

Integrated strategies to manage herbicide-resistant kochia and Palmer amaranth in Kansas
cropping systems

by

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B.S., CCS Haryana Agricultural University, 2018
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AN ABSTRACT OF A DISSERTATION

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Abstract

Kochia [*Bassia scoparia* (L.) A. J. Scott] and Palmer amaranth (*Amaranthus palmeri* S. Watson) are the two most troublesome summer annual weeds in the Central Great Plains (CGP). The widespread evolution of herbicide resistance in both species warrants alternate weed management strategies in the region. The major objectives of this dissertation were to 1) determine the response of kochia populations from Kansas, Oklahoma, and Texas to commonly used herbicides and quantify the level of resistance in putative multiple herbicide-resistant (MHR) kochia populations, 2) determine the interactions of 2,4-D, dichlorprop-p, dicamba, and halauxifen/fluroxypyr applied in two- or three-way combinations for controlling MHR kochia, 3) evaluate pyridate-based tank mixtures for controlling MHR kochia, 4) determine the effect of fall-planted cover crop (CC) and soil residual herbicides on emergence pattern and emergence periodicity of glyphosate-resistant Palmer amaranth, and 5) determine the effect of fall-planted CC in combination with soil residual herbicides on weed suppression and on grain yield and net returns in no-till dryland grain sorghum. Greenhouse screening experiments showed that 23% of tested kochia populations (82 total) from Kansas, Oklahoma, and Texas were potentially four-way resistant ($\geq 20\%$ survival) to chlorsulfuron, dicamba, fluroxypyr, and glyphosate. Dose-response studies further revealed resistance to dicamba (5- to 13-fold), fluroxypyr (3- to 6-fold), and glyphosate (3- to 5-fold) in kochia populations from Kansas and Oklahoma and resistance to dicamba (2- to 4-fold) and glyphosate (5-fold) in Texas populations compared to susceptible population. Three-way tank-mixtures of 2,4-D, dicamba, dichlorprop-p, and halauxifen/fluroxypyr in various combinations had synergistic interactions and provided 85 to 97% control of MHR kochia. Results from greenhouse experiments also showed synergistic interactions when pyridate was tank-mixed with atrazine, dicamba, dichlorprop-p, fluroxypyr,

glyphosate, or halauxifen/fluroxypyr and resulted in >94% control and shoot dry biomass reduction of MHR kochia. In a separate field study, a synergistic interaction was observed when pyridate was tank-mixed with glyphosate or atrazine. Results from a 3-year field study showed that fall-planted CC after winter wheat harvest and terminated before grain sorghum planting with glyphosate plus acetochlor/atrazine required 105 to 1257 more cumulative growing degree days for 90% emergence of glyphosate-resistant Palmer amaranth and reduced the cumulative emergence by 42 to 56% compared to chemical fallow. Furthermore, the same treatment reduced total weed density by 34 to 81% and total weed biomass by 45 to 73% compared to chemical fallow. Average grain sorghum yield was 790 to 1430 kg ha⁻¹ and did not differ between chemical fallow and CC terminated with glyphosate plus acetochlor/atrazine. However, net returns were lower with both CC treatments (USD -\$275 to \$66 ha⁻¹) in all three years than chemical fallow (USD -\$111 to \$120 ha⁻¹). These results suggest that integrating a fall-planted CC after wheat harvest can help suppress glyphosate-resistant kochia and Palmer amaranth in the subsequent grain sorghum. This practice was not profitable most years but could be profitable during wet years with average yields (3800 to 5000 kg ha⁻¹) of the region. Overall, the results confirm the widespread presence of MHR kochia populations in the southcentral Great Plains. Growers should adopt alternative herbicide tank-mixtures and CC strategies to manage herbicide-resistant kochia and Palmer amaranth populations in the region.

Keywords: *Amaranthus palmeri* S. Watson, *Bassia scoparia* (L.) A. J. Scott, cover crop, herbicide mixtures, integrated weed management

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Dedication

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Chapter 1 - Literature Review

Kochia [*Bassia scoparia* (L.) A. J. Scott] and Palmer amaranth (*Amaranthus palmeri* S. Watson) are two major troublesome weeds in the Central Great Plains (CGP). Both weeds compete with crops for resources such as nutrients, water, light, and space and reduce the crop yield. Both are summer annual broadleaf weeds but kochia can emerge early in the spring with an extended period of emergence (mid-Feb through mid-June) (Dille et al. 2017; Friesen et al. 2009; Kumar et al. 2018). Both weeds have several characteristics which make them problematic and hard to control in agronomic crops in the U.S.

Kochia is a C₄ summer annual broadleaf weed in the Chenopodiaceae family and has been reported as the second most troublesome weed in alfalfa (*Medicago sativa* L.), canola (*Brassica napus* L.), and spring cereals (Van Wychen 2022). Kochia is tolerant to abiotic stresses (heat, drought, and salinity), a prolific seed producer (one plant can produce >100,000 seeds), and exhibits wind-mediated tumble mechanism of seed dispersal (Christoffoleti et al. 1997; Friesen et al. 2009; Kumar et al. 2019). Season-long interference of kochia can reduce grain yield by 68% in corn (*Zea mays* L.), 62% in sorghum [*Sorghum bicolor* (L.) Moench ssp. *bicolor*], 46% in sugarbeet (*Beta vulgaris* L.), and 23% in sunflower (*Helianthus annuus* L.) (Geddes and Sharpe 2022). Similarly, kochia at densities of 240 to 520 plants m⁻² has been reported to reduce spring wheat (*Triticum aestivum* L.) grain yield by 60% (Friesen et al 1991). In addition, kochia plants escaping post-emergence herbicide applications in the spring or late-emerging cohorts can also hinder mechanical wheat harvest (Kumar and Jha 2015; Torbiak et al. 2021).

Palmer amaranth is a summer annual C₄ dioecious plant belonging to the Amaranthaceae family. It is native to the southwestern United States and northwestern Mexico and is widely

distributed in several countries across Africa, Asia, Europe, North America, and South America (Roberts and Florentine 2022; Sauer 1957). It exhibits an extended emergence period (late April to early September), high photosynthetic rate, prolific seed production (up to 600,000 seeds per female plant), high genetic diversity within and among field populations, and ability to evolve herbicide resistance (Chahal et al. 2015; Horak and Loughin 2000; Keeley et al. 1987; Ward et al. 2013). Palmer amaranth can cause significant yield losses in several crops. For instance, grain yield reduction up to 91% in corn and 79% in soybean [*Glycine max* (L.) Merr.] has been reported with Palmer amaranth interference at a density of 8 plants m⁻¹ of row (Klingaman and Oliver 1994; Massinga et al. 2001). Similarly, Palmer amaranth caused yield reduction up to 28% in peanut (*Arachis hypogaea* L.) at 1 plant m⁻¹ of row (Burke et al. 2007), and 54% in cotton (*Gossypium hirsutum* L.) at a density of 10 plants per 9.1 m of row (Morgan et al. 2001). Moore et al. (2004) reported that grain sorghum yield was reduced by 1.8 to 3.5% with one Palmer amaranth plant in 15 m of sorghum row.

Evolution of multiple herbicide resistance in kochia and Palmer amaranth is a serious management concern. Currently, kochia populations resistant to five different herbicide sites of action (SOA) (WSSA Groups 2, 4, 5, 9, and 14) have been documented (Heap 2024). Similarly, resistance to 10 different herbicide SOA has been identified in Palmer amaranth (WSSA Groups 2, 3, 4, 5, 6, 9, 10, 14, 15, and 27) (Heap 2024). Multiple herbicide-resistant (MHR) kochia and Palmer amaranth populations have been reported in the CGP region, including Kansas (Chahal et al. 2015; Heap 2024; Jhala et al. 2014; Kumar et al. 2019; 2020a).

Applying two or more herbicide SOA as a mixture is generally recommended to mitigate/delay herbicide resistance in weeds (Beckie and Reboud 2009; Green 1991). Auxinic herbicides (WSSA Group 4) are commonly used to control broadleaf weeds in a variety of crops

since the discovery of 2,4-D in the mid-1940s (Busi et al. 2018). The incidence of weed species resistant to auxinic herbicides is relatively low considering their long-term global application (Busi et al. 2018). The application of auxinic herbicides in combination with other SOA as herbicide mixtures 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. For instance, Torbiak et al. (2021) reported $\geq 90\%$ control of glyphosate-resistant kochia with a post-emergence application of bromoxynil + fluroxypyr + 2,4-D or dichlorprop-p + MCPA + mecoprop-p or fluroxypyr + 2,4-D. Similarly, 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 pre-plant burndown applications. In addition, synergistic interactions among auxinic herbicides have previously been reported with turf herbicide combination products (Ni et al. 2006).

Pyridate is a contact, post-emergence herbicide that belongs to WSSA Group 6. It controls both grass and broadleaf weeds by inhibiting electron transfer due to blocking amino acid histidine 215 at the A site of PS II (Székács 2021). Tonks and Westra (1997) have previously reported 16 to 60% control of sulfonyleurea-resistant kochia with pyridate alone. Wicks et al. (1994) reported 89% control of atrazine-resistant kochia with pyridate alone at 21 days after treatment (DAT). Wicks et al. (1993) reported $>95\%$ control of atrazine-resistant kochia

with pyridate + flurochloridone at 21 to 30 DAT, indicating the benefits of adding pyridate in herbicide mixtures.

Integration of cover crops (CC) in crop rotations has been proven as one of the effective tools to suppress herbicide-resistant weeds in the semiarid CGP region (Kumar et al. 2020b; Mesbah et al. 2019; Obour et al. 2022; Petrosino et al. 2015). In addition to weed suppression, growing CC in the semiarid CGP provides other benefits including reduced soil erosion, enhanced nutrient cycling, increased microbial activity, improved soil health, and increased plant diversity and pollinator resources (Blanco-Canqui et al. 2011, 2013; Simon et al. 2022). The CC residue on the soil surface could reduce the soil temperature in summer and reduce evaporation, thereby contributing to increased soil water storage (Holman et al. 2021a, 2021b). However, replacing the fallow period with CC in the semiarid cropping systems sometimes reduces the yield of successive crops because of the reduced plant available water (Holman et al. 2018; Nielsen et al. 2016). Previous studies have evaluated the effect of spring-planted CC on weed suppression and winter wheat yields when CC replaced the fallow phase of winter wheat-grain sorghum-fallow (W-S-F) rotation in this region (Holman et al. 2022; Mesbah et al. 2019; Obour et al. 2022). Obour et al. (2022) reported that spring-planted CC (oat/triticale/spring peas) in W-S-F rotation can reduce weed biomass by 86 to 99% compared to weedy fallow. Holman et al. (2022) reported that spring-planted CC had no significant effect on wheat and grain sorghum yields when conditions were either extremely dry with poor yields or very wet with above-average yields. However, the authors reported that replacing fallow with CC increased the cost of production by 16 to 97% compared to fallow.

Several researchers have documented the importance of residual herbicides in combination with CC to achieve season-long weed control (Perkins et al. 2021; Whalen et al.

2020). For instance, Whalen et al. (2020) reported that CC terminated with glyphosate plus 2,4-D in combination with residual herbicides (sulfentrazone plus chlorimuron) resulted in greater waterhemp [*Amaranthus tuberculatus* (Moq.) J. D. Sauer.] control (73 to 84%) compared to no residual herbicide (44 to 65%). Teasdale et al. (2005) reported that hairy vetch (*Vicia villosa* Roth.) residue (500 g m⁻²) and metolachlor (10 g ha⁻¹) had a synergistic effect and reduced smooth pigweed (*Amaranthus hybridus* L.) emergence by 86% compared to no metolachlor or no hairy vetch residue. Most of these research studies were conducted in cotton (*Gossypium hirsutum* L.), corn, or soybean in greater precipitation environments.

Combination of cultural and mechanical practices (CC, tillage, etc.) as well as herbicides are also known to influence the periodicity and emergence pattern of weed species. For instance, DeVore et al. (2013) reported a 94% reduction in glyphosate-resistant Palmer amaranth emergence when cereal rye (*Secale cereale* L.) CC was used in combination with deep tillage (30-cm depth) in soybean compared to no deep tillage or cereal rye CC in Arkansas. Palhano et al. (2018) reported that cereal rye CC reduced Palmer amaranth emergence by 83% in cotton (*Gossypium hirsutum* L.) as compared to no CC in Arkansas. Perkins et al. (2021) reported that CC mixture of cereal rye and hairy vetch in combination with pyroxasulfone + flumioxazin, flumioxazin, pyroxasulfone, or acetochlor delayed Palmer amaranth emergence by 27 to 34 days in soybean compared to CC with no residual herbicides. Similarly, Liu et al. (2022) documented a differential emergence pattern and periodicity among Palmer amaranth populations from Kansas, Nebraska, Colorado, Oklahoma, and Texas in a no-tillage common garden field study in western Kansas.

Given the topics mentioned above, the major objectives of this dissertation were to 1) determine the response of kochia populations from Kansas, Oklahoma, and Texas to commonly

used herbicides and quantify the level of resistance in putative MHR kochia populations, 2) determine the interactions of 2,4-D, dichlorprop-p, dicamba, and halauxifen/fluroxypyr applied in two- or three-way combinations for controlling MHR kochia, 3) evaluate the pyridate-based tank mixtures for controlling MHR kochia, 4) determine the effect of fall-planted CC and soil residual herbicides on emergence pattern and emergence periodicity of glyphosate-resistant Palmer amaranth, and 5) determine the effect of fall-planted CC in combination with soil residual herbicides on weed suppression and on grain yield and net returns in no-till dryland grain sorghum.

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Figure 1.1. Kochia plants (A) and heavy infestation of kochia in grain sorghum at 20 days after sorghum planting (B) at Kansas State University Agricultural Research Center near Hays, KS.



Figure 1.2. Palmer amaranth plants (A) and heavy infestation of Palmer amaranth in grain sorghum at 30 days after sorghum planting (B) at Kansas State University Agricultural Research Center near Hays, KS.

Chapter 2 - Distribution and characterization of herbicide resistance in kochia populations from southcentral Great Plains

Abstract

Herbicide-resistant kochia is a major challenge for growers for sustainable crop production. Matured seeds of 82 kochia populations were randomly collected in fall of 2018 and 2019 from western Kansas (KS), western Oklahoma (OK), and northern Texas (TX) to determine the frequency and distribution of resistance to atrazine, chlorsulfuron, dicamba, fluroxypyr, and glyphosate. Total 50 kochia seedlings (6 to 9 cm tall) from each population were tested with each selected herbicide using a field-use rate. The survival frequency (number of alive seedlings/total number of seedlings screened) was determined at 28 days after treatment. Results from single-dose experiments showed that 33%, 100%, 48%, 30%, and 70% of screened kochia populations were potentially resistant ($\geq 20\%$ survival) to atrazine, chlorsulfuron, dicamba, fluroxypyr, and glyphosate, respectively. Dose-response experiments revealed that two kochia populations from KS (KS 19-2 and KS 19-7) were 5- to 10-fold resistant to dicamba, 3- to 6-fold resistant to fluroxypyr, and 4- to 5-fold resistant to glyphosate as compared to the SUS population (KS-SUS). Similarly, two populations from OK (OK 20 and OK 21) were 10- to 13-fold resistant to dicamba, 3- to 4-fold resistant to fluroxypyr, and 3- to 4-fold resistant to glyphosate compared to the SUS population (OK-SUS). Two kochia populations from TX (TX 18-52 and TX 19-65) were 2- to 4-fold resistant to dicamba and one population (TX 18-52) was 5-fold resistant to glyphosate compared to the SUS population (TX-SUS). These results confirm the widespread distribution of multiple herbicide-resistant kochia in the southcentral Great Plains.

Keywords: *Bassia scoparia* (L.) A. J. Scott; multiple herbicide resistance; resistance index; U.S.
Great Plains

Introduction

Kochia [*Bassia scoparia* (L.) A. J. Scott] is a major troublesome summer annual broadleaf weed of the Chenopodiaceae family (Kumar et al. 2019a). Kochia can emerge early in the spring and exhibits an extended emergence period from mid-Feb through mid-June (Dille et al. 2017; Friesen et al. 2009; Kumar et al. 2018). Small kochia seedlings have leaves that are densely covered in short, fine, white hairs. These leaf hairs can reduce foliar absorption of herbicides by suspending droplets above the leaf cuticle and thus applications are recommended to larger plants (Friesen et al. 2009). Kochia is tolerant to heat, drought, and salinity stresses, which led to its adaptation and widespread distribution in the U.S. Great Plains (Christoffoleti et al. 1997; Friesen et al. 2009). Kochia flowers are inconspicuous (without petals and are protogynous) which means stigmas emerge prior to anther development and this facilitates cross-pollination between plants. Each kochia plant can produce >100,000 seeds that can be dispersed over long distances by wind via a tumbling mechanism (Christoffoleti et al. 1997; Friesen et al. 2009; Kumar et al. 2019a).

Kochia is a highly competitive weed in agronomic crops because of its early emergence and stress-tolerant ability. It can cause a yield reduction in many field crops including corn (*Zea mays* L.), grain sorghum [*Sorghum bicolor* (L.) Moench ssp. *bicolor*], soybean [*Glycine max* (L.) Merr.], sugarbeet (*Beta vulgaris* L.), sunflower (*Helianthus annuus* L.), alfalfa (*Medicago sativa* L.), canola (*Brassica napus* L.), and spring cereals (Geddes et al. 2022; Kumar and Jha 2015; Lewis and Gulden 2014; Wicks et al. 1994, 1997). The amount of yield loss depends on the kochia density; for instance, kochia can cause a 95% yield reduction in grain sorghum at a density of 184 plants m⁻² (Wicks et al. 1994), 23 to 77% in soybean at 20 to 135 plants m⁻² (Geddes et al. 2022; Wicks et al. 1997), 60% in sugarbeet at 268 plants m⁻² (Kumar and Jha

2015), and 62 to 95% in sunflower at 34 to 905 plants m⁻² (Lewis and Gulden 2014). Continuous and repeated use of herbicides with same site of action (SOA) resulted in the evolution of herbicide-resistant kochia (Kumar et al. 2019a; Sharpe et al. 2023; Westra et al. 2019).

Currently, kochia populations have evolved resistance to five different SOA, including inhibitors of acetolactate synthase (ALS) (Group 2), photosystem (PS) II (Group 5), 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS) (Group 9), protoporphyrinogen oxidase (PPO) (Group 14), and synthetic auxins (Group 4) (Heap 2024). Furthermore, a single kochia population has been reported from Kansas with multiple resistance to four herbicide SOA, including chlorsulfuron (Group 2), dicamba (Group 4), atrazine (Group 5), and glyphosate (Group 9) (Heap 2024). The widespread distribution of glyphosate-resistant kochia populations has been previously reported across various states in the Great Plains (Godar et al. 2015; Kumar et al. 2019a). However, the current status on distribution and frequency of MHR kochia in the southcentral Great Plains (SGP) is unknown. Therefore, the main objectives of this study were to (1) determine the response of kochia populations from Kansas, Oklahoma, and Texas to atrazine, chlorsulfuron, dicamba, dichlorprop/dicamba/2,4-D, fluroxypyr, and glyphosate, and (2) quantify the level of resistance to dicamba, glyphosate, and fluroxypyr in selected putative MHR kochia populations.

Materials and Methods

Seed Source

Matured seeds of 82 different kochia populations were collected during fall of 2018 and 2019 from pasture, wheat stubble, and fallow fields with a history of corn, cotton, grain sorghum, and wheat production across Kansas (n=13), Oklahoma (n=19), and Texas (n=58) (Appendix Table A.1). Geo-referenced collection sites for these populations were mapped. Seeds from each

population were cleaned with a combination of sieves and an air-blower and stored in plastic bags at 4 C for subsequent experiments.

