frac{(XY + XZ + YZ)}{100} + \frac{XYZ}{100,000} \quad [3]$$

where E is the expected kochia control or shoot dry weight reduction with application of three auxinic herbicides A + B + C in a mixture, and X, Y, and Z are the observed kochia control or shoot dry weight reduction with the individual application of auxinic herbicides A, B, and C, respectively. The expected and observed kochia control and shoot dry weight reduction for those two-way and three-way mixtures were compared using *t*-tests to determine whether those mean values differed. When the observed mean for the mixture was less than the expected mean, the interaction was considered to be antagonistic. If the observed mean was greater than the expected mean, the interaction was considered to be synergistic. The interaction of auxinic herbicide mixture was considered as additive when expected and observed means were not different ($P < 0.05$) according to the *t*-test (Colby 1967). Contrast analysis was performed to compare herbicide mixtures with dicamba vs. without dicamba, with (halauxifen/fluroxypyr) vs. without (halauxifen/fluroxypyr), with dichlorprop-p vs. without dichlorprop-p, and with 2,4-D vs. without 2,4-D.

Results and Discussion

Greenhouse Study

Percent Visual Control

The interaction between kochia populations and herbicide treatments was significant ($P < 0.0001$), indicating the differential responses of MHR and SUS populations to the tested herbicides. The three-way mixture of dicamba + halauxifen/fluroxypyr + dichlorprop-p resulted in the greatest control of MHR populations at 28 DAT; however, the control was relatively greater for the 4H population (96% ± 2.5%) than the 10A population (88% ± 2.5%; Table 3). For the SUS population, dicamba + dichlorprop-p provided the greatest control followed by dicamba + halauxifen/fluroxypyr + dichlorprop-p. The least kochia control (<50%) was obtained with the application of 2,4-D alone on both SUS and MHR populations. Several previous studies have reported poor kochia control with 2,4-D (Friesen et al. 1993; Nandula and Manthey 2002; Tonks and Westra 1997). Treatments of dichlorprop-p, dicamba, and 2,4-D alone provided only 26% to 58% control of MHR populations, but the control reached 85% to 94% when these three herbicides were mixed (Table 3). Greater control was probably due to the synergistic interaction among the three auxinic herbicides as indicated by significantly lower expected values than observed values for this mixture (Table 3). Similarly, Torbiak et al. (2021) reported >90% control of glyphosate-resistant kochia with a three-way mixture of dicamba, 2,4-D, and mecoprop-P. Synergistic interactions were also observed in our study with two-way mixtures of dicamba + 2,4-D and dicamba + dichlorprop-p resulting in 84% to 91% control of MHR populations compared to 26% to 58% control with dicamba, dichlorprop-p, or 2,4-D alone (Table 3). An antagonistic interaction was observed only for the dichlorprop-p + 2,4-D mixture for MHR populations because the observed values were significantly less than the expected values (Table 3).

Table 3. Observed and expected percent control and shoot dry weight reductions of multiple herbicide-resistant (10A and 4H) and susceptible kochia populations at 28 d after treatment in the greenhouse experiment.[a,b]

Treatment	10A				4H				Susceptible			
	Control		Shoot dry weight reduction		Control		Shoot dry weight reduction		Control		Shoot dry weight reduction	
	Observed	Expected	Observed	Expected	Observed	Expected	Observed	Expected	Observed	Expected	Observed	Expected
							%					
Dicamba	39 f	–	19 g	–	58 e	–	37 fg	–	63 g	–	73 c	–
Halauxifen/fluroxypyr	51 e	–	33 fg	–	69 d	–	51 ef	–	75 e	–	76 bc	–
Dichlorprop-p	56 de	–	26 fg	–	56 e	–	30 g	–	67 fg	–	77 bc	–
2,4-D	26 g	–	0 h	–	42 f	–	3 h	–	44 h	–	52 d	–
Dicamba + halauxifen/fluroxypyr	80 b	70*	76 bc	46*	78 c	87*	70 bcd	66	89 cd	91	89 abc	93
Dicamba + dichlorprop-p	85 ab	73*	86 ab	46*	90 ab	81*	86 ab	58*	99 a	88*	95 a	93
Dicamba + 2,4-D	84 ab	55*	88 ab	19*	91 ab	76*	86 ab	38*	91 bc	80	94 a	88*
Halauxifen/fluroxypyr + dichlorprop-p	72 c	78	53 de	49	80 c	86	76 abc	65	92 abc	92	91 ab	95
Halauxifen/fluroxypyr + 2,4-D	80 b	63*	62 cd	33	80 c	82	68 cd	52	83 d	86	92 a	89
Dichlorprop-p + 2,4-D	61 d	67*	40 ef	26	62 de	74*	56 de	32	74 ef	81	85 abc	88
Dicamba + halauxifen/fluroxypyr + dichlorprop-p	88 a	77*	89 ab	62*	96 a	74*	87 a	72	97 ab	68*	93 a	59*
Dicamba + halauxifen/fluroxypyr + 2,4-D	87 a	73*	92 a	46*	92 a	78*	85 ab	66*	97 ab	76*	94 a	71*
Dicamba + dichlorprop-p + 2,4-D	85 ab	75*	90 ab	46*	94 a	77*	84 ab	59*	93 abc	76*	91 ab	71*
Halauxifen/fluroxypyr + dichlorprop-p + 2,4-D	85 ab	77*	89 ab	49*	84 bc	77*	53 e	66	90 cd	75*	83 abc	70*

[a]Means followed by the same letter within a column are not different according to Fisher's protected LSD at $P < 0.05$.
[b]Asterisks (*) indicate that observed and expected values were different as determined by *t*-test ($P < 0.05$), indicating antagonistic or synergistic interactions of herbicides applied in mixtures based on Colby's equations (Equations 2 and 3).

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Table 4. Contrast analysis to compare various auxinic herbicide mixtures for percent control and shoot dry weight reductions of multiple herbicide-resistant (10A and 4H) and susceptible kochia populations at 28 d after treatment in the greenhouse experiment.

	10A				4H				SUS			
Contrasts[a]	Control	P-value	Shoot dry weight reduction	P-value	Control	P-value	Shoot dry weight reduction	P-value	Control	P-value	Shoot dry weight reduction	P-value
	%		%		%		%		%		%	
Dicamba vs. non-dicamba	78 vs. 61	<0.0001	77 vs. 43	<0.0001	85 vs. 68	<0.0001	75 vs. 48	<0.0001	90 vs. 75	<0.0001	90 vs. 79	<0.0001
Halauxifen/fluroxypyr vs. non-halauxifen/ fluroxypyr	78 vs. 62	<0.0001	70 vs. 50	<0.0001	83 vs. 70	<0.0001	69 vs. 54	<0.0001	89 vs. 76	<0.0001	88 vs. 81	<0.0001
Dichlorprop-p vs. non-dichlorprop-p	76 vs. 64	<0.0001	67 vs. 53	0.0003	80 vs. 73	<0.0001	67 vs. 56	0.0014	87 vs. 77	<0.0001	88 vs. 81	0.0005
2,4-D vs. non-2,4-D	73 vs. 67	0.0002	66 vs. 54	0.0040	75 vs. 75	0.0354	60 vs. 63	0.5131	82 vs. 83	0.2576	84 vs. 85	0.7034

[a]All orthogonal contrasts for percent visual control and shoot dry weight reduction (%) of each multiple herbicide-resistant and susceptible kochia population were compared using $P \leq 0.05$.

Shoot Dry Weight Reduction

Consistent with percent visual control, shoot dry weight reductions of MHR and SUS kochia populations followed a similar trend with all tested herbicides. Dicamba in mixtures with dichlorprop-p or 2,4-D alone, or in three-way mixtures with halauxifen/fluroxypyr + 2,4-D, halauxifen/fluroxypyr + dichlorprop-p, or dichlorprop-p + 2,4-D, resulted in ≥85% shoot dry weight reduction of MHR kochia populations (Table 3). Torbiak et al. (2021) also reported 73% to 84% biomass reduction of glyphosate-resistant kochia when dicamba was mixed with either fluroxypyr or 2,4-D as compared to the nontreated check. All two-way and three-way mixtures provided 83% to 95% shoot dry weight reductions of the SUS population. However, 2,4-D alone was not effective and provided only 0% to 3% shoot dry weight reduction of both MHR populations and 52% reduction of the SUS population relative to the nontreated. Synergistic interactions were observed for shoot dry weight reductions of MHR populations when dicamba was mixed with dichlorprop-p, 2,4-D, and dichlorprop-p + 2,4-D (Table 3). Furthermore, contrast analysis between dicamba vs. non-dicamba, dichlorprop-p vs. non-dichlorprop-p, and halauxifen/fluroxypyr vs. non-halauxifen/fluroxypyr mixtures were significant for percent kochia control and shoot dry weight reductions, indicating the importance of co-applications of these auxinic herbicides in mixtures for effective control of MHR kochia (Table 4).