Single-Dose Experiment

Seeds from each kochia population were separately sown in 50-cell plastic trays (54- by 28- by 10-cm) containing commercial potting mixture (Miracle-Gro[®] Moisture Control[®] Potting Mix, Miracle-Gro Lawn Products, Scottslawn Road, Marysville, OH) at Kansas State University Agricultural Research Center (KSU-ARC) near Hays, KS. The temperature in the greenhouse was maintained at 25/23 ± 3 C day/night and a 16/8 h (day/night) photoperiod supplemented with metal-halide lamps (560 $\mu\text{mol m}^{-2} \text{s}^{-1}$). Seedlings were watered daily to maintain optimum moisture for growth. Each tray with 50 seedlings from each population was separately treated with glyphosate (Roundup PowerMax, Bayer Crop Science) at 1260 g ha⁻¹ plus 2% w/v ammonium sulfate (AMS), chlorsulfuron (Glean XP, FMC Corporation) at 26 g ha⁻¹ with 0.25% v/v nonionic surfactant (NIS), atrazine (AAtrex 4L, Syngenta) at 1120 g ha⁻¹ with 1% v/v crop oil concentrate (COC), dicamba (Clarity, BASF) at 560 g ha⁻¹ with 0.25% v/v NIS, fluroxypyr (Starane Ultra, Corteva Agriscience) at 228 g ha⁻¹, and a premixture of dichlorprop/dicamba/2,4-D (Scorch EXT, Nufarm) at 374/186/186 g ha⁻¹ with 0.25% v/v NIS. Seedlings from each population were treated when they were 6- to 9-cm tall. Herbicides were applied using a stationary cabinet spray chamber (Research Track Sprayer, De Vries Manufacturing, Hollandale, MN), equipped with an even flat-fan nozzle tip (TeeJet 8001EXR, Spraying System, Wheaton, IL) calibrated to deliver 132 L ha⁻¹ of spray solution at 241 kPa. At 28 days after treatment (DAT), the survival frequency using a simple ratio (number of alive seedlings/total number of seedlings screened) was determined for each tested population for each herbicide. To describe the resistance level, we used categories previously described by Owen et al. (2007) and Westra et

al. (2019), classifying kochia populations as either susceptible (<2% survival), having low resistance (2% to 19% survival), or resistant ($\geq$ 20% survival) to the respective use rate for each herbicide.

Dose-Response Experiments

Based on results from screening experiment, two MHR kochia populations (resistant to dicamba, fluroxypyr, and glyphosate) from Kansas (KS 19-2 and KS 19-7) and Oklahoma (OK 20 and OK 21) were selected (Table 2.1). Seeds of most putative MHR kochia populations from Texas did not germinate, therefore, TX 18-52 (resistant to dicamba and glyphosate) and TX 19-65 (resistant to glyphosate only) were selected. Susceptible (SUS) populations from Kansas (KS-SUS), Oklahoma (OK-SUS), and Texas (TX-SUS) were also selected (Table 2.1). The experiments were conducted in greenhouse conditions as mentioned above at KSU-ARC near Hays, KS in the fall of 2023 and repeated in the spring of 2024 (two experimental runs). Seeds for MHR and SUS kochia populations from each state were separately sown in 50-cell plastic trays (54- by 28- by 10-cm) containing the same potting mixture as mentioned above. Kochia seedlings (2 to 3 cm tall) from each population were transplanted in 10- by 10-cm square plastic pots (1 seedling per pot) containing the same potting mixture as mentioned above. Kochia seedlings were watered daily to maintain optimum growth. Experiments were conducted in a randomized complete block design with 12 replications (one plant per pot indicates one replication). Dicamba, fluroxypyr, and glyphosate dose response experiments were conducted for Kansas and Oklahoma populations and only dicamba and glyphosate dose-response experiments were conducted for Texas populations. Dicamba doses including 0, 140, 280, 560 (field-use rate in fallow), 1120, 2240, 4480, 8960, and 17,920 g ae ha⁻¹ were tested. The tested doses for fluroxypyr were 0, 57, 114, 228 (field-use rate in wheat), 456, 912, 1824, and 3648 g ae ha⁻¹ and

glyphosate doses were 0, 315, 630, 1260 (field-use rate), 2520, 5040, 10,080, and 20,160 g ae ha⁻¹. The following adjuvants, NIS at 0.25% v/v with dicamba and AMS at 2% w/v with glyphosate, were used. All herbicides were applied to 6- to 9-cm tall kochia for each population using a stationary cabinet spray chamber as described above. Percent visual control ratings were recorded based on each herbicide injury symptoms such as chlorosis, epinasty (curling, twisting, and cupping), and necrosis of kochia seedlings at 7, 14, and 28 DAT on a scale of 0% (no control) to 100% (complete control). At 28 DAT, each plant was clipped at the soil surface, placed in paper bags, and dried at 65 C for 4 days to obtain shoot dry biomass. Dry biomass data from each replication were converted to percent reduction of biomass compared to nontreated biomass (average of 12 replications).

Statistical Analyses

The collection sites for 82 kochia populations were mapped using QGIS (Version 3.22) to visualize spatial distribution of resistance for each herbicide and state. Data from dose-response experiments from each state for percent visual control and reduction in shoot dry biomass were subjected to ANOVA using PROC MIXED procedure in SAS 9.3 (SAS Institute, Inc., SAS Campus Drive, Cary, NC). The fixed effects were experimental runs, herbicides, herbicide doses, selected kochia populations, and their interactions. Replications and all interactions involving replication were considered as random effects. The data followed all the ANOVA assumptions as tested by PROC UNIVARIATE in SAS. The experimental run-by-herbicide interaction for each state was non-significant (P value >0.05); therefore, data were pooled across experimental runs for each state. Data on percent visual control and reduction of shoot dry biomass for each kochia population were regressed over herbicide doses using a three-parametric log-logistic model in R software (Ritz et al. 2015).

$$Y = \{d/1 + \exp[b(\log x - \log e)]\} \quad \text{Eqn. 2.1}$$

where Y is percent control or percent reduction in shoot dry biomass, d is maximum percent control or biomass reduction (fixed to 100%), b is slope, x is herbicide dose, and e represents herbicide dose needed for 50% control or shoot dry biomass reduction (referred to as LD₅₀ or GR₅₀ values, respectively). All nonlinear regression parameters and LD₉₀ or GR₉₀ values (herbicide dose required for 90% kochia control or shoot dry biomass reduction, respectively) were estimated using the DRC package in R software. The resistance index was calculated by dividing the LD₅₀ or GR₅₀ value for each MHR population by the LD₅₀ or GR₅₀ value of the respective SUS kochia population for that state.

Results and Discussion

Single-dose Experiment

Glyphosate resistance frequency in all tested kochia populations ranged from 0 to 97% with 70% of populations (57 out of 82 total) classified as resistant to glyphosate, 8% classified as low resistant, and 22% were susceptible (Figure 2.1 and Table 2.2). Similarly, the resistance frequency for atrazine ranged from 0 to 100% with 33% of populations classified as resistant, 51% had low resistance, and 16% were susceptible. The proportion of dicamba resistance was as follows: 48% classified as resistant, 28% classified as low resistant, and 24% susceptible with a survival frequency of 0 to 96%. For fluroxypyr, 30% of populations were classified as resistant, 40% were low resistant, and 30% were susceptible with 0 to 100% survival frequency. All tested kochia populations were classified as resistant to chlorsulfuron with a survival frequency ranging from 30 to 100% (Figure 2.1 and Table 2.2). In contrast, the premixture of dichlorprop/dicamba/2,4-D provided complete control of all the populations with 0% survival frequency.

Grouping the classifications of resistant and low-resistant kochia populations, 78% were resistant to glyphosate, 84% to atrazine, 76% to dicamba, 70% to fluroxypyr, and 100% to chlorsulfuron (Tables 2.2). These results indicate a widespread distribution of resistance in kochia populations to glyphosate, atrazine, dicamba, fluroxypyr, and chlorsulfuron in the SGP region. About 12% of populations were found to be three-way resistant ($\geq 20\%$ survival) to atrazine, chlorsulfuron, and glyphosate while 15% of populations were four-way resistant to atrazine, chlorsulfuron, dicamba, and glyphosate (Figure 2.2). Only two kochia populations were found five-way resistant to atrazine, chlorsulfuron, dicamba, fluroxypyr, and glyphosate (Figure 2.2). These results are consistent with Beckie et al. (2019), who reported ALS-inhibitor resistance in all the surveyed kochia populations from Alberta, Canada. They also reported that 50% of populations were resistant to glyphosate, 18% to dicamba, and 10% of tested kochia populations were multiple-resistant to ALS inhibitors, glyphosate, and dicamba. Geddes et al. (2022a) reported that 41% of tested kochia populations from Alberta, Canada were resistant to both dicamba and fluroxypyr. Geddes et al. (2022b) also reported resistance to glyphosate, fluroxypyr, and dicamba in 78%, 44%, and 28% of tested kochia populations, respectively. Westra et al. (2019) reported glyphosate and dicamba resistance in 39 to 60% and 28 to 45% of surveyed kochia populations from Colorado, respectively. They also reported multiple resistance to dicamba and glyphosate in 14 to 20% of surveyed kochia populations. Kumar et al. (2019b) reported previously 3- to 9-fold dicamba and fluroxypyr resistance in kochia populations from Kansas.

Dose-response Experiments

Kansas Populations

Based on the percent kochia control, the MHR populations (KS 19-2 and KS 19-7) were 4- to 8-fold resistant to dicamba, 2- to 4-fold resistant to fluroxypyr, and 3- to 4-fold resistant to glyphosate as compared to the SUS population (Figure 2.3 to 2.5; Table 2.3). On the basis of shoot dry biomass reduction, Kansas populations (KS 19-2 and KS 19-7) were 5- to 10-fold resistant to dicamba, 3- to 6-fold resistant to fluroxypyr, and 4- to 5-fold resistant to glyphosate as compared to the SUS population (Figure 2.3 to 2.5; Table 2.4). About 3,489 to 7,485 g ha⁻¹ of dicamba was needed to achieve 90% control of the MHR populations compared to only 514 g ha⁻¹ for the SUS population (Table 2.3). Similarly, a 5-to 7-times greater rate of fluroxypyr and 5- to 6-times greater rate of glyphosate was needed to obtain 90% control of the MHR populations compared to the SUS population. Consistent with percent control, 3,436 to 8,677 g ha⁻¹ of dicamba was needed to achieve 90% shoot dry biomass reduction of MHR populations compared to 544 g ha⁻¹ for the SUS population (Figure 2.3; Table 2.4). Similarly, 1,787 to 2,757 g ha⁻¹ of fluroxypyr and 3,635 to 4,603 g ha⁻¹ of glyphosate were needed to achieve 90% shoot dry biomass reduction of MHR populations compared to 253 g ha⁻¹ and 646 g ha⁻¹ for the SUS populations, respectively (Figures 2.4 and 2.5; Table 2.4). These results are consistent with Kumar et al. (2019b) who reported 3- to 15-fold resistance to dicamba and 3- to 9-fold resistance to fluroxypyr from Kansas. Similarly, Godar et al. (2015) also reported 4- to 11-fold resistance to glyphosate in kochia populations from Kansas. Previously, three-to four-way resistance to glyphosate, chlorsulfuron, atrazine, and/or dicamba/fluroxypyr in kochia populations from Kansas has also been reported (Kumar et al. 2019a; Varanasi et al. 2015).

Oklahoma Populations

The MHR populations (OK 20 and OK 21) were 8- to 12-fold resistant to dicamba, 3- to 4-fold resistant to fluroxypyr, and 3- to 4-fold resistant to glyphosate based on percent kochia

control as compared to the SUS population (Figures 2.3 to 2.5; Table 2.3). Consistent with percent control, the populations were 10- to 13-fold resistant to dicamba, 3- to 4-fold to fluroxypyr, and 3- to 4-fold to glyphosate based on shoot dry biomass reduction as compared to the SUS population (Figures 2.3 to 2.5; Table 2.4). About 5,628 to 5,963 g ha⁻¹ of dicamba was needed to achieve 90% control of the MHR populations compared to only 341 g ha⁻¹ for the SUS population (Table 2.3). Similarly, a 6-to 7-times higher rate of fluroxypyr and glyphosate was needed to obtain 90% control of the MHR population from Oklahoma compared to the SUS population. Consistent with percent control, 7,527 to 9,629 g ha⁻¹ of dicamba was needed to achieve 90% shoot dry biomass reduction compared to 427 g ha⁻¹ for the SUS population (Figure 2.3; Table 2.4). Similarly, 891 to 1,191 g ha⁻¹ of fluroxypyr and 4,934 to 6,045 g ha⁻¹ of glyphosate were needed to achieve 90% shoot dry biomass reduction of MHR populations compared to 179 g ha⁻¹ and 674 g ha⁻¹ for the SUS populations, respectively (Figures 2.4 and 2.5; Table 2.4). These results indicate a low level of resistance to glyphosate and fluroxypyr (upto 4-fold) and a moderate level of resistance to dicamba (upto 13-fold). To date, herbicide resistance in kochia populations from Oklahoma has been reported to ALS inhibitors (chlorsulfuron, metsulfuron-methyl, sulfometuron-methyl) and glyphosate (Heap 2024). To our knowledge, the current study revealed the first case of multiple herbicide resistance to glyphosate, dicamba, and fluroxypyr in kochia populations from Oklahoma. Fallow-based cropping systems in western Oklahoma, such as winter wheat-fallow and winter wheat-summer crop-fallow, are prevalent, and application of glyphosate and dicamba are commonly used herbicides in fallow phase and at post-harvest time to control weeds (Manuchehri et al. 2019). This may have resulted in strong selection pressure and ultimately the evolution of herbicide resistance in kochia populations. Previous studies have reported 3- to 13-fold glyphosate resistance in kochia populations

collected from Kansas, Colorado, North Dakota, and South Dakota (Godar et al. 2015; Wiersma et al. 2015). Kumar et al. (2019b) reported 3-to 15-fold resistance to dicamba and 3- to 9-fold resistance to fluroxypyr in kochia populations collected from Kansas.

Texas Populations

Based on the resistance index calculated from percent kochia control and shoot dry biomass reduction, two MHR populations (TX 18-52 and TX 19-65) were 2- to 4-fold resistant to dicamba and one population (TX 18-52) was 5-fold resistant to glyphosate compared to the SUS population (Figures 2.3 and 2.5; Tables 2.3 and 2.4). Furthermore, to achieve 90% control of MHR populations, 1,064 to 2,272 g ha⁻¹ of dicamba was required as compared to 557 g ha⁻¹ for the SUS population. Similarly, a 4-times higher rate of glyphosate was needed to obtain 90% control of the MHR population from Texas compared to the SUS population (Table 2.3). Consistent with percent control, 1,838 to 2,542 g ha⁻¹ of dicamba was needed to achieve 90% shoot dry biomass reduction compared to 604 g ha⁻¹ for the SUS population (Table 2.4). Similarly, 1,655 g ha⁻¹ of glyphosate was required to achieve 90% shoot dry biomass reduction compared to 347 g ha⁻¹ for the SUS population. These results indicate the low level of resistance to glyphosate and dicamba. Previous studies have also reported glyphosate-resistant kochia from the Great Plains, including Colorado (Wiersma et al. 2015), Kansas (Godar et al. 2015), Montana (Kumar et al. 2014), Wyoming (Gaines et al. 2016), Nebraska (Rana and Jhala 2016), North Dakota (Heap 2024), Oklahoma (Heap 2024), South Dakota (Heap 2024), as well as the Canadian provinces of Alberta, Manitoba, and Saskatchewan (Beckie et al. 2015; Heap 2024; Hall et al. 2014). LeClere et al. (2018) reported MHR kochia populations to dicamba (8-fold) and fluroxypyr (13-fold) from western Nebraska. Westra et al. (2019) reported multiple resistance to dicamba and glyphosate in 14 to 20% of surveyed kochia populations from Colorado.

Glyphosate resistance has been reported from ten US states including Colorado, Idaho, Kansas, Montana, Nebraska, North Dakota, Oklahoma, Oregon, South Dakota, and Wyoming whereas dicamba resistance has been reported from six US states, including Colorado, Idaho, Kansas, Montana, Nebraska, and North Dakota (Heap 2024). To date, herbicide resistance in kochia populations from Texas has been reported to only metsulfuron-methyl (Heap 2024). The present study revealed resistance to glyphosate and dicamba in kochia populations collected from Texas.

Overall, the results from this study confirmed multiple herbicide resistance to dicamba, fluroxypyr, and glyphosate from SGP. Therefore, over-reliance on these herbicides for kochia control should be avoided in the region to maintain their utility for susceptible kochia control. The underlying mechanism(s) of resistance in these MHR kochia populations is unknown. Several previous studies showed that higher *EPSPS* gene copy conferred glyphosate resistance in kochia populations (Gaines et al. 2016; Godar et al. 2015; Kumar et al. 2019b; Wiersma et al. 2015). Pettinga et al. (2017) reported that a greater synthesis of flavonols would compete with the intercellular transport of dicamba molecules, thus impairing the dicamba translocation and resulted in dicamba resistance in kochia. LeClere et al. (2018) reported a point mutation within a highly conserved region of an AUX/IAA protein which conferred cross-resistance to dicamba, 2,4-D, and fluroxypyr in kochia. With the widespread evolution of MHR kochia populations in the SGP and limited herbicide options for kochia control warrant integrated weed management practices like diversified crop rotations, occasional tillage, cover crops, herbicide rotations, and multiple herbicide SOA to delay the further evolution of herbicide resistance in kochia. Future studies should investigate the underlying mechanism(s) of resistance in these MHR kochia populations from the SGP region.

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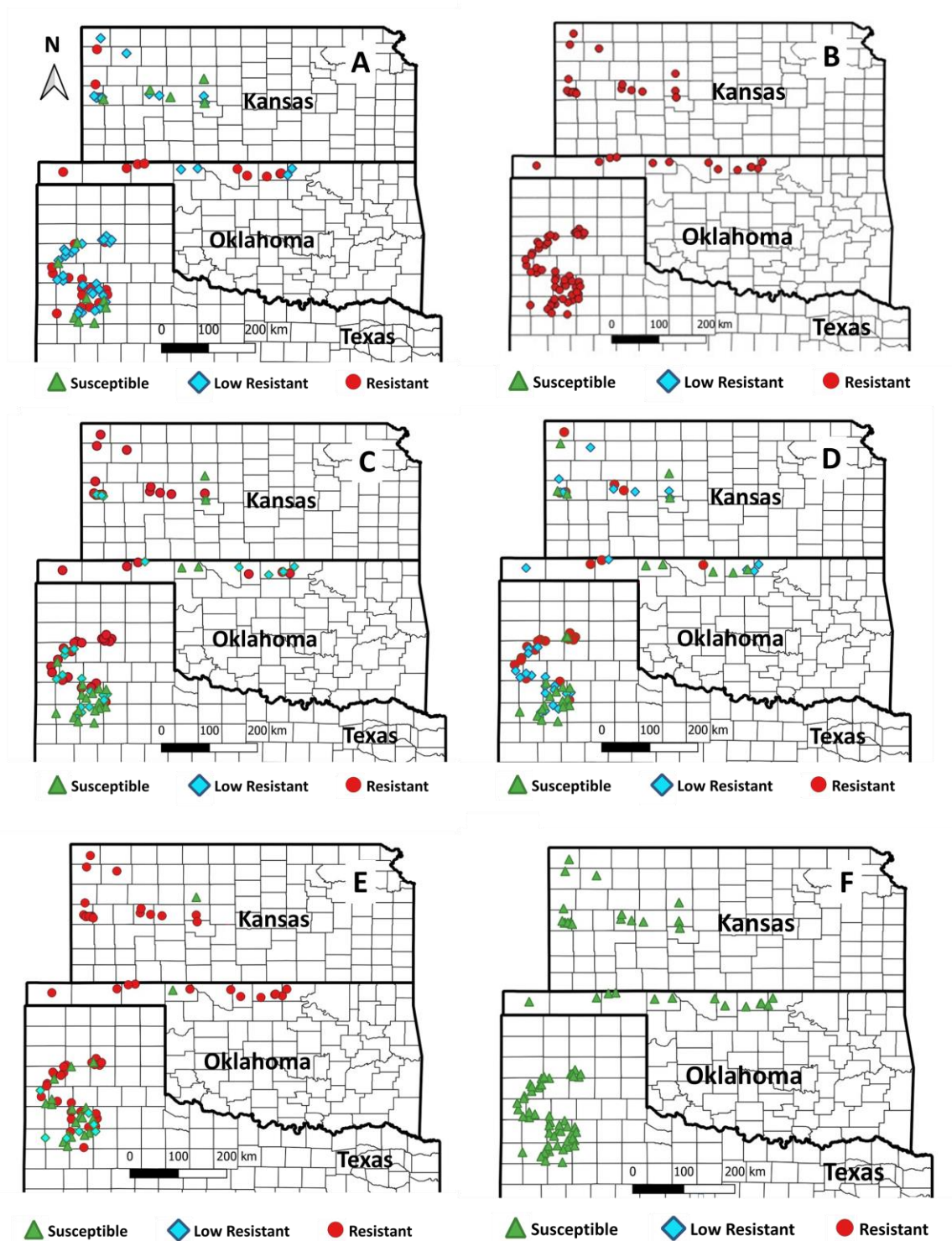


Figure 2.1. Distribution of kochia populations resistant to atrazine (A), chlorsulfuron (B), dicamba (C), fluroxypyr (D), glyphosate (E), and dichlorprop/dicamba/2,4-D (F) in Kansas, Oklahoma, and Texas.