Field Experiments

Monthly mean air temperatures during the study period (May to August) at KSU-ARC near Hays, KS, ranged from 16 to 26 C with a total precipitation of 359 mm in 2021 and 17 to 26 C with a total precipitation of 202 mm in 2022 (Table 2). Monthly mean air temperatures at KSU-SWREC near Garden City, KS were also similar and ranged from 18 to 26 C during the study period in 2022. However, the Garden City site was relatively drier than the Hays site, with a total precipitation of 151 mm during the study period.

Percent Visual Control

Mixing dicamba with dichlorprop-p, dichlorprop-p + 2,4-D, halauxifen/fluroxypyr, halauxifen/fluroxypyr + 2,4-D, and halauxifen/fluroxypyr + dichlorprop-p resulted in 90% to 96% control of MHR kochia at 14 and 28 DAT at Hays (Table 5). Based on Colby's equation, the expected kochia control was significantly less than the observed values for dicamba + halauxifen/fluroxypyr + 2,4-D, dicamba + halauxifen/fluroxypyr + dichlorprop-p, and dicamba + dichlorprop-p + 2,4-D at 28 DAT, indicating synergistic interactions of these mixtures (Table 5). A recent study also reported ≥90% control of glyphosate-resistant kochia with mixtures of bromoxynil + fluroxypyr + 2,4-D, dichlorprop-p + MCPA + mecoprop-p, or fluroxypyr + 2,4-D (Torbiak et al. 2021). The least kochia control (23% to 30%) was observed with 2,4-D alone at 14 and 28 DAT.

At the Garden City site, mixtures of dicamba with dichlorprop-p + 2,4-D, halauxifen/fluroxypyr, halauxifen/fluroxypyr + dichlorprop-p, and halauxifen/fluroxypyr + 2,4-D resulted in ≥84% control of MHR kochia at 28 DAT (Table 6). Based on Colby's equation, synergistic interactions for MHR kochia control were observed when dicamba was mixed with 2,4-D, dichlorprop-p + 2,4-D, halauxifen/fluroxypyr, halauxifen/fluroxypyr + dichlorprop-p, and halauxifen/fluroxypyr + 2,4-D at 28 DAT (Table 6). However, antagonistic interactions were observed for dichlorprop-p + 2,4-D and halauxifen/fluroxypyr + dichlorprop-p mixtures for controlling MHR kochia at both sites (Tables 5 and 6). The least control (13%) of MHR kochia was observed when 2,4-D was applied alone. The contrast analysis between dicamba vs. non-dicamba, halauxifen/fluroxypyr vs. non-halauxifen/fluroxypyr, dichlorprop-p vs. non-dichlorprop-p, and 2,4-D vs. non-2,4-D mixtures were significant for MHR kochia control at 28 DAT at both sites, indicating the importance of co-applications of these herbicides in mixtures for effective kochia control (Table 7).

Shoot Dry Biomass Reduction

The greatest shoot dry weight reduction (97%) of MHR kochia was observed with the three-way mixture of dicamba + halauxifen/fluroxypyr + 2,4-D at the Hays site (Table 5). Furthermore, mixing dicamba with dichlorprop-p + 2,4-D, halauxifen/fluroxypyr, and halauxifen/fluroxypyr + dichlorprop-p reduced shoot dry weights of MHR kochia by ≥90%. Based on Colby's equation, dicamba + halauxifen/fluroxypyr + 2,4-D and dicamba + dichlorprop-p + 2,4-D mixtures showed synergistic interactions for controlling MHR kochia. The least shoot dry weight reductions (44% to 48%) were observed when dichlorprop-p + 2,4-D and 2,4-D were applied alone. The contrast analysis showed greater shoot dry weight reduction (82%) of MHR kochia when dicamba was added to the mixture compared to no dicamba in the mixture (55%; Table 7). Similarly, 74% shoot dry weight reduction was obtained when halauxifen/ fluroxypyr was added to the mixture compared to a 62% reduction in shoot dry weights of MHR kochia without

Table 5. Observed and expected percent control and shoot dry weight reduction of glyphosate- and chlorsulfuron-resistant kochia with different auxinic herbicides applied alone or in mixtures in a field study during 2021 and 2022 at KSU-ARC near Hays, KS.[a,b,c]

		Control				Shoot dry weight reduction	
		14 DAT		28 DAT			
Treatments	Rate	Observed	Expected	Observed	Expected	Observed	Expected
	g ai or ae ha-1			%		%	
Dicamba	560	63 df	–	67 dfg	–	63 cde	–
Halauxifen/fluroxypyr	5/123	65 cdf	–	69 df	–	50 e	–
Dichlorprop-p	560	67 bcd	–	64 fg	–	51 e	–
2,4-D	538	23 g	–	30 h	–	48 e	–
Dicamba + halauxifen/fluroxypyr	560 + 5/123	92 a	87	94 a	90	90 ab	78
Dicamba + dichlorprop-p	560 + 560	92 a	88	90 a	88	73 bcd	83
Dicamba + 2,4-D	560 + 538	74 bc	71	80 b	77	63 cde	80
Halauxifen/fluroxypyr + dichlorprop-p	5/123 + 560	72 bcd	88*	74 bcd	89*	55 de	79*
Halauxifen/fluroxypyr + 2,4-D	5/123 + 538	71 bcd	73	71 cdf	78	52 de	72
Dichlorprop-p + 2,4-D	560 + 538	56 f	74*	60 g	75*	44 e	75*
Dicamba + halauxifen/fluroxypyr + dichlorprop-p	560 + 5/123 + 560	94a a	71*	95 a	70*	91 ab	77
Dicamba + halauxifen/fluroxypyr + 2,4-D	560 + 5/123 + 538	95a a	82*	96 a	80*	97 a	71*
Dicamba + dichlorprop-p + 2,4-D	560 + 560 + 538	93 a	82*	95 a	80*	95 a	78*
Halauxifen/fluroxypyr + dichlorprop-p + 2,4-D	5/123 + 560 + 538	75 b	82	78 bc	80	84 abc	78

[a]Abbreviation: KSU-ARC, Kansas State University Agricultural Research Center.
[b]Means followed by the same letter within a column are not different according to Fisher's protected LSD at P < 0.05.
[c]Asterisks (*) indicate that observed and expected values were different as determined by *t*-test (P < 0.05), indicating antagonistic or synergistic interactions of herbicides applied in mixtures based on Colby's equations (Equations 2 and 3).

Table 6. Observed and expected percent control and shoot dry weight reduction of multiple herbicide-resistant kochia with different auxinic herbicides applied alone or in mixtures in a field study during 2022 at KSU-SWREC near Garden City, KS.[a,b,c]

		Control				Shoot dry weight reduction	
		14 DAT		28 DAT			
Treatments	Rate	Observed	Expected	Observed	Expected	Observed	Expected
	g ae or ai ha-1	%				%	
Dicamba	560	35 f	–	45 j	–	78 abc	–
Halauxifen/fluroxypyr	5/123	45 e	–	58 gi	–	85 abc	–
Dichlorprop-p	560	33 f	–	63 fg	–	70 bc	–
2,4-D	538	10 g	–	13 k	–	48 d	–
Dicamba + halauxifen/ fluroxypyr	560 + 5/123	75 ab	64	86 abc	77*	88 abc	96
Dicamba + dichlorprop-p	560 + 560	70 abc	56*	80 cd	79	83 abc	92
Dicamba + 2,4-D	560 + 538	50 e	42	70 ef	52*	71 bc	87
Halauxifen/fluroxypyr + dichlorprop-p	5/123 + 560	60 cd	63	73 e	84*	72 bc	95
Halauxifen/fluroxypyr + 2,4-D	5/123 + 538	68 bc	51*	78 de	63*	81 abc	90
Dichlorprop-p + 2,4-D	560 + 538	53 de	39	53 i	67*	70 bc	79
Dicamba + halauxifen/ fluroxypyr + dichlorprop-p	560 + 5/123 + 560	78 a	71	91 a	76*	94 a	58*
Dicamba + halauxifen/ fluroxypyr + 2,4-D	560 + 5/123 + 538	78 a	67*	89 ab	76*	87 abc	67
Dicamba + dichlorprop-p + 2,4-D	560 + 560 + 538	75 ab	60*	84 bcd	78*	80 abc	75
Halauxifen/fluroxypyr + dichlorprop-p + 2,4-D	5/123 + 560 + 538	75 ab	65*	73 e	81	91 a	74

[a]Abbreviation: KSU-SWREC, Kansas State University Southwest Research and Extension Center.
[b]Means followed by the same letter within a column are not different according to Fisher's protected LSD at P < 0.05.
[c]Asterisks (*) indicate that observed and expected values were different as determined by *t*-test (P < 0.05), indicating antagonistic or synergistic interactions of herbicides applied in mixtures based on Colby's equations (Equations 2 and 3).

halauxifen/fluroxypyr (Table 7). These results indicate the importance of dicamba and halauxifen/fluroxypyr in the mixtures for effective shoot dry weight reduction of MHR kochia.