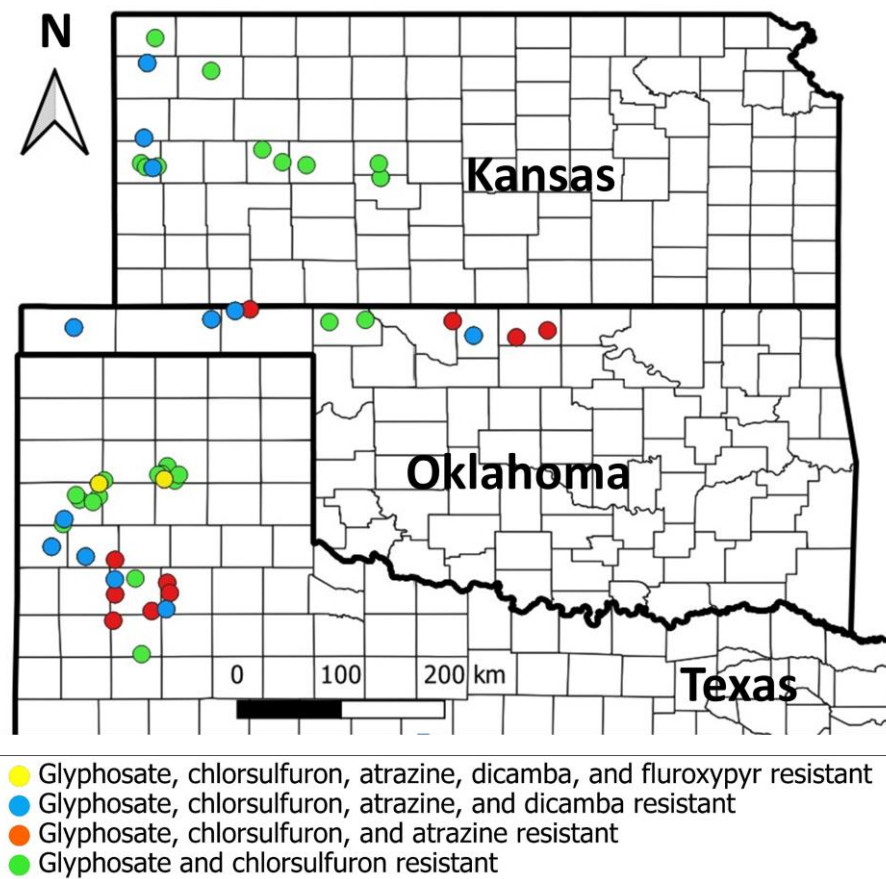


Figure 2.2. Distribution of putative multiple herbicide-resistant kochia populations in Kansas, Oklahoma, and Texas.

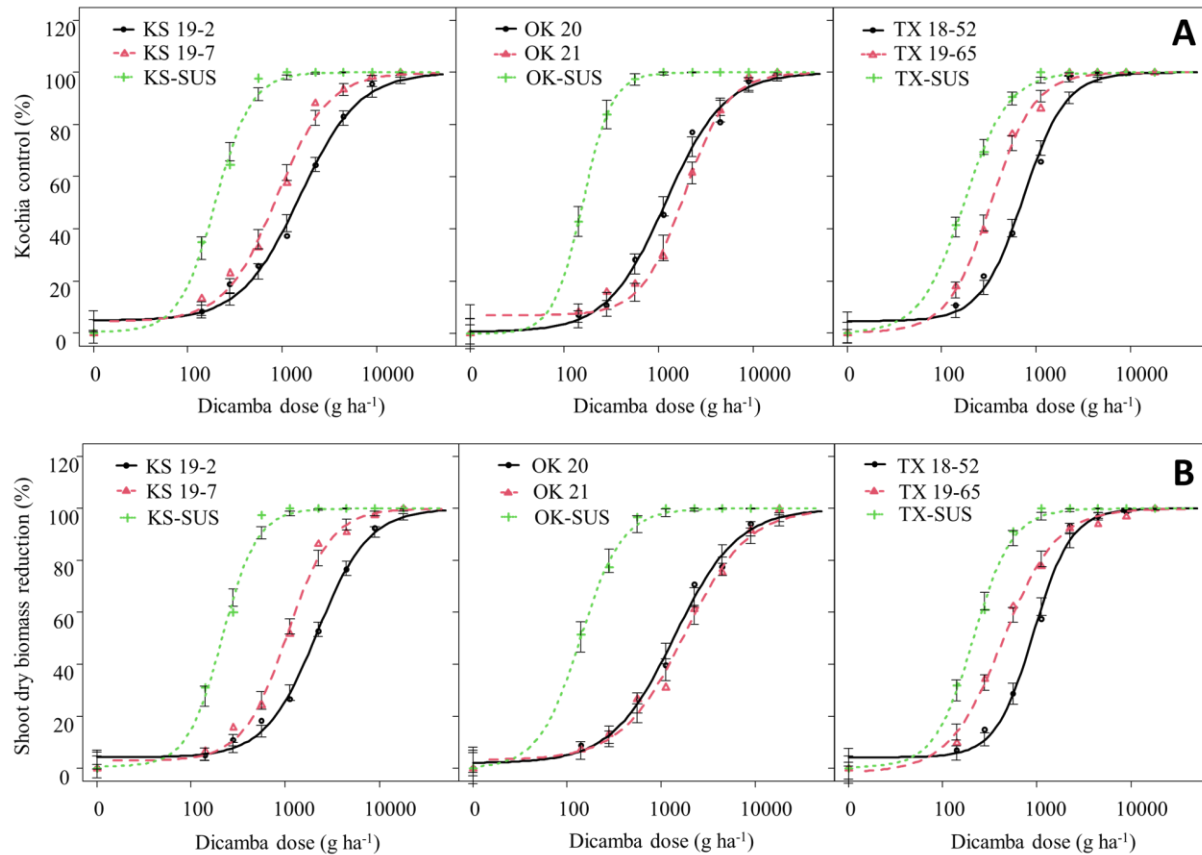


Figure 2.3. Percent control (A) and shoot dry biomass reduction (B) response of kochia populations to various doses of dicamba. KS 19-2 and KS 19-7 were multiple herbicide-resistant (MHR) populations from Kansas; OK 20 and OK 21 were MHR populations from Oklahoma; TX 18-52 and TX 19-65 were MHR populations from Texas. KS-SUS, OK-SUS, and TX-SUS were susceptible kochia populations collected from Kansas, Oklahoma, and Texas, respectively.

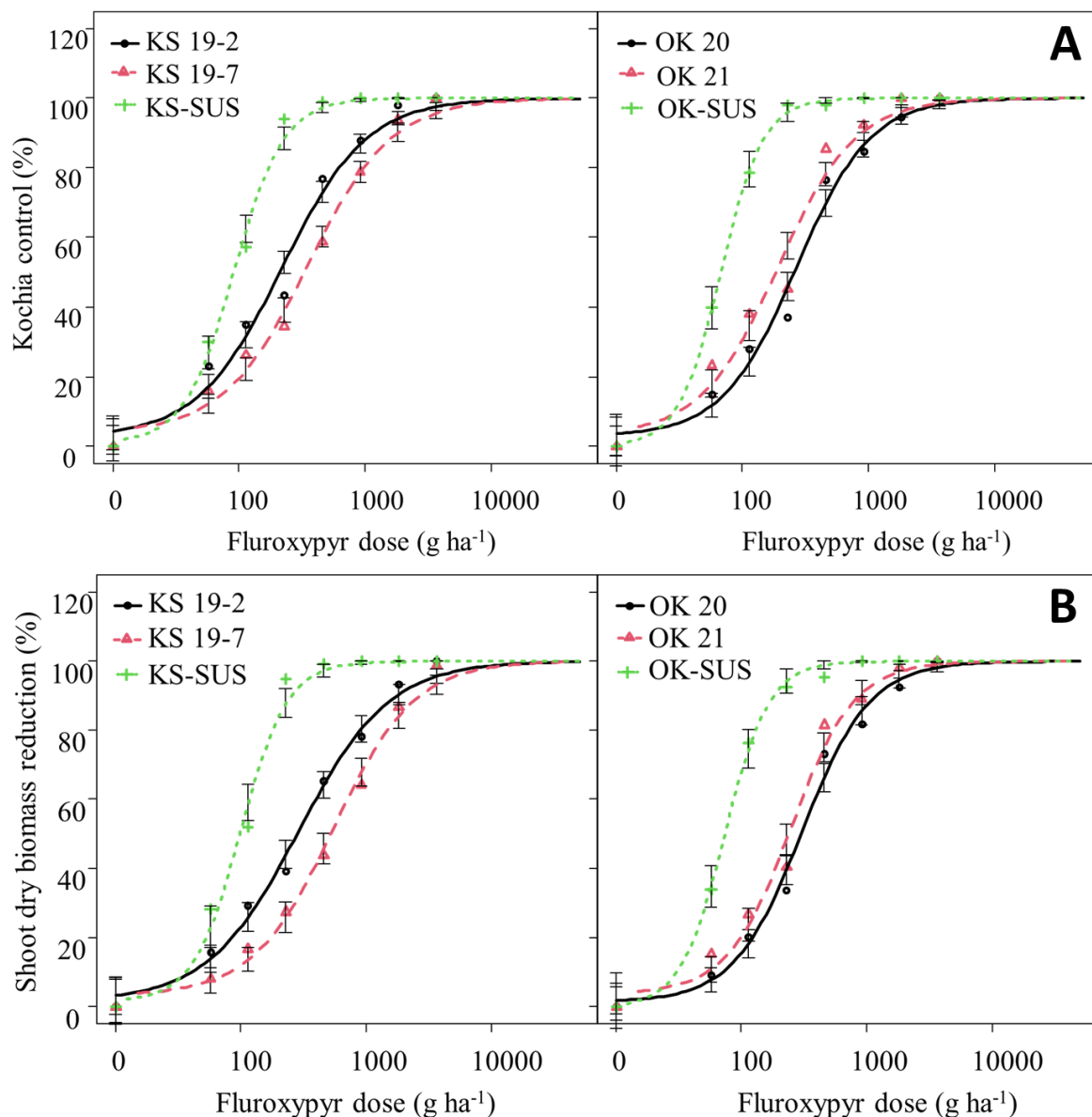


Figure 2.4. Percent control (A) and shoot dry biomass reduction (B) response of kochia populations to various doses of fluroxypyr. KS 19-2 and KS 19-7 were multiple herbicide-resistant (MHR) populations from Kansas; OK 20 and OK 21 were MHR populations from Oklahoma. KS-SUS and OK-SUS were and susceptible kochia populations collected from Kansas and Oklahoma, respectively.

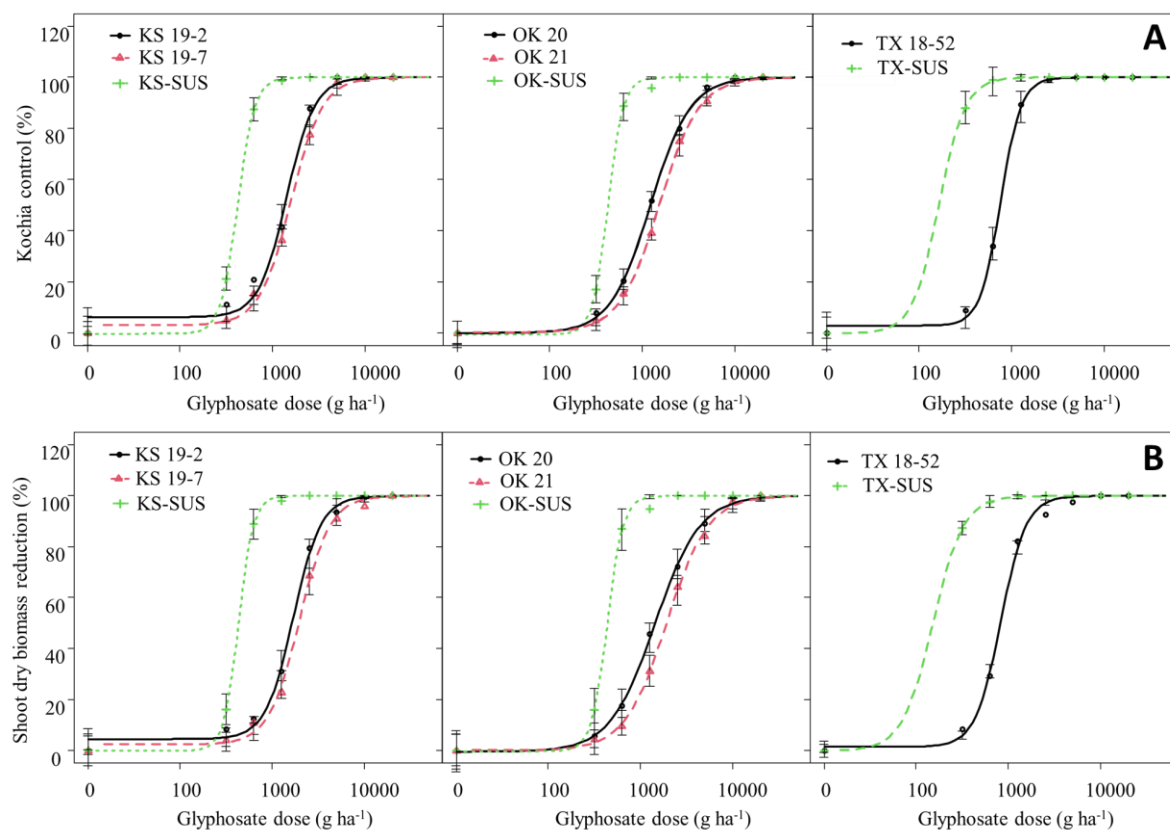


Figure 2.5. Percent control (A) and shoot dry biomass reduction (B) response of kochia populations to various doses of glyphosate. KS 19-2 and KS 19-7 were multiple herbicide-resistant (MHR) populations from Kansas; OK 20 and OK 21 were MHR populations from Oklahoma; TX 18-52 was MHR population from Texas. KS-SUS, OK-SUS, and TX-SUS were susceptible kochia populations collected from Kansas, Oklahoma, and Texas, respectively.

Table 2.1. Selected putative multiple herbicide-resistant (MHR) and susceptible (SUS) kochia populations for dose response experiment.

Population	Longitude	Latitude	Comments
KS 19-2	-101.6250	39.7570	Wheat Stubble
KS 19-7	-101.7200	38.4300	Wheat Stubble
KS-SUS	-101.5511	38.3900	Sorghum Stubble
OK-20	-100.8032	36.9518	Wheat Stubble
OK-21	-101.0475	36.8646	Wheat Stubble
OK-SUS	-99.8351	36.8365	Wheat Stubble
TX 18-52	-101.5936	35.2661	Cotton Field
TX 19-65	-102.4100	34.9100	Sorghum Field Edge
TX-SUS	-101.5567	35.2739	Sorghum Field Edge

Table 2.2. The number and (percentage) of kochia populations determined to be susceptible, low resistant, and resistant categories for each tested herbicide.

Herbicide	Susceptible	Low resistant	Resistant
Atrazine	13 (16)	42 (51)	27 (33)
Chlorsulfuron	0 (0)	0 (0)	82 (100)
Dicamba	20 (24)	23 (28)	39 (48)
Dichlorprop/dicamba/2,4-D	82 (100)	0 (0)	0 (0)
Fluroxypyr	25 (30)	32 (40)	25 (30)
Glyphosate	18 (22)	7 (8)	57 (70)

Resistance level classified as three categories based on survival %; <2% = susceptible, 2% to 19% = low resistant, and 20% to 100% = resistant

Table 2.3. Regression parameter estimates (Equation 2.1) for percent control of multiple herbicide-resistant and susceptible kochia populations from Kansas, Oklahoma, and Texas to dicamba, fluroxypyr, and glyphosate under separate dose-response experiments, and calculated resistance index.

Herbicide	Population	Parameter estimates			Resistance index
		b ($\pm$ SE)	LD ₅₀ (95% CI)	LD ₉₀ (95% CI)	
Dicamba	KS 19-2	-1.3 (0.1)	1533 (1347-1719)	7485 (6103-8868)	7.9
	KS 19-7	-1.5 (0.1)	874 (773-975)	3489 (2886-4093)	4.5
	KS SUS	-2.2 (0.2)	195 (178-211)	514 (446-582)	
	OK 20	-1.4 (0.1)	1176 (1010-1341)	5628 (4324-6931)	7.6
	OK 21	-1.8 (0.1)	1867 (1647-2087)	5963 (4680-7245)	12.0
	OK-SUS	-2.7 (0.3)	155 (141-168)	341 (277-404)	
	TX 18-52	-1.9 (1.1)	733 (668-797)	2272 (1985-2560)	4.3
	TX 19-65	-1.8 (0.1)	332 (305-359)	1064 (919-1209)	1.9
	TX-SUS	-1.8 (0.1)	172 (157-185)	557 (474-640)	
Fluroxypyr	KS 19-2	-1.3 (0.1)	218 (186-250)	1183 (942-1423)	2.4
	KS 19-7	-1.3 (0.1)	348 (297-399)	1873 (1480-2266)	3.8
	KS-SUS	-2.2 (0.1)	91 (82-100)	247 (208-285)	
	OK 20	-1.4 (0.1)	267 (225-307)	1168 (898-1438)	4.0
	OK 21	-1.4 (0.1)	191 (160-222)	900 (704-1096)	2.9
	OK SUS	-2.5 (0.2)	67 (60-73)	157 (129-186)	
Glyphosate	KS 19-2	-2.8 (0.3)	1414 (1306-4523)	3052 (2618-3486)	3.4
	KS 19-7	-2.5 (0.2)	1573 (1453-1693)	3737 (3244-4231)	3.8
	KS-SUS	-4.6 (0.3)	416 (391-441)	665 (600-730)	
	OK 20	-2.0 (0.1)	1224 (1099-1349)	3659 (3045-4274)	2.9
	OK 21	-2.0 (0.1)	1527 (1375-1678)	4527 (3747-5306)	3.6
	OK-SUS	-5.1 (0.5)	424 (394-454)	648 (579-718)	
	TX 18-52	-3.9 (0.5)	756 (697-814)	1324 (1106-1541)	4.6
	TX SUS	-3.0 (0.5)	163 (110-215)	334 (264-404)	

Abbreviations: KS-SUS, OK-SUS, and TX-SUS were susceptible kochia populations collected from Kansas, Oklahoma, and Texas, respectively; KS 19-2 and KS 19-7 were multiple herbicide-resistant populations from Kansas; OK 20 and OK 21 were multiple herbicide-resistant populations from Oklahoma; TX 18-52 and TX 19-65 were multiple herbicide-resistant populations from Texas; SE, standard error of mean; CI, confidence interval; LD₅₀, effective dose (g ha⁻¹) required for 50% kochia control; Resistance index is the ratio of LD₅₀ of a multiple herbicide-resistant population to LD₅₀ of the susceptible kochia population from each respective state.

Table 2.4. Regression parameter estimates (Equation 2.1) for shoot dry biomass reduction of multiple herbicide-resistant and susceptible kochia populations from Kansas, Oklahoma, and Texas to dicamba, fluroxypyr, and glyphosate under separate dose-response experiments and calculated resistance index.

Herbicide	Population	Parameter estimates			Resistance index
		b ($\pm$ SE)	GR ₅₀ (95% CI)	GR ₉₀ (95% CI)	
Dicamba	KS 19-2	-1.5 (0.1)	2170 (1972-2368)	8677 (7331-10023)	10.2
	KS 19-7	-1.8 (0.1)	1046 (958-1135)	3436 (2916-3955)	4.9
	KS SUS	-2.3 (0.2)	213 (196-229)	544 (482-606)	
	OK 20	-1.2 (0.1)	1387 (1167-1608)	7527 (5653-9400)	10.1
	OK 21	-12 (0.1)	1762 (1473-2051)	9629 (7216-12043)	12.8
	OK-SUS	-1.9 (0.2)	138 (119-157)	427 (327-525)	
	TX 18-52	-2.1 (0.1)	921 (841-1000)	2542 (2186-2899)	4.3
	TX 19-65	-1.5 (0.1)	432 (390-475)	1838 (1523-2115))	2.0
	TX-SUS	-2.0 (0.1)	212 (194-229)	604 (519-690)	
Fluroxypyr	KS 19-2	-1.2 (0.1)	287 (232-343)	1787 (1296-2277)	2.9
	KS 19-7	-1.2 (0.2)	545 (449-641)	2757 (2036-3477)	5.6
	KS-SUS	-2.3 (0.2)	98 (86-111)	253 (207-299)	
	OK 20	-1.6 (0.1)	303 (268-345)	1191 (910-1472)	4.2
	OK 21	-1.7 (0.1)	248 (212-285)	891 (689-1093)	3.4
	OK SUS	-2.4 (0.2)	73 (66-80)	179 (142-162)	
Glyphosate	KS 19-2	-2.8 (0.2)	1685 (1538-1833)	3635 (2948-4323)	3.9
	KS 19-7	-2.6 (0.2)	1969 (1791-2147)	4603 (3696-5509)	4.6
	KS-SUS	-5.3 (0.5)	427 (393-461)	646 (567-725)	
	OK 20	-1.7 (0.1)	1431 (1196-1667)	4934 (3902-5966)	3.3
	OK 21	-1.9 (0.2)	1913 (1622-2204)	6045 (4803-7287)	4.4
	OK-SUS	-5.0 (0.3)	435 (388-482)	674 (594-754)	
	TX 18-52	-3.1 (0.1)	822 (793-852)	1655 (1363-1947)	5.4
	TX SUS	-2.6 (0.1)	151 (105-198)	347 (264-430)	

Abbreviations: KS-SUS, OK-SUS, and TX-SUS were susceptible kochia populations collected from Kansas, Oklahoma, and Texas, respectively; KS 19-2 and KS 19-7 were multiple herbicide-resistant populations from Kansas; OK 20 and OK 21 were multiple herbicide-resistant populations from Oklahoma; TX 18-52 and TX 19-65 were multiple herbicide-resistant populations from Texas; SE, standard error of mean; CI, confidence interval; GR₅₀, effective dose (g ha⁻¹) required for 50% shoot dry biomass reduction; Resistance index is the ratio of GR₅₀ of a multiple herbicide-resistant population to GR₅₀ of the susceptible kochia population from each respective state.

Chapter 3 - Synergistic interactions of 2,4-D, dichlorprop-p, dicamba, and halauxifen/fluroxypyr for controlling multiple herbicide-resistant kochia

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 days 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.

Keywords: *Bassia scoparia* (L.) A. J. Scott; multiple herbicide resistance; herbicides interaction; U.S. Great Plains

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-enolpyruvylshikimate-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 $\geq 90\%$ 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 3.1.

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 $25/23 \pm 3$ C day/night temperatures and 16/8 h (day/night) photoperiod supplemented with metal-halide lamps ($560 \mu\text{mol m}^{-2} \text{s}^{-1}$). 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 3.1) 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 3.1:

$$\text{Shoot dry weight reduction (\%)} = \left[\frac{c-T}{c} \right] \times 100 \quad [\text{Eqn. 3.1}]$$

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 Experiments

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 the Hays site (38°51'08.7"N, 99°20'16.3"W) was Roxbury silt loam, pH 7.6, with 2.1% organic matter. The soil type at the Garden City site (38°00'10.00"N, 100°48'54.80"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 3.1 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 3.2. 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 3.1) were applied on young actively growing kochia seedlings (8 to 10 cm tall) using a CO₂-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-m² 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 3.1.

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 3.2) was used to calculate the expected values for two-way mixtures to determine the interaction of tested auxinic herbicides (Colby 1967):

$$E = (X + Y) - \frac{(XY)}{100} \quad [\text{Eqn. 3.2}]$$

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 and B, respectively.

Similarly, the expected values for three-way mixtures were calculated using Equation 3.3 (Colby 1967):

$$E = (X + Y + Z) - \frac{(XY + XZ + YZ)}{100} + \frac{XYZ}{100,000} \quad [\text{Eqn. 3.3}]$$

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 ($96\% \pm 2.5\%$) than the 10A population ($88\% \pm 2.5\%$; Table 3.3). 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 3.3). 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 3.3). 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

3.3). 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 3.3).

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 $\geq 85\%$ shoot dry weight reduction of MHR kochia populations (Table 3.3). 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 3.3). 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 3.4).

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 3.2). 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 3.5). 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 3.5). A recent study also reported $\geq 90\%$ 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 $\geq 84\%$ control of MHR kochia at 28 DAT (Table 3.6). 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 3.6). 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 3.5 and 3.6). 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 3.7).

Shoot Dry Weight 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 3.5). Furthermore, mixing dicamba with dichlorprop-p + 2,4-D, halauxifen/fluroxypyr, and halauxifen/fluroxypyr + dichlorprop-p reduced shoot dry weights of MHR kochia by $\geq 90\%$. 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 3.7). 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 halauxifen/fluroxypyr (Table 3.7). 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 3.6). All the two-way or three-way mixtures resulted in $\geq 80\%$ 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 3.7). 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.

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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Table 3.1. 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 g ai or ae ha ⁻¹	Manufacturer
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

^aTreatments containing dichlorprop-p had nonionic surfactant (NIS) at 0.5% v/v.

Table 3.2. 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

Table 3.3. 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 +											bc							
halauxifen/fluroxypyr	80	b	70*	76	bc	46*	78	c	87*	70	d	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 +											ab							
dichlorprop-p	72	c	78	53	de	49	80	c	86	76	c	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*

^aMeans followed by the same letter within a column are not different according to Fisher's protected LSD at $P < 0.05$.

^bAsterisks (*) 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 3.4. 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.

Contrast	10A				4H				SUS			
	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

Table 3.5. 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}

Treatments	Rate	Control						Shoot dry weight reduction		
		14 DAT			28 DAT					
		Observed	Expected	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

^aAbbreviation: KSU-ARC, Kansas State University Agricultural Research Center.

^bMeans followed by the same letter within a column are not different according to Fisher's protected LSD at $P < 0.05$.

^cAsterisks (*) 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 3.6. 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}

Treatments	Rate g ai or ae ha ⁻¹	Control						Shoot dry weight reduction		
		14 DAT			28 DAT					
		Observed	Expected	Observed	Expected	Observed	Expected	Observed	Expected	
						%				
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

^aAbbreviation: KSU-SWREC, Kansas State University Southwest Research and Extension Center.

^bMeans followed by the same letter within a column are not different according to Fisher's protected LSD at $P < 0.05$.

^cAsterisks (*) 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 3.7. 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

Contrast	Hays				Garden City			
	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.

Chapter 4 - Control of multiple herbicide-resistant kochia with pyridate-based herbicide mixtures

Abstract

Multiple herbicide-resistant (MHR) kochia is an increasing concern in the Central Great Plains, including western Kansas. Greenhouse and field experiments were conducted at Kansas State University Research and Extension Centers near Hays and Garden City, KS to evaluate several pyridate-based tank mixtures for controlling MHR kochia. One previously confirmed MHR (resistant to atrazine, glyphosate, dicamba, and fluroxypyr) and a susceptible (SUS) kochia population were tested in a greenhouse study. Kochia population at Hays field site was resistant to atrazine, dicamba, and glyphosate while kochia at Garden City field site was resistant to atrazine and glyphosate. Colby analysis revealed synergistic interactions when pyridate was tank-mixed with atrazine, dicamba, dichlorprop-p, fluroxypyr, glyphosate, or halauxifen/fluroxypyr for MHR kochia control (>95%) and shoot dry biomass reduction (94 to 100%) in a greenhouse study. Similarly, synergistic interactions were observed in fallow field studies when pyridate was tank-mixed with glyphosate or atrazine across both sites. Kochia control was increased from 26 to 90% with the application of glyphosate + pyridate and 28 to 95% with atrazine + pyridate as compared to separate applications of glyphosate or atrazine across both sites. All other pyridate-based mixtures showed an additive interaction and resulted in better control (87 to 100%) as compared to their individual applications (23 to 92%) except 2,4-D across both sites. These results suggest that pyridate can have a crucial role in various herbicide tank mixtures for effective control of MHR kochia.

Keywords: *Bassia scoparia* (L.) A. J. Scott; dicamba; glyphosate; pyridate; U.S. Great Plains

Introduction

Kochia [*Bassia scoparia* (L.) A. J. Scott] is a C₄ summer annual broadleaf weed in the Chenopodiaceae family and is a major challenge to crop production in the North American Great Plains (Kumar et al. 2019a). Kochia has been reported as the second most troublesome weed in alfalfa (*Medicago sativa* L.), canola (*Brassica napus* L.), and spring cereals (Van Wychen 2022). Kochia is an early emerging summer annual weed, appearing early in the spring with an extended period of emergence (from mid-February through mid-June) (Dille et al. 2017; Friesen et al. 2009; Kumar et al. 2018). Kochia is tolerant to abiotic stresses (heat, drought, and salinity), a prolific seed producer (one plant can produce >100,000 seeds), and exhibits wind-mediated tumble mechanism of seed dispersal (Christoffoleti et al. 1997; Friesen et al. 2009; Kumar et al. 2019a). Season-long interference of kochia can reduce grain yield by 68% in corn (*Zea mays* L.), 62% in sorghum [*Sorghum bicolor* (L.) Moench ssp. *bicolor*], 46% in sugarbeet (*Beta vulgaris* L.), and 23% in sunflower (*Helianthus annuus* L.) (Geddes and Sharpe 2022). Similarly, kochia at densities of 240 to 520 plants m⁻² has been reported to reduce spring wheat (*Triticum aestivum* L.) grain yield by 60% (Friesen et al 1991). Kochia plants escaping post-emergence (POST) herbicide applications in the spring or late-emerging cohorts can also hinder mechanical wheat harvest (Kumar and Jha 2015a; Torbiak et al. 2021). In addition to various crop situations, kochia infestation is also highly problematic during fallow periods of the crop rotations (winter wheat-fallow or winter wheat-summer crop-fallow) (Kumar et al. 2019a).

Kochia populations resistant to five different herbicide site of actions (SOA) (WSSA Groups 2, 4, 5, 9, and 14) have been documented (Heap 2024; Kumar et al. 2019a; Sharpe et al. 2023; Westra et al. 2019). The widespread evolution of glyphosate- and acetolactate synthase (ALS) inhibitors-resistant kochia populations have been reported across the Great Plains region

(Godar et al. 2015; Kumar et al. 2019a). With the frequent use of dicamba and fluroxypyr, resistance to these herbicides has also become more prevalent (Heap 2024; Kumar et al. 2019b; Westra et al. 2019). Furthermore, few kochia populations with resistance to four herbicide SOAs, including inhibitors of photosystem (PS) II (Group 5), ALS (Group 2), 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS) (Group 9), and synthetic auxins (Group 4) have also been reported from Kansas (Heap 2024). This rapid evolution of multiple herbicide-resistant (MHR) kochia populations poses a serious management challenge by limiting the effectiveness of commonly used POST herbicides.

Pyridate is a contact, POST herbicide that belongs to PS II inhibitors (WSSA Group 6). It controls both grass and broadleaf weeds by inhibiting electron transfer due to blocking amino acid histidine 215 at the A site of PS II (Székács 2021). Tonks and Westra (1997) have previously reported 16 to 60% control of sulfonylurea-resistant kochia with pyridate alone. Wicks et al. (1994) reported 89% control of atrazine-resistant kochia with pyridate alone at 21 days after treatment (DAT). The application of two or more herbicide SOA as a tank-mixture is generally recommended to mitigate/delay evolution of herbicide resistance in weeds as a component of integrated weed management (Beckie and Reboud 2009; Green 1991). For example, Wicks et al. (1993) reported >95% control of atrazine-resistant kochia with pyridate + flurochloridone at 21 to 30 DAT. However, limited research is available regarding effectiveness of pyridate-based tank mixtures to control MHR kochia. Therefore, the objective of this study was to evaluate pyridate-based herbicide tank mixtures for controlling MHR kochia in greenhouse and field experiments in western KS.

Materials and Methods

Greenhouse Study

Greenhouse experiments were conducted at Kansas State University Agricultural Research Center (KSU-ARC) near Hays, KS in the spring of 2022 and repeated in the fall of 2022. A previously confirmed MHR kochia population (KS-10A) and a susceptible population (SUS) were used. The MHR kochia population was originally collected from Garden City, KS, and was confirmed resistant to atrazine, glyphosate, dicamba, and fluroxypyr (Kumar et al. 2019b; 2020) and the SUS population was collected from a pasture field at KSU-ARC. Seeds for both MHR and SUS kochia populations were separately sown in plastic trays (54- by 28- by 10-cm) containing commercial potting mixture (Miracle-Gro® Moisture Control® Potting Mix, Miracle-Gro Lawn Products, Scottslawn Road, Marysville, OH). The temperature in the greenhouse was maintained at $25/23 \pm 3$ C day/night and a 16/8 h (day/night) photoperiod supplemented with metal-halide lamps ($560 \mu\text{mol m}^{-2} \text{s}^{-1}$). When kochia seedlings reached 2 to 3 cm tall from each population, they were transplanted into 10- by 10-cm squared plastic pots (1 seedling per pot) containing the same potting mixture as mentioned above. Experiments were conducted in a randomized complete block design with 12 replications (one plant per pot = one replication). Kochia seedlings from both populations were watered once daily to avoid moisture stress. Actively growing young kochia seedlings (6 to 9 cm tall) from each population were treated with selected POST treatments (Table 4.1) using a stationary cabinet spray chamber (Research Track Sprayer, De Vries Manufacturing, Hollandale, MN), equipped with an even flat-fan nozzle tip (TeeJet 8001EXR, Spraying System, Wheaton, IL) calibrated to deliver 132 L ha^{-1} of spray solution at 241 kPa. Percent visual control ratings were recorded at 7, 21, and 28 DAT. Ratings were based on herbicide injury symptoms such as chlorosis, epinasty (curling, twisting,

and cupping), and necrosis of kochia seedlings on a scale of 0% (no control) to 100% (complete control). At 28 DAT, each plant was clipped at the soil surface, placed in a paper bag and dried at 65 C for 4 days to obtain shoot dry biomass. The data for shoot dry biomass was used to calculate the percent reduction of shoot dry biomass using equation 4.1:

$$\text{Shoot dry biomass reduction (\%)} = \left[\frac{BC - BT}{BC} \right] \times 100 \quad \text{Eqn 4.1}$$

where *BC* is the average shoot dry biomass from the nontreated check treatment and *BT* is the shoot dry biomass from a treated pot. Price estimates for herbicides were obtained from the average prices listed in the K-State Chemical Weed Control Guide for 2022 and 2023 (Lancaster et al. 2022, 2023)

Field Experiments

Field experiments were conducted in fallow fields at KSU-ARC near Hays, KS and at Kansas State University Southwest Research and Extension Center (KSU-SWREC) near Garden City, KS in 2023 (sorghum stubble at Hays and wheat stubble in Garden City). Soil type at Hays site was Roxbury silt loam with a pH of 7.4 and 2.1% organic matter while the soil type at Garden City was Ulysses silt loam with a pH of 8.0 and 1.4% organic matter. The field site at Hays had a history of kochia population surviving field-use rate of atrazine (1120 g ha⁻¹), dicamba (560 g ha⁻¹), and glyphosate (1260 g ha⁻¹) (personal observations). Similarly, kochia population at Garden City site had previously survived POST applications of atrazine and glyphosate. At both sites, treatments were laid out in a randomized complete block design with four replications. Plot size was 3 m wide by 6 m long at both sites. Data for daily maximum and minimum air temperature and precipitation at both field sites during the study period were obtained from nearby Kansas Mesonet (<https://mesonet.k-state.edu>) weather stations and are presented in Figures 1 and 2. All treatments (Table 4.1) were applied at their recommended field-

use rates on young actively growing kochia seedlings (5- to 9-cm tall) using a CO₂ pressurized backpack sprayer equipped with four flat-fan nozzles (TeeJet 8001XR, Spraying Systems Co., P.O. Box 7900, Wheaton, IL), calibrated to deliver 132 L ha⁻¹ of spray solution at 276 kPa. Treatments were applied on May 23 at Hays and on May 29 at Garden City. Data for percent visual control of kochia was recorded at 7, 14, and 28 DAT on a scale of 0 to 100%, where 0% indicates no control and 100% indicates complete control as compared to nontreated check. Kochia density was recorded at 28 DAT using a 1-m² quadrat placed at the center of each plot and the kochia plants were manually clipped at the soil level and shoot dry biomass was determined after oven-drying the samples at 65 C for 4 days. Similar to the greenhouse study, kochia density and aboveground shoot dry biomass data were converted to a percent reduction compared to nontreated.

Statistical Analyses

All the data collected from greenhouse and field studies were subjected to ANOVA using PROC MIXED procedure in SAS 9.3 (SAS Institute, Inc., SAS Campus Drive, Cary, NC). The data from nontreated plots were not included in the analyses. Data were checked for the ANOVA assumptions using the PROC UNIVARIATE procedure in SAS. Data on percent visual control, percent reduction in density, and percent reduction in shoot dry biomass 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. Treatments, experimental runs, kochia populations, and their interactions were considered fixed effects for greenhouse experiments, whereas sites, treatments, and their interactions were considered fixed effects for field experiments. The replication and all interactions involving replication were considered as random effects for both greenhouse and

field experiments. Data from both runs from greenhouse experiments were pooled because of non-significant experimental run-by-herbicide treatment interaction ($P = 0.36$). Data from Hays and Garden City sites were analyzed separately because of significant treatment-by-site interaction ($P < 0.0001$). Treatment means were separated using Fisher's Protected LSD test ($P < 0.05$).

Expected values with herbicide mixtures for percent kochia control and percent reduction in shoot dry biomass for greenhouse study and for percent kochia control, percent reduction in density, and percent reduction in shoot dry biomass for field studies were calculated using Colby's analysis with equation (Eqn. 4.2) to determine the interaction of tested herbicides (Colby 1967):

$$E = (X + Y) - \frac{(XY)}{100} \quad \text{Eqn. 4.2}$$

where E is the expected values of the response variable to the application of herbicides A + B in a mixture, and X and Y are the respective observed values of the response variables to the individual application of herbicide A and B, respectively. The expected and observed percent kochia control, percent reduction in density, and percent reduction in shoot dry biomass were compared using t -tests to determine if 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 herbicide mixture was considered as additive when expected and observed means were not different according to t -test $P < 0.05$ (Colby 1967).

Results and Discussion

Greenhouse Study

Percent Visual Control

Both MHR and SUS kochia populations showed differential responses with a significant interaction between populations and herbicide treatments ($P < 0.0001$). All pyridate-based herbicide mixtures, except 2,4-D + pyridate, resulted in >90% control of MHR kochia at both 14 and 28 DAT (Table 4.2). All other herbicides (without pyridate) resulted in less than 50% control of MHR kochia population at 14 and 28 DAT, except for bromoxynil/pyrasulfotole. Application of atrazine, halauxifen/fluroxypyr, glyphosate, fluroxypyr, or dicamba provided 11 to 49% control of MHR kochia population at 28 DAT, however, the control increased to 98 to 100% after adding pyridate with these herbicides. The cost of adding pyridate in the mixtures was \$27 ha⁻¹ (Table 4.1). Colby's analysis further revealed synergistic interactions for MHR kochia control when pyridate was tank-mixed with atrazine, halauxifen/fluroxypyr, glyphosate, fluroxypyr, or dicamba (Table 4.2). These results indicate the benefit of adding pyridate with these POST herbicides to control MHR kochia. Application of bromoxynil/pyrasulfotole alone or with pyridate resulted in 92 to 100% control of MHR kochia population. Similarly, all pyridate-based herbicide mixtures (except 2,4-D + pyridate) provided >95% control of SUS kochia population at 14 and 28 DAT (Table 4.2). Bromoxynil/pyrasulfotole and atrazine resulted in 97 to 100% control of SUS kochia population at 28 DAT followed by dicamba (91%), fluroxypyr (88%), glyphosate (88%), and halauxifen/fluroxypyr (85%). Previous studies have also reported effective kochia control (85 to 100%) with bromoxynil + pyrasulfotole (Kumar and Jha 2015b; Sbatella et al. 2019). Application of pyridate alone provided 52 to 72% control of both MHR and SUS populations at 28 DAT. Tonks and Westra (1997) have also previously reported 16 to 60% control of sulfonylurea-resistant kochia with pyridate alone. Kousta et al. (2024) reported 53% control of common lambsquarters (*Chenopodium album* L) with pyridate alone. Wicks et al. (1993) reported >95 control of atrazine-resistant kochia with pyridate + flurochloridone at 21 to

30 DAT. The application of 2,4-D provided only 12 to 30% control for both MHR and SUS populations at 28 DAT. Several previous studies have also reported poor kochia control with 2,4-D alone (Friesen et al. 1993; Nandula and Manthey 2002; Tonks and Westra 1997).