At the Garden City location, dicamba + halauxifen/fluroxypyr + dichlorprop-p and halauxifen/fluroxypyr + dichlorprop-p + 2,4-D provided >90% shoot dry weight reduction of MHR kochia (Table 6). All the two-way or three-way mixtures resulted in ≥80% shoot dry weight reductions, except dichlorprop-p + 2,4-D and halauxifen/fluroxypyr + dichlorprop-p. Consistent with percent visual control, 2,4-D had the least (48%) shoot dry weight reduction of MHR kochia. The expected reduction in shoot dry weights of MHR kochia with dicamba + halauxifen/fluroxypyr + dichlorprop-p was significantly lower than the observed value, indicating synergistic interaction of the mixture. The contrast analysis further indicated that co-applications of dicamba or halauxifen/fluroxypyr in the mixtures resulted in 9% to 14% more shoot dry weight reduction of MHR kochia (Table 7). Torbiak et al. (2021) previously reported 99% biomass reduction of glyphosate-resistant kochia with a three-way mixture of dicamba, 2,4-D, and mecoprop-P.

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Table 7. Contrast analysis to compare various auxinic mixtures for percent control and shoot dry weight reductions of multiple herbicide-resistant kochia at 28 d after treatment in field studies conducted at KSU-ARC near Hays, KS and KSU-SWREC near Garden City, KS.[a]

	Hays				Garden City			
Contrasts[b]	Control	p-value	Shoot dry weight reduction	p-value	Control	p-value	Shoot dry weight reduction	p-value
	%		%		%		%	
Dicamba vs non-dicamba	88 vs 64	<0.0001	82 vs 55	<0.0001	78 vs 58	<0.0001	83 vs 74	0.0820
Halauxifen/fluroxypyr vs non-halauxifen/fluroxypyr	82 vs 69	<0.0001	74 vs 62	0.0051	78 vs 58	<0.0001	85 vs 71	0.0069
Dichlorprop-p vs non-dichlorprop-p	79 vs 72	<0.0001	70 vs 66	0.3168	74 vs 63	<0.0001	80 vs 77	0.5635
2,4-D vs non-2,4-D	73 vs 79	0.0001	69 vs 68	0.7239	65 vs 71	0.0003	75 vs 81	0.2482

[a]Abbreviations: KSU-ARC, Kansas State University Agricultural Research Center; KSU-SWREC, Kansas State University Southwest Research and Extension Center.
[b]All orthogonal contrasts for percent visual control and shoot dry weight reduction (%) at each experimental site were compared using $P \leq 0.05$.

Practical Implications

Results from the current study suggest that mixtures of auxinic herbicides, including dicamba + halauxifen/fluroxypyr + dichlorprop-p, dicamba + halauxifen/ fluroxypyr + 2,4-D, and dicamba + dichlorprop-p + 2,4-D were synergistic for control of MHR kochia. Furthermore, two-way mixtures such as dicamba + halauxifen/fluroxypyr or dicamba + dichlorprop-p resulted in greater control of MHR kochia compared to their standalone treatments. Considering the rapid spread of MHR kochia coupled with dwindling new herbicide SOAs, the synergistic interactions of auxinic herbicides in two- or three-way mixtures tested in this study can play a crucial role in managing MHR kochia populations (especially those that are resistant to dicamba/fluroxypyr) in fallow-fields or burndown (prior to crop planting or after crop harvesting) after careful considerations of the rotational crops. Development of new crop trait technologies with resistance to multiple auxinic herbicides (such as 2,4-D, dicamba, dichlorprop, or fluroxypyr) can also allow the use of these auxinic mixtures for in-season control of MHR kochia populations. However, proper auxinic herbicide stewardship guidelines, including vapor or physical drift mitigation practices, need to be followed to avoid off-target movement of these auxinic mixtures. It is also important to note that overreliance on these mixtures should be avoided to further prevent evolution of cross-resistance to auxinic herbicides among kochia populations. Growers should adopt these effective auxinic-based mixtures along with other effective weed control tactics, including cultural (competitive crop rotations, optimum time of crop planting, narrow crop row spacing, cover crops, etc.), mechanical (occasional/strategic tillage, harvest weed seed control, etc.), and precision spray technologies as part of integrated weed management strategies for kochia control. Future studies will investigate the possible underlying mechanism(s) of synergistic interactions among auxinic herbicides in various mixtures for MHR kochia control.

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