Percent Reduction in Shoot Dry Biomass

Consistent with percent visual control, all pyridate-based herbicide mixtures provided 94 to 100% reduction in shoot dry biomass of MHR kochia, except 2,4-D + pyridate (Table 4.2). Application of 2,4-D and glyphosate provided the least shoot dry biomass reduction (14 to 17%) of MHR kochia. Percent reduction in shoot dry biomass changed from 17 to 99% with glyphosate + pyridate, 33 to 99% with fluroxypyr + pyridate, and 45 to 99% with dicamba + pyridate compared to glyphosate, fluroxypyr, or dicamba alone, respectively. Greater observed percent reduction of shoot dry biomass for MHR kochia as compared to the expected values for dicamba + pyridate, glyphosate + pyridate, halauxifen/fluroxypyr + pyridate, fluroxypyr + pyridate, dichlorprop-p + pyridate, and atrazine + pyridate indicated the synergistic interactions of pyridate-based mixtures (Table 4.2). Consistent with percent control ratings, all herbicide treatments except dichlorprop-p, pyridate, 2,4-D, and 2,4-D + pyridate resulted in >90% reduction in shoot dry biomass of the SUS population. Atrazine, bromoxynil/pyrasulfotole, and all pyridate-based herbicide mixtures resulted in 97 to 100% shoot dry biomass reduction of SUS population except for 2,4-D + pyridate. These results are consistent with previous researchers, who have reported excellent levels of reduction in shoot dry biomass (83 to 99%) of glyphosate-resistant kochia with bromoxynil/pyrasulfotole and bromoxynil/MCPA (Burton et al. 2014; Kumar et al. 2014). Dichlorprop-p + pyridate provided 94 to 98% reduction in shoot dry biomass for both MHR and SUS kochia populations. In our previous research, we have also observed 87 to 89% shoot dry biomass reduction of MHR kochia population with dicamba +

halauxifen/fluroxypyr + dichlorprop-p (Dhanda et al. 2023). In that same study, synergistic interactions were also observed when dicamba was tank-mixed with dichlorprop-p, 2,4-D, dichlorprop-p + 2,4-D, and halauxifen/fluroxypyr + 2,4-D for shoot dry biomass reductions (86 to 92%) of MHR kochia (Dhanda et al. 2023).

Field Experiments

Daily mean air temperatures during the study period (end of May through end of June) at KSU-ARC ranged from 14 to 24 C with a total precipitation of 111 mm at Hays site (Figure 4.1). Daily mean air temperatures at KSU-SWREC were similar and ranged from 14 to 29 C during the study period (end of May through end of June). However, the Garden City site had relatively more precipitation of 156 mm during the study period (Figure 4.2).

At the KSU-ARC site, all pyridate-based herbicide mixtures provided $\geq 85\%$ control of MHR kochia population at 14 and 28 DAT, except 2,4-D + pyridate (Table 4.3). The lower control with 2,4-D + pyridate might be because of low efficacy of 2,4-D for kochia control as reported by several previous studies (Friesen et al. 1993; Nandula and Manthey 2002; Tonks and Westra 1997). The relatively lower kochia control with pyridate-based herbicide mixtures in field conditions as compared to greenhouse might be due to the dry field conditions (Figure 4.1) and higher density of kochia in the field ($165 \text{ plants m}^{-2}$) forming a thick mat, which could potentially reduce the herbicide coverage. The proper coverage for contact herbicides like pyridate is important to achieve effective control (Butts et al. 2021). Tank-mixing pyridate with either bromoxynil/pyrasulfotole, fluroxypyr, or atrazine provided 88 to 90% visual control at 14 DAT and 91 to 94% at 28 DAT (Table 4.3). These same treatments reduced kochia density by 96 to 98% compared to nontreated weedy check at 28 DAT. Similarly, two-way tank mixes increased percent visual kochia control from 28 to 88% with glyphosate + pyridate, 33 to 91%

with atrazine + pyridate, and 48 to 89% with dicamba + pyridate as compared to glyphosate, atrazine, or dicamba alone, respectively (Table 4.3). Synergistic interactions were observed when pyridate was tank-mixed with glyphosate, atrazine, or dicamba which resulted in greater percent visual control with mixtures than their standalone applications (Table 4.3). Wicks et al. (1993) also reported 56 to 84% control of atrazine-resistant kochia with cyanazine + atrazine + pyridate at 21 to 30 DAT as compared to cyanazine + atrazine (0 to 7% control). In that same study, higher control of atrazine-resistant kochia was reported with pyridate + fluorchloridone (98 to 100%) as compared to fluorchloridone alone (70 to 96%). In our current study, synergistic interactions were also observed for density reduction when pyridate was tank-mixed with atrazine, dicamba, dichlorprop-p, fluroxypyr, glyphosate, or halauxifen/fluroxypyr (Table 4.3). Consistent with percent control, all pyridate-based mixtures, except 2,4-D + pyridate provided $\geq 90\%$ shoot dry biomass reduction. The application of 2,4-D + pyridate provided 63% kochia control and 65% shoot dry biomass reduction at 28 DAT while 2,4-D alone provided the least kochia control (14%) and shoot dry biomass reduction (23%) (Table 4.3). Tank-mixing of pyridate with glyphosate, atrazine, or dicamba resulted in $\geq 90\%$ shoot dry biomass reduction as compared to glyphosate, atrazine, or dicamba alone treatments (39 to 53% reduction). Colby's analysis for shoot dry biomass reduction revealed a synergistic interaction between glyphosate and pyridate (Table 4.3). For all other pyridate-based mixtures, no significant differences were reported between observed values for shoot dry biomass reduction and expected values indicating additive effects of tank-mixing pyridate for better control of MHR kochia as compared to not adding pyridate in the mixtures. These results are consistent with Wicks et al. (1993), who previously reported greater reduction in shoot dry biomass of atrazine-resistant kochia with pyridate + fluorchloridone (93%) as compared to fluorchloridone alone (76%).

At the KSU-SWREC site, percent visual control and percent reduction of shoot dry biomass of MHR kochia were relatively greater compared to KSU-ARC site. In general, kochia density was less at Garden City (13 plants m⁻²) as compared to Hays (165 plants m⁻²), which might have improved herbicide coverage at KSU-SWREC than on a dense mat of kochia at KSU-ARC (Table 4.4). Also, more precipitation at Garden City (156 mm) than Hays (111 mm) may have resulted in actively growing kochia at time of herbicide application (Figures 4.1 and 4.2). All pyridate-based mixtures, provided $\geq 85\%$ control at 14 DAT and $>90\%$ at 28 DAT, except 2,4-D + pyridate. Bromoxynil/pyrasulfotole alone or with pyridate resulted in 92 to 100% control and 81 to 98% reduction in kochia density at 28 DAT compared to nontreated (Table 4.4). Furthermore, tank-mixing pyridate with atrazine, fluroxypyr, halauxifen/fluroxypyr, and dicamba resulted in 94 to 98% kochia control at 28 DAT. These treatments reduced kochia density by 85 to 97% compared to nontreated (Table 4.4). Colby's analysis further revealed synergistic interactions between pyridate and atrazine as well as pyridate and glyphosate for kochia control (Table 4.4). Consistent with percent control, bromoxynil/pyrasulfotole and all pyridate-based mixtures, except 2,4-D + pyridate provided $\geq 90\%$ shoot dry biomass reduction. Based on Colby's analysis, synergistic interactions for kochia density reduction and shoot dry biomass reduction were observed when pyridate was tank-mixed with atrazine or glyphosate. The shoot dry biomass reduction increased from 18% to 99% with atrazine + pyridate and 15% to 90% with glyphosate + pyridate compared to atrazine or glyphosate alone. Similarly, kochia density reduction was greater (85 to 96%) when pyridate was tank mixed with atrazine or glyphosate as compared to their individual application (23%) without pyridate. The least reduction in kochia density (18 to 23%) and shoot dry biomass reduction (15 to 23%) was obtained with the 2,4-D, glyphosate, and atrazine alone treatments (Table 4.4). These results

indicate that pyridate can play an important role as tank-mix partner with other herbicides for effective control of MHR kochia. These findings are consistent with Seidel and Russell (1990), who previously reported greater control (91 to 97%) of lanceleaf sage (*Salvia reflexa* L. Hornem.) and turnipweed (*Rapistrum rugosum* L. All) when pyridate was tank-mixed with metribuzin as compared to pyridate alone (59 to 78% control).

Practical Implications

With the widespread evolution and spread of MHR weed populations, the use of herbicide mixtures containing multiple SOA could play a crucial role in managing these MHR weed populations and to further delay/mitigate evolution of herbicide resistance (Norsworthy et al. 2012). Results from the current study suggest that tank-mixing of pyridate with atrazine, dicamba, dichlorprop-p, fluroxypyr, glyphosate, or halauxifen/fluroxypyr had synergistic or additive interactions and resulted in greater control of MHR kochia than without adding pyridate in mixtures. These pyridate-based mixtures can effectively control MHR kochia in fallow fields or burndown scenarios (prior to crop planting or after crop harvest) with careful considerations of the rotational crops to be grown. However, it is important to note that these pyridate-based mixtures incur higher costs, ranging from \$33 to \$60 ha⁻¹ compared to their standalone applications, ranging from \$6 to \$33 ha⁻¹ (Table 4.1). At present, according to pyridate label (Anonymous 2024), it is labeled to apply in field corn, seed corn, popcorn, chickpea (*Cicer arietinum* L.), lentils (*Lens culinaris* Medik.) and mint (*Mentha arvensis* L.). It is critical to note that overreliance on these pyridate-based tank mixtures should be avoided to prevent further evolution and spread of MHR kochia populations. Along with these pyridate-based tank-mixtures, growers should also integrate other weed control tactics in their crop rotations, including the use of effective pre-emergence herbicides, competitive crop rotations, narrow crop

row spacing, cover crops, occasional tillage, harvest weed seed control technologies (chaff lining, impact mills), and precision spray technologies for controlling the seed bank of MHR kochia.

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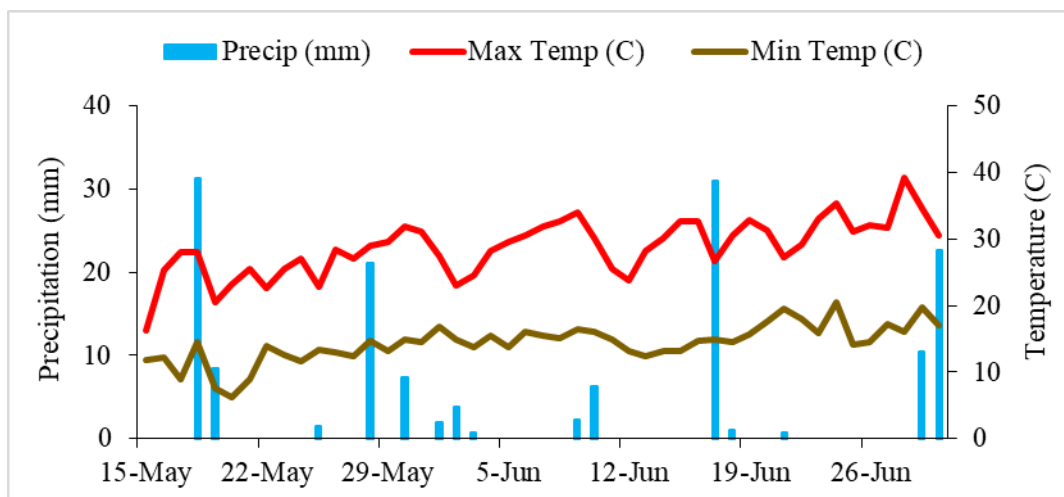


Figure 4.1. Daily minimum and maximum air temperature (C) and precipitation (mm) during the growing season at Kansas State University Agricultural Research Center (KSU-ARC) near Hays, KS.

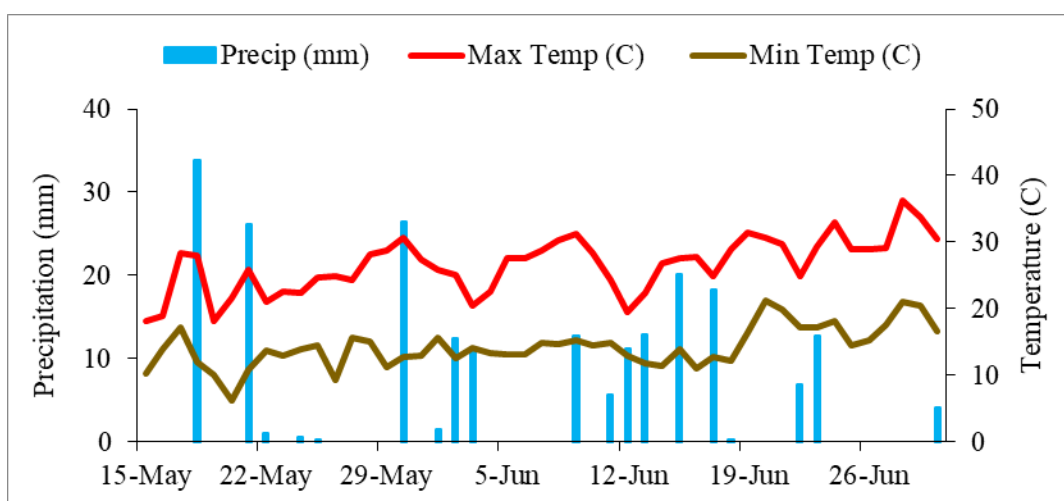


Figure 4.2. Daily minimum and maximum air temperature (C) and precipitation (mm) during the growing season at Kansas State University Southwest Research and Extension Center (KSU-SREC) near Garden City, KS.

Table 4.1. List of herbicides alone or in mixtures with pyridate tested for controlling multiple herbicide-resistant kochia under greenhouse and field experiments.

Herbicide(s)	Trade name	Rate	Cost of herbicide(s)	Manufacturer	Adjuvant ^{ab}
		g ai or ae ha ⁻¹	US\$ ha ⁻¹		
2,4-D	Weedone LV4	538	8	Nufarm	-
Atrazine	AAtrex 4L	560	6	Syngenta	-
Bromoxynil/pyrasulfotole	Huskie	(230 + 41)	33	Bayer Crop Science	NIS
Dicamba	Clarity	560	31	BASF	NIS
Dichlorprop-p	Duplosan	560	14	Nufarm	NIS
Fluroxypyr	Starane Ultra	157	27	Corteva Agriscience	-
Glyphosate	Roundup PowerMax	1260	31	Bayer Crop Science	AMS
Halauxifen/fluroxypyr	Pixxaro	(5 + 123)	17	Corteva Agriscience	-
Pyridate	Tough 5EC	350	27	Belchim Crop Protection	NIS
2,4-D + pyridate	Weedone LV4 + Tough 5EC	538 + 350	35	Nufarm and Belchim Crop Protection	NIS
Atrazine + pyridate	AAtrex 4L + Tough 5EC	560 + 350	33	Syngenta and Belchim Crop Protection	NIS
Bromoxynil/pyrasulfotole + pyridate	Huskie + Tough 5EC	(230 + 41) + 350	60	Bayer Crop Science and Belchim Crop Protection	NIS
Dicamba + pyridate	Clarity + Tough 5EC	560 + 350	58	BASF and Belchim Crop Protection	NIS
Dichlorprop-p + pyridate	Duplosan + Tough 5EC	560 + 350	41	Nufarm and Belchim Crop Protection	NIS
Fluroxypyr + pyridate	Starane Ultra + Tough 5EC	157 + 350	54	Corteva Agriscience and Belchim Crop Protection	NIS
Glyphosate + pyridate	Roundup PowerMax + Tough 5EC	1260 + 350	58	Bayer Crop Science and Belchim Crop Protection	AMS
Halauxifen/fluroxypyr + pyridate	Pixxaro + Tough 5EC	(5 + 123) + 350	44	Corteva Agriscience and Belchim Crop Protection	NIS

^aAbbreviations: AMS, ammonium sulphate; NIS, nonionic surfactant

^bAdjuvant rates: AMS (2% w/v); NIS, 0.25% v/v

Table 4.2. Percent control at 14 and 28 days after treatment (DAT) and shoot dry biomass reduction of multiple herbicide-resistant (MHR) and susceptible (SUS) kochia populations in the greenhouse experiment.^{1, 2}

Treatments	MHR						SUS						
	Weed control			Shoot dry biomass reduction			Weed control			Shoot dry biomass reduction			
	14 DAT		28 DAT	28 DAT		14 DAT		28 DAT		28 DAT			
	Observed	Observed	Expected	Observed	Expected	Observed	Observed	Expected	Observed	Expected			
	%												
2,4-D	28	g	12	f	-	14	e	-	36	h	30	d	-
Atrazine	51	f	49	de	-	55	bc	-	97	ab	97	a	-
Bromoxynil/pyrasulfotole	89	cd	92	b	-	96	a	-	100	a	100	a	-
Dicamba	50	f	42	e	-	45	cd	-	85	de	91	b	-
Dichlorprop-p	39	f	45	de	-	41	cd	-	66	g	70	c	-
Fluroxypyr	40	f	42	e	-	33	d	-	84	ef	88	b	-
Glyphosate	10	h	11	f	-	17	e	-	90	cd	88	b	-
Halauxifen/fluroxypyr	66	e	45	de	-	48	cd	-	84	ef	85	b	-
Pyridate	48	f	52	d	-	53	bc	-	69	g	72	c	-
2,4-D + pyridate	79	d	67	c	58	65	b	60	86	de	83	b	81
Atrazine + pyridate	100	a	100	a	77*	100	a	79*	100	a	100	a	99
Bromoxynil/pyrasulfotole + pyridate	100	a	100	a	96	100	a	98	100	a	100	a	100
Dicamba + pyridate	94	bc	98	a	72*	99	a	75*	94	bc	99	a	99
Dichlorprop-p + pyridate	93	bc	94	ab	74*	94	a	70*	96	ab	97	a	91
Fluroxypyr + pyridate	97	a	98	a	72*	99	a	69*	98	a	99	a	96
Glyphosate + pyridate	98	a	99	a	57*	99	a	65*	98	ab	100	a	97
Halauxifen/fluroxypyr + pyridate	96	ab	100	a	71*	100	a	78*	97	ab	100	a	95

¹Means followed by the same letter within a column are not different according to Fisher's protected LSD at $P < 0.05$.

²Asterisks indicate that observed and expected values were different as determined by t-test ($P < 0.05$), indicating synergistic interaction of herbicides applied in tank-mixtures based on Colby's equation (Eqn. 4.2)

Table 4.3. Average percent control at 14 and 28 days after treatment (DAT), density reduction, and shoot dry biomass reduction of atrazine-, dicamba-, and glyphosate-resistant kochia in a field study during 2023 at Kansas State University Agricultural Research Center (KSU-ARC) near Hays, KS.^{1, 2}

Treatments	Weed control			Density reduction		Shoot dry biomass reduction	
	14 DAT	28 DAT		28 DAT		28 DAT	
	Observed	Observed	Expected	Observed	Expected	Observed	Expected
	%						
2,4-D	10 f	14 i	-	31 j	-	23 h	-
Atrazine	22 f	33 gh	-	41 hi	-	47 fg	-
Bromoxynil/pyrasulfotole	74 bc	81 bc	-	81 c	-	85 bc	-
Dicamba	41 e	48 fg	-	53 fg	-	53 fg	-
Dichlorprop-p	53 de	61 def	-	55 ef	-	65 de	-
Fluroxypyr	39 e	69 cd	-	65 d	-	74 cd	-
Glyphosate	19 f	28 h	-	37 ij	-	39 g	-
Halauxifen/fluroxypyr	45 e	75 cd	-	63 de	-	71 d	-
Pyridate	46 e	51 ef	-	46 gh	-	58 ef	-
2,4-D + pyridate	70 cd	63 de	57	71 d	63	65 de	68
Atrazine + pyridate	88 a	91 a	66*	97 a	65*	91 ab	78
Bromoxynil/pyrasulfotole + pyridate	90 a	94 a	91	98 a	90	95 a	94
Dicamba + pyridate	84 abc	89 ab	74*	90 ab	79*	90 ab	79
Dichlorprop-p + pyridate	85 ab	87 ab	82	91 ab	78*	90 ab	85
Fluroxypyr + pyridate	87 a	92 a	85	96 a	81*	90 ab	89
Glyphosate + pyridate	85 ab	88 ab	65*	86 bc	66*	91 ab	74*
Halauxifen/fluroxypyr + pyridate	81 bc	90 ab	88	95 a	80*	91 ab	88

¹Means followed by the same letter within a column are not different according to Fisher's protected LSD at $P < 0.05$.

²Asterisks indicate that observed and expected values were different as determined by t-test ($P < 0.05$), indicating synergistic interaction of herbicides applied in tank-mixtures based on Colby's equation (Eqn. 4.2)

Table 4.4. Average percent control at 14 and 28 days after treatment (DAT), density reduction, and shoot dry biomass reduction of atrazine- and glyphosate-resistant kochia in a field study during 2023 at Kansas State University Southwest Research and Extension Center (KSU-SREC) near Garden City, KS.^{1, 2}

Treatments	Weed control					Density reduction			Shoot dry biomass reduction		
	14 DAT		28 DAT			28 DAT			28 DAT		
	Observed		Observed	Expected		Observed	Expected		Observed	Expected	
	%										
2,4-D	33	i	24	g	-	18	f	-	23	e	-
Atrazine	23	i	23	g	-	23	f	-	18	e	-
Bromoxynil/pyrasulfotole	89	cd	92	bc	-	81	bc	-	91	abc	-
Dicamba	70	gh	78	de	-	65	d	-	75	cd	-
Dichlorprop-p	69	gh	75	e	-	52	e	-	71	cd	-
Fluroxypyr	75	fg	86	cd	-	70	cd	-	88	bc	-
Glyphosate	25	i	24	g	-	23	f	-	15	e	-
Halauxifen/fluroxypyr	84	de	85	cd	-	70	cd	-	81	bcd	-
Pyridate	60	h	66	f	-	63	de	-	62	d	-
2,4-D + pyridate	80	ef	77	de	76	70	cd	70	75	cd	73
Atrazine + pyridate	96	a	98	a	65*	96	a	77*	99	a	73*
Bromoxynil/pyrasulfotole + pyridate	98	a	100	a	99	98	a	93	99	a	96
Dicamba + pyridate	90	bc	94	ab	95	85	ab	83	92	abc	92
Dichlorprop-p + pyridate	85	de	91	bc	91	85	ab	84	91	abc	93
Fluroxypyr + pyridate	94	ab	96	ab	94	97	a	88	96	ab	95
Glyphosate + pyridate	87	de	92	bc	80*	85	ab	70*	90	abc	69*
Halauxifen/fluroxypyr + pyridate	95	a	95	ab	96	92	ab	85	94	ab	96

¹Means followed by the same letter within a column are not different according to Fisher's protected LSD at $P < 0.05$.

²Asterisks indicate that observed and expected values were different as determined by t-test ($P < 0.05$), indicating synergistic interaction of herbicides applied in tank-mixtures based on Colby's equation (Eqn. 4.2)

Chapter 5 - Influence of cover crop residue and residual herbicide on emergence dynamics of glyphosate-resistant Palmer amaranth in grain sorghum

Abstract

A field study was conducted from 2020 to 2023 at Kansas State University Agricultural Research Center near Hays, KS to understand the emergence dynamics and periodicity of glyphosate-resistant (GR) Palmer amaranth (*Amaranthus palmeri* S. Watson) as influenced by cover crop (CC) residue and residual herbicide in grain sorghum. Study site was under wheat-sorghum-fallow rotation with a natural seedbank of GR *A. palmeri*. Treatments included (1) fall-planted CC mixture (triticale/winter peas/radish/rapeseed) after wheat harvest and terminated at triticale heading stage (next spring prior to sorghum planting) with glyphosate alone or (2) glyphosate plus acetochlor/atrazine, (3) chemical fallow (no CC but treated with acetochlor/atrazine and dicamba prior to sorghum planting), and (4) nontreated control (no CC and no herbicide). Results indicated that CC terminated with glyphosate plus acetochlor/atrazine had a delayed and reduced cumulative emergence of GR *A. palmeri* as compared to chemical fallow and CC terminated with glyphosate alone across all 3 years. Compared to chemical fallow, the CC terminated with glyphosate alone and glyphosate plus acetochlor/atrazine required 66 to 643 and 105 to 1257 more cumulative growing degree days, respectively, to achieve 90% cumulative emergence of GR *A. palmeri* across all three years. The combined effect of CC residue with glyphosate plus acetochlor/atrazine reduced the cumulative emergence of GR *A. palmeri* by 42 to 56% and 82 to 94% as compared to chemical fallow and nontreated control,

respectively. These results suggest that fall-planted CC combined with a residual herbicide at termination can be utilized for GR *A. palmeri* suppression in grain sorghum.

Keywords: Cover crop; emergence dynamics; grain sorghum; Central Great Plains

Introduction

Grain sorghum (*Sorghum bicolor* (L.) Moench) is the fifth most important crop grown in arid and semiarid regions worldwide (Visarada and Aruna 2019). It is a drought-tolerant crop that is productive in moisture-limited environments and is mainly used for food, feed, and fuel (Mundia et al. 2019). Grain sorghum is the fourth largest crop after corn (*Zea mays* L.), soybean [*Glycine max* (L.) Merr.], and wheat (*Triticum aestivum* L.) in the U.S. (USDA-NASS 2023). In 2021, grain sorghum was harvested from 2.63 million ha in the U.S., yielding 11,375 million kg of grain with Kansas contributing 59% of this production (USDA-NASS 2023). Weeds are a serious production challenge in grain sorghum and if weeds are not controlled in grain sorghum, U.S. farmers would lose approximately 5,700 million kg of grain sorghum annually valued at approximately US \$953 million (Dille et al. 2020). Moore et al. (2004) reported that grain sorghum yield was reduced by 1.8 to 3.5% with one Palmer amaranth (*Amaranthus palmeri* S. Watson) plant in 15 m of sorghum row.

Amaranthus palmeri is the most troublesome broadleaf weed in grain sorghum in the United States (Van Wyche 2020). It is a summer annual C₄ dioecious plant belonging to the Amaranthaceae family. It is native to the southwestern United States and northwestern Mexico and is widely distributed in several countries across Africa, Asia, Europe, North America, and South America (Roberts and Florentine 2022; Sauer 1957). *Amaranthus palmeri* exhibits an extended emergence period, high photosynthetic rate, prolific seed production (up to 600,000 seeds per female plant), high genetic diversity within and among field populations, and ability to evolve herbicide resistance (Chahal et al. 2015; Horak and Loughin 2000; Keeley et al. 1987; Ward et al. 2013). Resistance to 10 different herbicide sites of action has been identified in populations of *A. palmeri* (WSSA Groups 2, 3, 4, 5, 6, 9, 10, 14, 15, and 27) (Heap 2023).

Multiple herbicide-resistant (MHR) *A. palmeri* populations have been reported in the Central Great Plains (CGP) region, including Kansas (Chahal et al. 2015; Heap 2023; Jhala et al. 2014; Kumar et al. 2019a; 2020a).

Several studies have demonstrated the importance of different management strategies that can impact *A. palmeri* emergence. For instance, Chahal et al. (2021) reported that an early-season shallow tillage (10-cm depth) in the first week of June, increased cumulative emergence of *A. palmeri* (1674 plants m⁻²) in the fallow field as compared to mid (mid-June)- and late (first week of July)-season tillage operations (533 to 869 plants m⁻²) in southcentral Nebraska. DeVore et al. (2013) reported a 94% reduction of GR *A. palmeri* emergence when cereal rye (*Secale cereale* L.) CC was used in combination with deep tillage (30-cm depth) in soybean compared to no deep tillage or cereal rye CC in Arkansas. Palhano et al. (2018) reported that cereal rye CC reduced *A. palmeri* emergence by 83% in cotton (*Gossypium hirsutum* L.) as compared to no CC in Arkansas. In a greenhouse study, Teasdale et al. (2005) found that hairy vetch (*Vicia villosa* Roth.) residue (500 g m⁻²) and metolachlor (10 g ha⁻¹) had a synergistic effect and reduced smooth pigweed (*Amaranthus hybridus* L.) emergence by 86% compared to no metolachlor or no hairy vetch residue. Perkins et al. (2021) reported that CC mixture of cereal rye and hairy vetch in combination with pyroxasulfone + flumioxazin, flumioxazin, pyroxasulfone, or acetochlor delayed *A. palmeri* emergence by 27 to 34 days in soybean compared to CC with no residual herbicides. Similarly, Liu et al. (2022) documented a differential emergence pattern and periodicity among *A. palmeri* populations from Kansas, Nebraska, Colorado, Oklahoma, and Texas in no-tillage (NT) field in western Kansas.

Winter wheat-sorghum-fallow (WSF) is a dominant crop rotation in the semiarid CGP region, where producers generally rely on POST herbicides for weed control during the fallow

period (Holman et al. 2022; Kumar et al. 2019a; 2019b). In this 3-yr crop rotation, a fallow period of about 10 months occurs after the winter wheat harvest and prior to grain sorghum planting in the following year (Kumar et al. 2020b). During this fallow period, late-season infestation of *A. palmeri* in postharvest wheat stubble can produce significant number of seeds (58,300 seeds m⁻² if left uncontrolled), contributing to the soil seedbank for infestations in the subsequent summer crops, including grain sorghum (Kumar et al. 2021). In these NT-based cropping systems, the majority of *A. palmeri* seeds are concentrated on the soil surface and can emerge easily (Buhler 1992; Buhler et al. 1996; Cardina et al. 1991; Oryokot et al. 1997). Replacing a portion of this fallow period with a CC may provide effective weed suppression in NT cropping systems of the CGP region (Kumar et al. 2020b). However, limited information exists for the combined effect of CC residue and residual herbicides on emergence of *A. palmeri* in NT grain sorghum in this dryland region. The main objective of this study was to determine the effect of fall-planted CC after wheat harvest and terminated in the following spring with glyphosate alone or glyphosate with acetochlor/atrazine on (1) emergence pattern and (2) emergence periodicity of GR *A. palmeri* in NT dryland grain sorghum.

Materials and Methods

Field Experiment

A field study was conducted at Kansas State University Agricultural Research Center near Hays (KSU-ARCH), KS (38.85196°N, 99.34279°W) from fall 2020 through fall 2023. Field site was under a NT dryland WSF rotation with a history of natural seedbank of GR *A. palmeri*. All three phases of the crop rotation (WSF) were present in each experimental year. The soil type at the experimental site was Roxbury silt loam with a pH of 6.9 and organic matter of 1.6%. A separate greenhouse study confirmed that *A. palmeri* population from the study site survived

5,040 g ha⁻¹ (4 times the field-use rate) of glyphosate (data not shown). Each year, a CC mixture of winter triticale ($\times$ Triticosecale Wittm.) (60%)/winter peas (*Pisum sativum* L.) (30%)/rapeseed (*Brassica napus* L.) (5%)/radish (*Raphanus sativus* L.) (5%) was drilled at a seeding rate of 67 kg ha⁻¹ in wheat stubble during fall (September/October) and terminated in the following spring at the triticale heading stage (Table 1). During each spring, four treatments were established: (1) nontreated control (where no CC was planted and no herbicides were applied to control weeds), (2) chemical fallow [where no CC was planted but the plot area was treated with glyphosate (Roundup PowerMax, Bayer Crop Science) at 1260 g ae ha⁻¹ plus a premix of acetochlor/atrazine (Degree Xtra, Bayer Crop Science) at 1665/826 g ai ha⁻¹ plus dicamba (Clarity, BASF) at 560 g ae ha⁻¹ at the same time as CC termination], (3) CC terminated with glyphosate at 1260 g ae ha⁻¹, and (4) CC terminated with glyphosate at 1260 g ae ha⁻¹ plus a premix of acetochlor/atrazine at 1665/826 g ai ha⁻¹. Each year, treatments were arranged in a randomized complete block design with four replications. During 2020-2021 experimental year, the nontreated control was not present and there were only three treatments. The individual plot size was 45-m long and 13-m wide each year. During 2021-2022 and 2022-2023, the original chemical fallow plot was further divided into two to have both nontreated control and chemical fallow treatments (each plot 45-m long and 6.5-m wide). A grain sorghum hybrid ‘DKS 38-16’ was planted at a seeding rate of 114,855 seeds ha⁻¹ in rows spaced 76-cm apart within 3 to 4 weeks of CC termination each year. The details for planting and termination dates of CC as well as grain sorghum planting and harvesting dates for each experimental year are presented in Table 1. All local agronomic practices for grain sorghum production as recommended by Kansas State University were followed (Ciampitti et al. 2022).

Data Collection

Each year, the aboveground shoot biomass of CC was manually harvested from two 1-m² quadrats from each plot just prior to CC termination and oven-dried at 72 C for 4 days to obtain dry biomass. Two permanent 1-m² quadrats were established in each plot immediately after the termination of CC for GR *A. palmeri* emergence counts. Newly emerged GR *A. palmeri* seedlings from each permanent quadrat were counted when cotyledons were fully expanded and removed manually every week starting from termination of CC throughout sorghum growing season (Hartzler et al. 1999). Kochia (*Bassia scoparia* L.) and puncturevine (*Tribulus terrestris* L.) were also present each year and were removed manually along with GR *A. palmeri* every week. The end date for the emergence count of GR *A. palmeri* was chosen when no new emergence occurred over 21 days period each year. The average number of *A. palmeri* seedlings from the two permanent quadrats in each plot at each count timing was used in the data analysis. Data on daily minimum and maximum air temperature and precipitation during each growing season were obtained from the Kansas State University mesonet weather station (<https://mesonet.k-state.edu>) located approximately 400 m away from the study site (38.8495°N, 99.3446°W). Growing degree days (GDD) and cumulative GDD (cGDD) were calculated (started at the day of CC termination) from the daily minimum and maximum air temperatures using equations 5.1 and 5.2, respectively (McMaster and Wilhelm 1997).

$$GDD_{daily} = \left[\frac{T_{max} + T_{min}}{2} \right] - T_{base} \quad \text{Eqn. 5.1}$$

$$cGDD = \sum_{i=1}^n GDD_{daily} \quad \text{Eqn. 5.2}$$

where GDD_{daily} is the daily accumulated GDD (it is a nonnegative value, therefore negative GDD_{daily} was replaced by 0), T_{max} is the daily maximum air temperature (C), T_{min} is the daily minimum air temperature (C), T_{base} is the base temperature below which plant growth ceases and was considered as 10 C for *A. palmeri* (Norsworthy et al. 2008), and n is the number of days in each growing season for which the emergence counts were recorded.

Statistical Analyses

Data on weekly GR *A. palmeri* emergence were subjected to ANOVA using PROC MIXED procedure and were tested for homogeneity of variance and normality of the residuals using the PROC UNIVARIATE procedure in SAS 9.3 (SAS Institute, Inc., SAS Campus Drive, Cary, NC). Data were log-transformed to improve the normality of the residuals and homogeneity of variance; however, back-transformed data were presented with mean separation based on the transformed data. The fixed effects in the ANOVA model included treatment, year, weekly emergence timings, and their interactions. The random effects in the ANOVA model included replication and all interactions involving replication. Due to significant year-by-treatment interaction ($P = 0.0321$), data were analyzed separately for each year. The interaction between treatment and weekly emergence timing was significant ($P = <0.001$), therefore, data were sorted by weekly emergence timings using PROC SORT. Treatment means were separated using Fisher's protected LSD test ($P < 0.05$) for each emergence timing.

The percent cumulative emergence of GR *A. palmeri* in each treatment from each replication was calculated as the sum of emergence on a sample date and all previous sample dates and expressed as a percentage of total emergence in each growing season (Jha and Norsworthy 2009). Data on percent cumulative emergence of GR *A. palmeri* from all four

treatments were regressed against cGDD with a three-parameter log-logistic model using the ‘drc’ package in R software (Knezevic et al. 2007; Ritz and Streibig 2005; Seefeldt et al. 1995).

$$Y = \{d/1 + \exp[b(\log X - \log E_{50})]\} \quad \text{Eqn. 5.3}$$

where Y is the percent cumulative emergence of GR *A. palmeri*, X is the cGDD, parameter d in the model is the maximum cumulative emergence which was fixed to “100” because the percent cumulative emergence was based on the total emergence, E_{50} is the cGDD required to reach 50% cumulative emergence, and the parameter b is the slope at the inflection point. The slope (b) represents the emergence rate of GR *A. palmeri* over cGDD, and the negative value of the slope indicates rapid emergence (Kumar et al. 2018). This model was selected based on Akaike’s criteria as described in Ritz and Spiess (2008). The P-value > 0.05 for the lack-of-fit test indicated that the model (Equation 3) adequately explained the percent cumulative emergence of GR *A. palmeri* (Ritz and Streibig 2005). Parameter estimates, standard errors, and E_{50} values for each treatment were determined. The cGDD required for 10% (ED_{10}) and 90% (ED_{90}) cumulative emergence of GR *A. palmeri* and duration (cGDDs for 10% to 90% emergence, i.e., $E_{90} - E_{10}$) for each treatment were predicted from the fitted model. ED_{10} , ED_{50} , and ED_{90} values were compared among treatments using the approximate t -test with the ‘compParm’ and ‘EDcomp’ functions in the ‘drc’ package of the R software (Knezevic et al. 2007; Ritz et al. 2015).

Results and Discussion

Variable amount and frequency of precipitation were observed at KSU-ARCH during the experimental periods from 2020 to 2023 (Figure 5.1). The total amount of precipitation received during the CC growing season (Sep to May) in 2020-2021, 2021-2022 and 2022-2023 were 217, 99, and 130 mm, respectively (Figure 5.1). The dry CC biomass at the time of termination was

1520, 1130, and 1470 kg ha⁻¹ in 2021, 2022, and 2023, respectively. Total precipitation amount during the grain sorghum growing seasons (June to Oct) in 2021, 2022, and 2023 was 256, 171, and 237 mm, respectively (Figure 5.1).

Emergence Periodicity of GR *A. palmeri*

Amaranthus palmeri seedlings emerged from May 12 through Sep 15 in 2021, May 12 through July 14 in 2022, and June 2 through July 14 in 2023. The prolonged emergence in 2021 might be due to relatively higher precipitation events (17) of each >10 mm from May to Sep as compared to 2022 (9 events) and 2023 (10 events) growing seasons (Figure 5.1). Across three years, the emergence initiation of GR *A. palmeri* was delayed by 0 to 21 days and 7 to 28 days under the CC terminated with glyphosate only and CC terminated with glyphosate plus acetochlor/atrazine, respectively, compared to chemical fallow (Figures 5.2 to 5.4). The CC terminated with glyphosate plus acetochlor/atrazine reduced the cumulative emergence of GR *A. palmeri* by 42 to 56% compared to the chemical fallow and by 82 to 94% compared to nontreated control across three years (Figures 5.2 to 5.4).

2021 Growing Season

The total cumulative emergence of GR *A. palmeri* at the end of the grain sorghum growing season was 22 (SE = 4), 23 (SE = 4), and 11 (SE = 3) seedlings m⁻² under chemical fallow, CC terminated with glyphosate, and CC terminated with glyphosate plus acetochlor/atrazine, respectively (Figure 5.2). GR *A. palmeri* in chemical fallow started to emerge on May 12 with a major emergence observed from end of May through end-June (Figure 5.2). In contrast, both CC treatments had a delayed GR *A. palmeri* emergence by 21 to 28 days with the first seedlings emerging between June 2 and June 9 and a major emergence occurred from early-June to mid-July. Out of the total rainfall (358 mm), 64% occurred from mid-May to

mid-July with ten rainfall events of >10 mm which coincided with the major emergence period of GR *A. palmeri* (Figures 5.1 and 5.2). These results indicate that CC residue suppressed GR *A. palmeri* emergence and provided less initial weed competition to grain sorghum in 2021.

2022 Growing Season

The total cumulative emergence of GR *A. palmeri* at the end of the grain sorghum growing season was 33 (SE = 6), 12 (SE = 4), 31 (SE = 6), and 7 (SE = 2) seedlings m⁻² under nontreated, chemical fallow, CC terminated with glyphosate, and CC terminated with glyphosate plus acetochlor/atrazine, respectively (Figure 5.3). *Amaranthus palmeri* seedlings started to emerge nearly at the same time (May 12 through May 19) under all treatments except CC terminated with glyphosate plus acetochlor/atrazine (delayed by 7 days) in 2022 growing season (Figure 5.3). This was possibly due to relatively less accumulated dry biomass of CC (1130 kg ha⁻¹) at termination because of relatively low precipitation (99 mm) during the CC growing season (Figures 5.1 and 5.3). Although GR *A. palmeri* seedlings emerged nearly at the same time, the total seasonal cumulative emergence of GR *A. palmeri* was reduced by 68% under chemical fallow, by 14% under CC terminated with glyphosate, and by 82% under CC terminated with glyphosate plus acetochlor/atrazine as compared to nontreated control. Interestingly, both CC treatments significantly reduced early-season (May 12 through June 2) emergence of GR *A. palmeri* by 66 to 92% as compared to nontreated control. Our results are consistent with Cornelius and Bradley (2017), who also reported that cereal rye CC reduced early-season emergence of common waterhemp (*Amaranthus rudis* Sauer) by 35% compared to nontreated control in soybean. Majority of the GR *A. palmeri* seedlings emerged from mid-May through early June under nontreated control and chemical fallow treatments. In contrast, the emergence of GR *A. palmeri* was mostly observed from late-May through end-June under CC

terminated with glyphosate alone and from late-May through early June under CC terminated with glyphosate plus acetochlor/atrazine (Figure 5.3). All the major emergence periods of *GR A. palmeri* in May and June coincided with the precipitation events with 60% of the total seasonal rainfall occurred during these two months (Figure 5.1). These findings are consistent with Wiggins et al (2015), who concluded that CC alone was not sufficient for season-long control of *GR A. palmeri* and suggested integrating residual herbicides to complement the suppressive effect of CC.

2023 Growing Season

The total cumulative emergence of *GR A. palmeri* at the end of the grain sorghum growing season was 60 (SE = 8), 9 (SE = 2), 29 (SE = 6), and 4 (SE = 1) seedlings m⁻² under nontreated, chemical fallow, CC terminated with glyphosate, and CC terminated with glyphosate plus acetochlor/atrazine, respectively (Figure 5.4). The *GR A. palmeri* seedlings started to emerge on June 2 in nontreated control and on June 9 in chemical fallow and CC terminated with glyphosate alone (Figure 5.4). In contrast, the emergence of *GR A. palmeri* in CC terminated with glyphosate plus acetochlor/atrazine was delayed and first observed on June 30 indicating the emergence delay by 28 days as compared to nontreated control and by 21 days compared to chemical fallow (Figure 5.4). Majority of the *GR A. palmeri* seedlings emerged from June 2 through June 30 in nontreated control, June 9 through July 7 in chemical fallow, June 9 through July 14 in CC terminated with glyphosate alone, and June 30 through July 7 in CC terminated with glyphosate plus acetochlor/atrazine. Most of the *GR A. palmeri* seedlings emerged in June and July, which coincided with eight major precipitation events (each event > 5 mm) and two events of (>20 mm) contributing 44% of the total season rainfall (312 mm) in 2023 (Figures 5.1 and 5.4). The cumulative seasonal emergence of *GR A. palmeri* was reduced by 85% in chemical

fallow, 51% in CC terminated with glyphosate alone, and 94% in CC terminated with glyphosate plus acetochlor/atrazine as compared to nontreated control (Figure 5.4). These results agree with DeVore et al. (2012), who reported that fall-planted cereal rye CC reduced the emergence of GR *A. palmeri* by 67% in cotton compared to no CC. Similarly, McCall (2018) observed a delayed emergence of GR *A. palmeri* under winter wheat, spring oat, or spring oat/spring pea CC terminated with glyphosate plus flumioxazin/pyroxasulfone as compared to CC terminated with glyphosate only.

Emergence Pattern of GR *A. palmeri*

Across three years, 66 to 643 and 105 to 1257 more cGDD were required to achieve 90% cumulative emergence of GR *A. palmeri* under the CC terminated with glyphosate alone and CC terminated with glyphosate plus acetochlor/atrazine, respectively, compared to chemical fallow (Tables 5.2 to 5.4).

2021 Growing Season

Only 230 to 286 cGDD were required for 10% cumulative emergence (E_{10} value) of GR *A. palmeri* under chemical fallow and CC terminated with glyphosate alone, whereas about 498 cGDD were required to achieve 10% cumulative emergence of GR *A. palmeri* under CC terminated with glyphosate plus acetochlor/atrazine (Table 5.2). Similarly, about 205 and 580 more cGDD were required to achieve 50% cumulative emergence (E_{50} values) of GR *A. palmeri* for CC terminated with glyphosate and CC terminated with glyphosate plus acetochlor/atrazine, respectively, as compared to chemical fallow. For 90% cumulative emergence (E_{90} value) of GR *A. palmeri*, about 1000, 1643, and 2257 cGDD were required for chemical fallow, CC terminated with glyphosate, and CC terminated with glyphosate plus acetochlor/atrazine, respectively (Table 5.2). The CC terminated with glyphosate or with glyphosate plus acetochlor/atrazine exhibited a

prolonged emergence duration ($E_{90} - E_{10}$) of GR *A. palmeri* and required additional 587 and 989 cGDD, respectively, as compared to chemical fallow. The greater cGDD requirements for 10, 50 and 90% cumulative emergence of GR *A. palmeri* under CC treatments might be due to alteration in soil microenvironment with decreasing light penetration and soil temperature as compared to no CC treatments (Teasdale and Mohler 1993). Our results are consistent with Perkins et al. (2021), who previously reported that applications of pyroxasulfone + flumioxazin, flumioxazin, pyroxasulfone, or acetochlor at three weeks after fall-planted cereal rye + hairy vetch CC termination delayed *A. palmeri* emergence by 27 to 34 days compared to CC termination with glyphosate + dicamba in soybeans.

2022 Growing Season

GR *A. palmeri* seedlings emerged quickly in the nontreated control and required only 27 cGDD to achieve 10% cumulative emergence in the absence of CC and residual herbicide (Table 5.3). In contrast, GR *A. palmeri* under the chemical fallow treatment required 47 cGDD for 10% cumulative emergence followed by CC terminated with glyphosate alone (70 cGDD), and CC terminated with glyphosate plus acetochlor/atrazine (116 cGDD) (Table 5.3). To reach 50% cumulative emergence, the CC terminated with glyphosate alone required 103 and 52 more cGDD compared to nontreated and chemical fallow, respectively. Similarly, the CC terminated with glyphosate plus acetochlor/atrazine required 187 and 136 additional cGDD to achieve 50% cumulative emergence of GR *A. palmeri* compared to nontreated and chemical fallow, respectively. Furthermore, to reach 90% cumulative emergence, the CC terminated with glyphosate plus acetochlor/atrazine required 557 cGDD and had a prolonged period of emergence ($E_{90} - E_{10} = 441$ cGDD). In contrast, the nontreated control required only 168 cGDD to achieve 90% cumulative emergence of GR *A. palmeri* and had a short period of emergence

($E_{90} - E_{10} = 141$ cGDD) (Table 5.3). These results are consistent with McCall (2018), who reported that an additional 80 cGDD were required to reach 50% emergence of GR *A. palmeri* under winter wheat CC terminated with glyphosate plus flumioxazin/pyroxasulfone as compared to CC terminated with glyphosate only in soybean.

2023 Growing Season

Only 97 cGDD were required to achieve 10% cumulative emergence of GR *A. palmeri* in nontreated control. Compared to 2021 and 2022 growing seasons, the CC terminated with glyphosate alone required fewer cGDD (132) than chemical fallow (192) to achieve 10% cumulative emergence of GR *A. palmeri* (Table 5.4). This might be because of frequent precipitation events during late May and early June. Among all treatments, CC terminated with glyphosate plus acetochlor/atrazine required the greatest cGDD (427) for 10% cumulative emergence of GR *A. palmeri* (Table 5.4). In nontreated control, GR *A. palmeri* required 204 cGDD for 50% cumulative emergence, while CC terminated with glyphosate plus acetochlor/atrazine required significantly higher cGDD (562) (Table 5.4). Similarly, to achieve 90% cumulative emergence of GR *A. palmeri*, the highest cGDD (739) were required under the CC terminated with glyphosate plus acetochlor/atrazine, while only 429 cGDD were required under nontreated control. These results are consistent with Teasdale et al. (2003), who previously reported that hairy vetch residue and pre-emergence application of metolachlor delayed weed emergence by 7 to 10 days compared to herbicide alone. The delay in emergence of GR *A. palmeri* would provide an initial competitive benefit to grain sorghum.

Practical Implications

Grain sorghum producers face a real challenge to control GR *A. palmeri* in the NT dryland CGP region. Results from this study indicate that fall-planted CC after winter wheat

harvest and terminated in the following spring with glyphosate plus acetochlor/atrazine can delay and reduce the cumulative emergence of GR *A. palmeri* in grain sorghum. Different *A. palmeri* populations within a CGP region could exhibit a differential emergence pattern as previously observed by Liu et al. (2022). Therefore, the findings of the current study will further help to improve the prediction models for *A. palmeri* emergence under CC plus residual herbicide strategy to build decision support tools (DSTs) for cost-effective and sustainable management of GR *A. palmeri* in the region. These combined practices of CC and residual herbicides would also help growers to target smaller GR *A. palmeri* seedlings with POST herbicides for effective in-season control. Fewer and late emerging GR *A. palmeri* plants would have lesser biomass accumulation, and ultimately lesser seed production, that can further help to prevent replenishment of the soil seedbank. Although this study investigated only one residual herbicide, yet sorghum producers should diversify their residual herbicide programs annually to prevent the selection of *A. palmeri* populations with resistance to residual herbicides. Future studies should investigate weed suppression with CC plus residual herbicides by considering the soil water budget and subsequent crop yield implications in the WSF rotation.

Nonetheless, the extended emergence duration of the GR *A. palmeri* observed under CC terminated with glyphosate plus acetochlor/atrazine further warrants the need for a season-long integrated *A. palmeri* management strategy in grain sorghum. Previous studies have also established that adequate CC biomass is highly crucial to achieve suppression and delayed emergence of *A. palmeri* (Devore et al. 2013; Palhano et al. 2018; Perkins et al. 2021) and infrequent rainfall patterns is a major constraint to the widescale adoption of CC in the NT dryland CGP region. As evident from this research, limited precipitation during winter months in some years can result in lower CC biomass accumulation and inactivation of residual herbicides

resulting into loss of suppressive effect of CC and residual herbicide on *A. palmeri* emergence. Considering the aforementioned challenges with CC plus residual herbicides and rapid evolution of *A. palmeri* with multiple resistance to POST herbicides, additional novel weed control strategies [such as electric weeder and harvest weed seed control (chaff lining and seed impact mill)] should also be integrated for managing GR *A. palmeri* seedbanks in this NT dryland region. Based on available soil moisture, the fallow phase after grain sorghum harvest in the WSF rotation could also be replaced with the spring-planted CC such as barley, oat, or pea for summer annual weed suppression (Obour et al. 2022).

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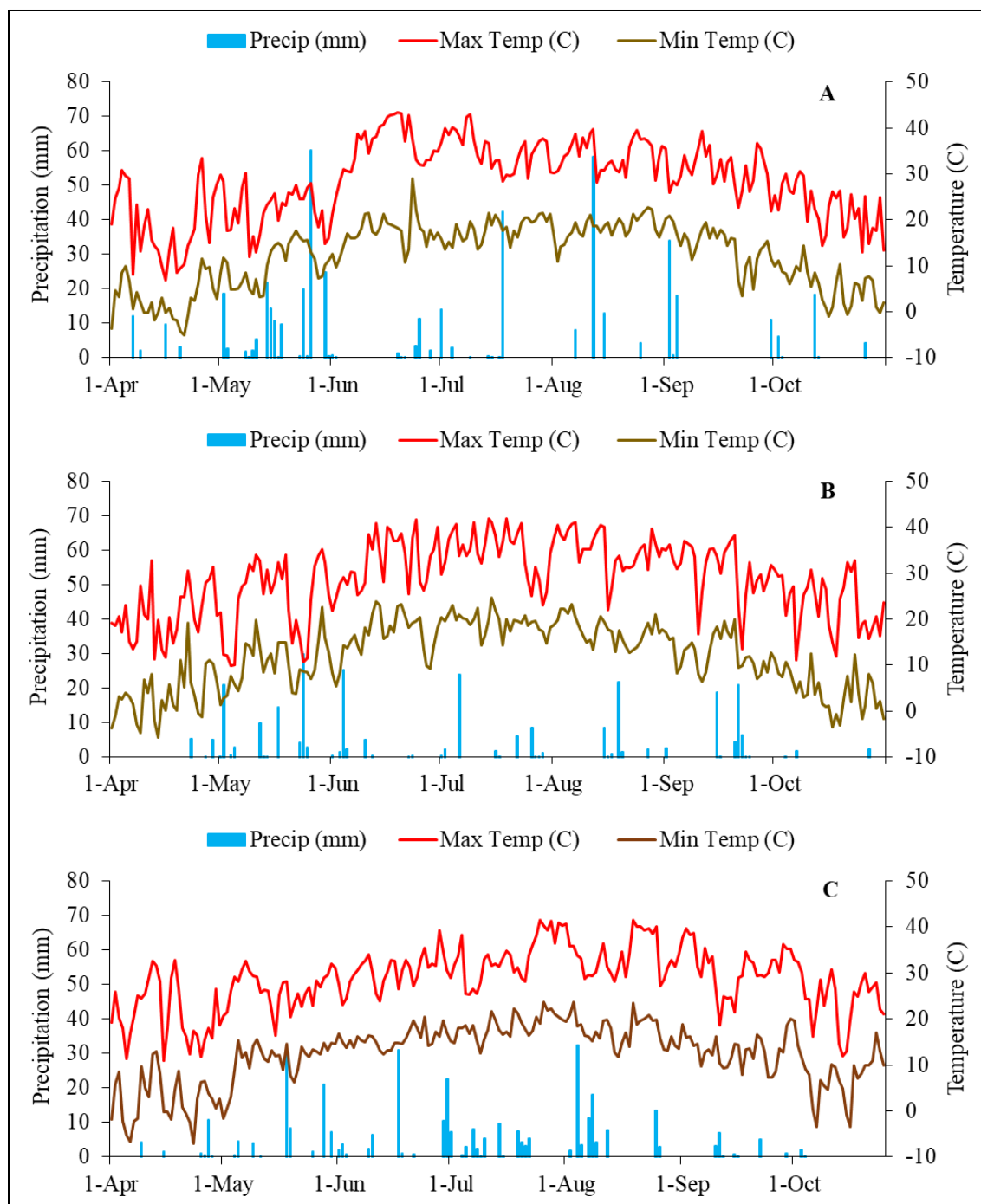


Figure 5.1. Daily minimum and maximum air temperature (°C) and precipitation (mm) during the growing season of 2021 (A), 2022 (B), and 2023 (C).

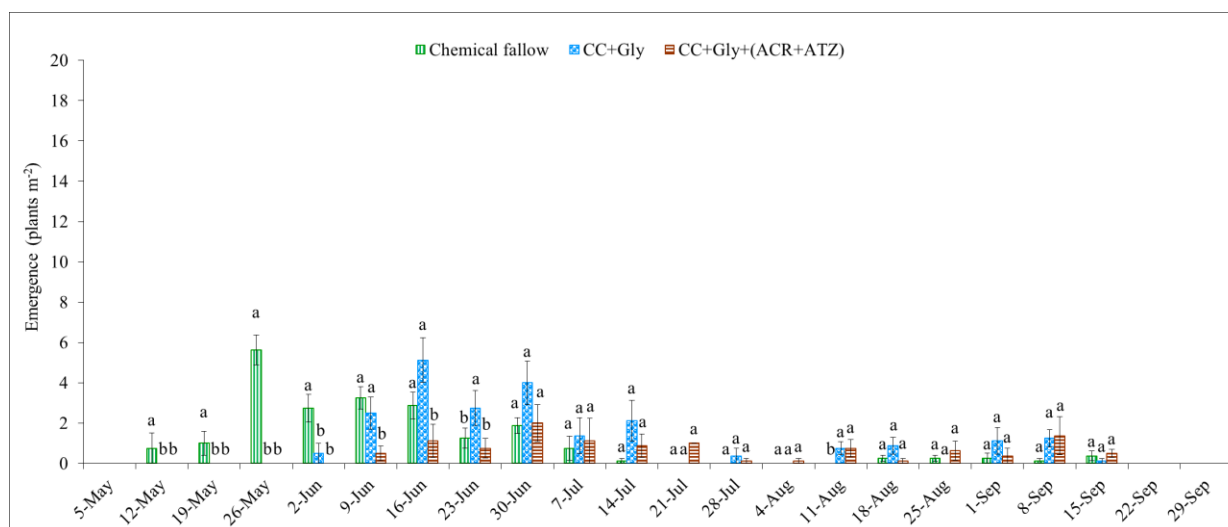


Figure 5.2. Weekly emergence counts of glyphosate-resistant *Amaranthus palmeri* under different treatments in 2021. CC+Gly indicates cover crop terminated with glyphosate; CC+Gly+(ACR+ATZ) indicates cover crop terminated with glyphosate plus acetochlor/atrazine. Means followed by the same letter within each timing are not significantly different according to Fisher's protected LSD test at $P < 0.05$. Error bars represent standard error of the mean.

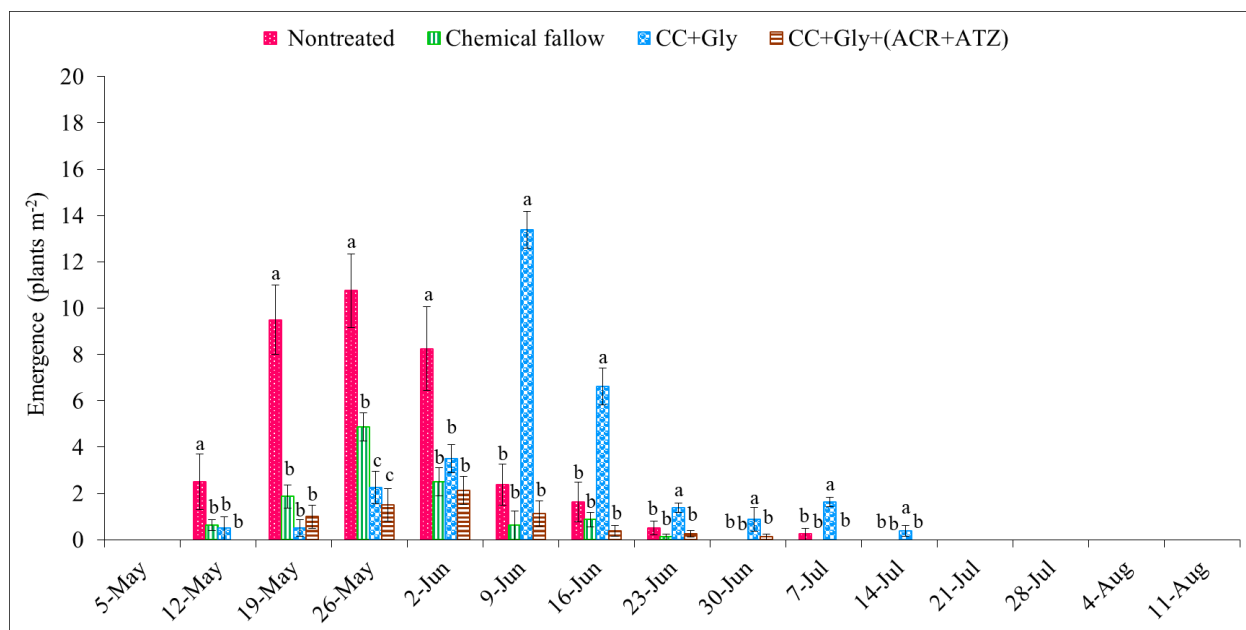


Figure 5.3. Weekly emergence counts of glyphosate-resistant *Amaranthus palmeri* under different treatments in 2022. CC+Gly indicates cover crop terminated with glyphosate; CC+Gly+(ACR+ATZ) indicates cover crop terminated with glyphosate plus acetochlor/atrazine. Means followed by the same letter within each timing are not significantly different according to Fisher's protected LSD test at $P < 0.05$. Error bars represent standard error of the mean.

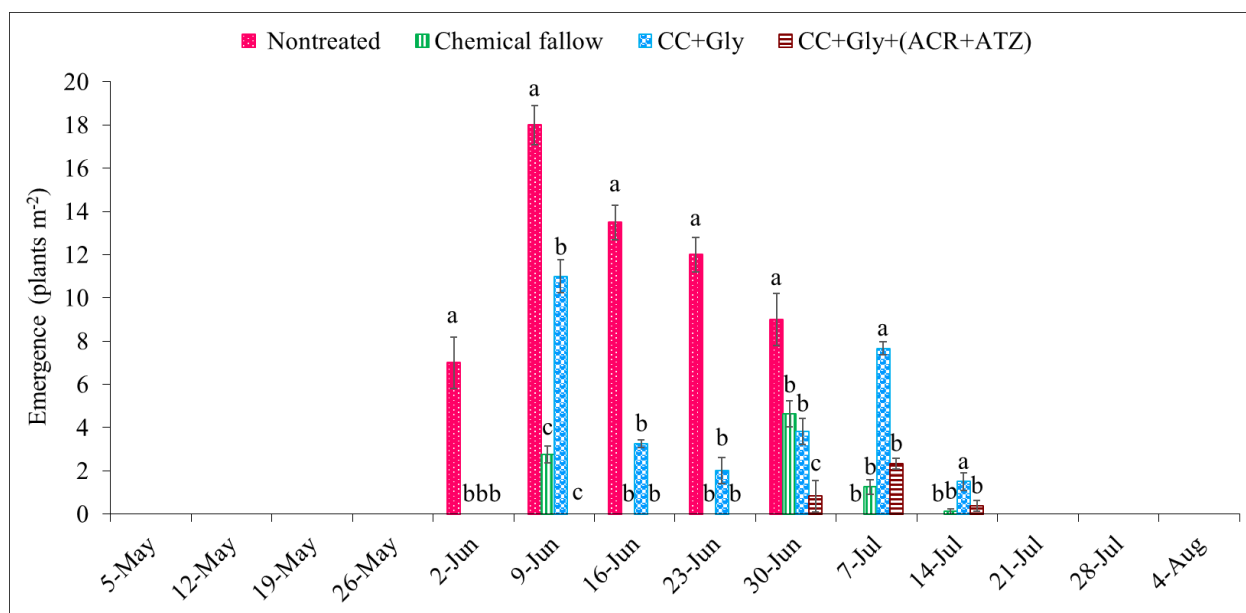


Figure 5.4. Weekly emergence counts of glyphosate-resistant *Amaranthus palmeri* under different treatments in 2023. CC+Gly indicates cover crop terminated with glyphosate; CC+Gly+(ACR+ATZ) indicates cover crop terminated with glyphosate plus acetochlor/atrazine. Means followed by the same letter within each timing are not significantly different according to Fisher's protected LSD test at $P < 0.05$. Error bars represent standard error of the mean.

Table 5.1. Planting and termination dates for cover crops and planting and harvesting dates for grain sorghum over three growing seasons at Kansas State University Agricultural Research Center near Hays, KS.

Growing season	Cover crop		Grain sorghum	
	Planting date	Termination date	Planting date	Harvesting date
2020-21	Sep 28, 2020	May 13, 2021	June 9, 2021	Nov 4, 2021
2021-22	Oct 7, 2021	May 11, 2022	June 2, 2022	Oct 26, 2022
2022-23	Sep 30, 2022	May 22, 2023	June 15, 2023	Oct 19, 2023

Table 5.2. Regression parameters estimated from the log-logistic model (Equation 5.3) for percent cumulative emergence of glyphosate-resistant (GR) *Amaranthus palmeri* and the duration of *A. palmeri* emergence in 2021.

Treatment*	Regression parameters								Duration of emergence (E_{90} - E_{10})
	b (SE)	E_{10} (SE)		E_{50} (SE)		E_{90} (SE)			
	GDD								
Chemical fallow	-2.9 (0.5)	230 (46)	b	480 (45)	c	1000 (100)	c	770	
CC+Gly	-2.5 (0.2)	286 (45)	b	685 (50)	b	1643 (142)	b	1357	
CC+Gly+(ACR+ATZ)	-2.9 (0.3)	498 (74)	a	1060 (66)	a	2257 (167)	a	1759	

* CC+Gly indicates cover crop terminated with glyphosate; CC+Gly+(ACR+ATZ) indicates cover crop terminated with glyphosate plus acetochlor/atrazine;
 b is the slope at the inflection point of each curve; E_{10} , E_{50} , and E_{90} are cumulative GDD required for 10%, 50%, and 90% cumulative emergence of GR *A. palmeri*, respectively; SE is the standard error of mean. E_{10} , E_{50} , and E_{90} estimates followed by the same letter are not different based on an approximate t-test using the “CompParm” and “EDcomp” functions in the drc package in R software (Ritz et al. 2015)

Table 5.3. Regression parameters estimated from the log-logistic model (Equation 5.3) for percent cumulative emergence of glyphosate-resistant (GR) *Amaranthus palmeri* and the duration of *A. palmeri* emergence in 2022.

Treatment*	Regression parameters								Duration of emergence (E_{90} - E_{10})
	b (SE)	E_{10} (SE)		E_{50} (SE)		E_{90} (SE)			
GDD									
Nontreated	-2.4 (0.5)	27 (8)	d	67 (8)	d	168 (24)	d	141	
Chemical fallow	-2.3 (0.2)	47 (6)	c	118 (8)	c	296 (31)	c	249	
CC+Gly	-2.5 (0.2)	70 (9)	b	170 (11)	b	409 (36)	b	339	
CC+Gly+(ACR+ATZ)	-2.8 (0.3)	116 (16)	a	254 (17)	a	557 (37)	a	441	

* CC+Gly indicates cover crop terminated with glyphosate; CC+Gly+(ACR+ATZ) indicates cover crop terminated with glyphosate plus acetochlor/atrazine; b is the slope at the inflection point of each curve; E_{10} , E_{50} , and E_{90} are cumulative GDD required for 10%, 50%, and 90% cumulative emergence of GR *A. palmeri*, respectively; SE is the standard error of mean. E_{10} , E_{50} , and E_{90} estimates followed by the same letter are not different based on an approximate t-test using the “CompParm” and “EDcomp” functions in the drc package in R software (Ritz et al. 2015)

Table 5.4. Regression parameters estimated from the log-logistic model (Equation 5.3) for percent cumulative emergence of glyphosate-resistant (GR) *Amaranthus palmeri* and the duration of *A. palmeri* emergence in 2023.

Treatment*	Regression parameters							Duration of emergence (E_{90} - E_{10})
	b (SE)	E_{10} (SE)		E_{50} (SE)		E_{90} (SE)		
	GDD							
Nontreated	-2.9 (0.4)	97 (16)	d	204 (15)	c	429 (42)	c	332
Chemical fallow	-3.6 (0.6)	192 (28)	b	349 (21)	b	634 (53)	b	442
CC+Gly	-2.6 (0.3)	132 (18)	c	304 (20)	b	700 (26)	b	568
CC+Gly+(ACR+ATZ)	-8 (1.5)	427 (28)	a	562 (16)	a	739 (39)	a	312

* CC+Gly indicates cover crop terminated with glyphosate; CC+Gly+(ACR+ATZ) indicates cover crop terminated with glyphosate plus acetochlor/atrazine;
 b is the slope at the inflection point of each curve; E_{10} , E_{50} , and E_{90} are cumulative GDD required for 10%, 50%, and 90% cumulative emergence of GR *A. palmeri*, respectively; SE is the standard error of mean. E_{10} , E_{50} , and E_{90} estimates followed by the same letter are not different based on an approximate t-test using the “CompParm” and “EDcomp” functions in the drc package in R software (Ritz et al. 2015)

Chapter 6 - Effect of fall-planted cover crops on weed suppression, grain sorghum yield, and profitability in the semi-arid Central Great Plains

Abstract

Integrating cover crops (CC) in dryland crop rotations could help in controlling herbicide-resistant weeds. A field study was conducted at Kansas State University Agricultural Research Center near Hays, KS from 2020-2023 to determine the effect of fall-planted CC on weed suppression in grain sorghum, crop yield, and net returns in no-till dryland winter wheat (*Triticum aestivum* L.)-grain sorghum [*Sorghum bicolor* (L.) Moench]-fallow (W-S-F) rotation. The field site had a natural seedbank of glyphosate-resistant (GR) kochia [*Bassia scoparia* (L.) A. J. Scott] and Palmer amaranth (*Amaranthus palmeri* S. Watson). A CC mixture (triticale/winter peas/radish/rapeseed) was planted after wheat harvest and terminated at triticale heading stage before sorghum planting. Treatments included nontreated control, chemical fallow, CC terminated with glyphosate (GLY), and CC terminated with GLY+ acetochlor/atrazine (ACR/ATZ). Across three years, CC terminated with GLY+ACR/ATZ reduced total weed density by 34-81% and total weed biomass by 45-73% compared to chemical fallow during sorghum growing season. Average grain sorghum yield was 786 to 1432 kg ha⁻¹ and did not differ between chemical fallow and CC terminated with GLY+ACR/ATZ. However, net returns were lower with both CC treatments (USD -\$275 to \$66 ha⁻¹) in all three years than chemical fallow (USD -\$111 to \$120 ha⁻¹). These results suggest that fallow replacement with fall-planted CC in the W-S-F rotation can help suppress GR kochia and Palmer amaranth in the subsequent

grain sorghum. This practice was not profitable most years but could be profitable during wet years with average yields (3800 to 5000 kg ha⁻¹) of the region.

Keywords: Kochia, Palmer amaranth, central Great Plains, residual herbicide

Introduction

The Central Great Plains (CGP) is characterized by a semi-arid climate with relatively low annual precipitation (~300 to ~1200 mm) (Lenssen et al. 2007; NOAA 2024). To conserve the soil water received from limited annual precipitation and to prevent soil erosion by wind, the no-tillage (NT-) and fallow-based cropping systems were widely adopted in the region. Successful adoption of these soil conservation practices was achieved with chemical-based weed control (Hansen et al. 2012; Kumar et al. 2020). However, adoption of NT-based production systems has resulted in weed species shifts towards more small-seeded weeds like kochia [*Bassia scoparia* (L.) A.J. Scott], Palmer amaranth (*Amaranthus palmeri* S. Watson), horseweed [*Erigeron canadensis* (L.) Cronquist], common lambsquarters (*Chenopodium album* L.), Russian thistle (*Salsola tragus* L.), downy brome (*Bromus tectorum* L.), wild oat (*Avena fatua* L.), foxtail species (*Setaria* spp.), and tumble windmill grass (*Chloris verticillata* Nutt.) (Jha et al. 2016; Nichols et al. 2015).

Winter wheat (*Triticum aestivum* L.)-grain sorghum [*Sorghum bicolor* (L.) Moench]-fallow (W-S-F) is a dominant crop rotation in the CGP region (Holman et al. 2022). This 3-year crop rotation includes a fallow period of about 10 months between winter wheat harvest and sorghum planting as well as 10 months of fallow period between sorghum harvest and the next winter wheat planting (Kumar et al. 2020). Continuous reliance on herbicides with the same site(s) of action (SOA) for weed control during these fallow periods has resulted in the evolution of herbicide-resistant weeds, including kochia, Palmer amaranth, and horseweed (Heap 2024). For instance, glyphosate resistance is widespread among kochia and Palmer amaranth populations in Kansas and other neighboring states in the CGP region (Heap 2024; Kumar et al. 2019a, 2019b, 2020; Westra et al. 2019). Evolution of these glyphosate-resistant (GR) weed populations and

limited availability of alternative effective herbicide options pose a serious production challenge for grain sorghum producers in the region. Previous researchers have reported that season-long weed interference can result in an average grain yield loss of 47% in grain sorghum, which is an estimated loss of around US \$953 million annually (Dille et al. 2020). Therefore, alternative integrated weed management (IWM) strategies are needed to achieve effective control of herbicide-resistant weed populations in grain sorghum.

Integration of cover crop (CC) in crop rotations has been proven as one of the effective tools to suppress herbicide-resistant weeds in the CGP region (Kumar et al. 2020; Mesbah et al. 2019; Obour et al. 2022a; Petrosino et al. 2015). In addition to weed suppression, growing CC in the semiarid CGP provides several benefits, including reduced soil erosion, enhanced nutrient cycling, increased microbial activity, improved soil health, and increased plant diversity and pollinator resources (Blanco-Canqui et al. 2011, 2013; Simon et al. 2022). The CC residue on the soil surface could reduce the soil temperature in summer and reduce evaporation, thereby contributing to increased soil water storage (Holman et al. 2020, 2021). However, replacing the fallow period with CC in the semiarid cropping systems sometimes reduces the yield of successive crops because of the reduced plant available water (Holman et al. 2018; Nielsen et al. 2016). However, the United States Department of Agriculture Natural Resources Conservation Service (USDA NRCS) provides some financial support to growers under the Environmental Quality Incentives Program (EQIP) to cover some of the cost of growing cover crops and improve the net returns (USDA NRCS 2024). Previous studies have evaluated the effect of spring-planted CC on weed suppression and winter wheat yields when CCs replaced the fallow phase of W-S-F rotation in this region (Holman et al. 2022; Mesbah et al. 2019; Obour et al. 2022a). For instance, Obour et al. (2022a) reported that spring-planted CC (oats/triticale/spring

peas) in W-S-F rotation can reduce weed biomass by 86 to 99% compared to weedy fallow. Holman et al. (2022) reported that spring-planted CC had no significant effect on wheat and grain sorghum yields when conditions were either extremely dry with poor yields or very wet with above-average yields, however, replacing fallow with CC increased the cost of production by 16 to 97% compared to fallow.

Several researchers have documented the importance of residual herbicides in combination with CC to achieve season-long weed control (Perkins et al. 2021; Whalen et al. 2020). For instance, Whalen et al. (2020) reported that CC terminated with glyphosate plus 2,4-D in combination with residual herbicides (sulfentrazone plus chlorimuron) resulted in greater waterhemp [*Amaranthus tuberculatus* (Moq.) J. D. Sauer.] control (73 to 84%) compared to no residual herbicide (44 to 65%). Teasdale et al. (2005) reported that hairy vetch (*Vicia villosa* Roth.) residue (500 g m⁻²) and metolachlor (10 g ha⁻¹) had a synergistic effect and reduced smooth pigweed (*Amaranthus hybridus* L.) emergence by 86% compared to no metolachlor or no hairy vetch residue. Most of these research studies were conducted in cotton (*Gossypium hirsutum* L.), corn (*Zea mays* L.), or soybean [*Glycine max* (L.) Merr.] in greater precipitation environments. However, limited information exists regarding the integration of fall-planted CC in combination with soil residual herbicides at the termination of CC on weed suppression in subsequent grain sorghum in the semiarid CGP region. The main objectives of this study were to determine the effect of fall-planted CC in combination with soil residual herbicides on (1) weed suppression (density and biomass) in subsequent grain sorghum and grain yield, and (2) net returns with integrating CC in the no-till dryland W-S-F cropping system.

Materials and Methods

Study Design and Treatments

A field study was conducted at Kansas State University Agricultural Research Center near Hays (KSU-ARC), KS (38.85196°N, 99.34279°W) during 2020-2021, 2021-2022, and 2022-2023 growing seasons. This is a long-term study started in the fall of 2020. The soil type at the experimental site was a Roxbury silt loam with a pH of 6.9 and organic matter of 1.6%. The study site was under no-till W-S-F rotation for >10 years prior to study initiation and had a natural uniform seedbank of GR kochia and Palmer amaranth (Kumar, personal observations). All three phases of the W-S-F crop rotation were present each year. A CC mixture of winter triticale ($\times$ Triticosecale Wittm.) (60%)/winter peas (*Pisum sativum* L.) (30%)/rapeseed (*Brassica napus* L.) (5%)/radish (*Raphanus sativus* L.) (5%) was drilled in wheat stubble at a rate of 67 kg ha⁻¹ during each fall and terminated in the following spring at the triticale heading stage. The CC planting dates were Sep 28, Oct 7, and Sep 30 in 2020, 2021, and 2022, respectively. The CC was terminated on May 13, May 11, and May 22 in 2021, 2022, and 2023, respectively. During each spring, four treatments were established: (1) weedy fallow, (2) chemical fallow, (3) CC terminated with glyphosate (GLY) alone, and (4) CC terminated with GLY + residual herbicide. In weedy fallow, no CC was planted and no herbicides were applied to control weeds. In chemical fallow, no CC was planted but the plot area was treated with GLY (Roundup PowerMax, Bayer Crop Science) at 1260 g ae ha⁻¹ plus a premix of acetochlor/atrazine (ACR/ATZ) (Degree Xtra, Bayer Crop Science) at 1665/826 g ai ha⁻¹ plus dicamba (Clarity, BASF) at 560 g ae ha⁻¹ at the same time as CC termination. For CC termination, GLY at 1260 g ae ha⁻¹ was used and the residual herbicide was a premix of ACR/ATZ at 1665/826 g ai ha⁻¹. All treatments were established in a randomized complete block design with four replications.

During 2020-2021 experimental year, the weedy fallow was not present and there were only three treatments with individual plot sizes of 45-m long and 13-m wide each year. During 2021-2022 and 2022-2023 experimental years, the initial chemical fallow plot was sub-divided into two to have both weedy fallow and chemical fallow treatments (45-m long and 6.5-m wide each) for comparison of weed suppression. A sorghum hybrid ‘DKS 38-16’ was planted at a seeding rate of 114,855 seeds ha⁻¹ in rows spaced 76-cm apart on June 9, June 2, and June 15 during 2021, 2022, and 2023, respectively. All local agronomic practices for grain sorghum production as recommended by Kansas State University were followed (Ciampitti et al. 2022a). Grain sorghum was harvested on Nov 4, Oct 26, and Oct 19 during 2021, 2022, and 2023, respectively. Data on monthly precipitation and air temperature over the 3-year study period were obtained from the Kansas State University Mesonet weather station (<https://mesonet.k-state.edu>) located approximately 400 m from the study site (38.8495°N, 99.3446°W) (Figure 1).

Cover Crop Biomass, Weed Density, and Weed Biomass

Each year, the aboveground shoot biomass of CC was manually harvested from two 1-m² quadrats from each plot just before CC termination and oven-dried at 72 C for 4 days to obtain dry biomass. Weed density by species (number of emerged seedlings for each species) was recorded from two randomly placed 1-m² quadrats from each plot at CC termination and at monthly intervals until sorghum harvest (except 2021, where data at 90 days after CC termination was not collected) and aboveground weed biomass was manually harvested and oven-dried at 72 C for 4 days to obtain total weed dry biomass. The average of CC biomass, total weed density, and total weed dry biomass from the two quadrats in each plot at each time were used in the data analysis. The weed species composition was characterized by calculating the relative abundance of each species in each plot using the method described by Thomas (1985)

and used by Obour et al. (2022a). Relative abundance was determined using equation 6.1.

Relative density and relative frequency were calculated using equations 6.2 and 6.3, respectively.

$$\text{Relative abundance} = \frac{\text{Relative density} + \text{Relative frequency}}{2} \quad \text{Eqn. 6.1}$$

Relative density

$$= \left[\frac{\text{Number of plants for each species within the quadrat per plot}}{\text{Total number of plants in that sampled quadrat}} \right] \times 100 \quad \text{Eqn. 6.2}$$

Relative frequency

$$= \left[\frac{\text{Proportion of quadrats in which the species was present in a plot}}{\text{Frequency of all species in that sampled quadrat}} \right] \times 100 \quad \text{Eqn. 6.3}$$

Volumetric Water Content and Grain Sorghum Yield

The CC effect on soil water content at grain sorghum planting was determined gravimetrically in 30 cm increments up to 150 cm depth. Two soil cores were taken from each plot using a hydraulic probe (Giddings Machine Company) in June 2021 and 2023 before grain sorghum planting. Soil samples were not collected in 2022. Portions of the soil samples from each 30-cm depth were weighed fresh and then dried at 105 C for 4 days to calculate bulk density by dividing the mass of oven-dry soil to the volume of the core. Gravimetric water content was calculated using equation 6.4.

$$\text{Gravimetric water content} = \left[\frac{\text{Wet soil weight} - \text{Dry soil weight}}{\text{Dry soil weight}} \right] \quad \text{Eqn. 6.4}$$

The data from both soil cores were averaged to obtain a single soil water measurement which was converted to volumetric water content by multiplying with measured bulk density at each sampling depth. Data for volumetric water content were averaged for both CC treatments as both treatments were the same before termination. The grain sorghum yield was recorded by harvesting grain from each whole plot using a Massey Ferguson 8XP small plot combine harvester (Massey Ferguson) and yield was adjusted to 13.5% moisture content.

Economic Analyses

Gross returns were calculated by multiplying the grain sorghum yield and the price of sorghum grain. Net returns were calculated as gross returns minus total variable costs for each treatment for each year. Fixed costs were ignored in this analysis as they were assumed to be consistent across treatments. Four-year average custom rate values published by Kansas State University Land Use Survey Program and the Kansas Department of Agriculture (AgManager 2022) were used for current field operations and input costs. Total variable cost was calculated by adding all the expenses for planting (CC and sorghum), inputs (fertilizer, herbicides, etc., and their application costs), and harvesting. Grain sorghum price for each experimental year was taken from the USDA Economic Research Services market reports (USDA ERS 2024). Prices for grain sorghum were calculated on a per kg basis and ranged from \$0.20 to \$0.24. All costs and revenue were calculated in U.S. dollars per hectare.

Statistical Analyses

Data were tested for homogeneity of variance and normality of the residuals using the PROC UNIVARIATE procedure in SAS 9.3 (SAS Institute, Inc., SAS Campus Drive, Cary, NC). Data for total weed density and total dry biomass were log-transformed to improve the normality of the residuals and homogeneity of variance; however, back-transformed data were

presented with mean separation based on the transformed data, whereas the rest of the data met both ANOVA assumptions. All data for CC biomass, total weed density, and total weed dry biomass at each time, volumetric water content, grain yield, and net returns were subjected to ANOVA using PROC MIXED procedure. For CC biomass data, year was considered as fixed effect and replication was considered as random effect. For total weed density and total weed dry biomass data, CC treatment, year, monthly timing, and their interactions were considered as fixed effects whereas replication and their interactions were considered as random effects. For volumetric water content, CC treatment, year, soil depth, and their interactions were considered as fixed effects whereas replication and their interactions were considered as random effects. For data on grain sorghum yield and net returns, CC treatment, year, and their interactions were considered as fixed effects whereas replication and their interactions were considered as random effects. Data for total weed density, total weed dry biomass, volumetric water content, grain sorghum yield, and net returns were analyzed separately for each year because of significant year-by- treatment interaction ($P < 0.01$). Treatment-by-monthly evaluation interaction for total weed density and total weed dry biomass was significant ($P < 0.001$); therefore, data were sorted by monthly evaluation timings using PROC SORT. Treatment means were separated using Fisher's protected LSD test ($P < 0.05$). The grain sorghum yields were low because of the drought conditions during the study period; therefore, a sensitivity analysis was conducted to obtain net returns to possible grain sorghum yield (700 to 7400 kg ha⁻¹) and prices (\$0.09 to \$0.24 kg⁻¹) in the region.

Results and Discussion

Variable precipitation amount and frequency were observed at KSU-ARCH during the experimental periods 2020-2021, 2021-2022 and 2022-2023 (Figure 6.1). The total amount of

precipitation received during the CC growing season (Sept. to May) in 2020-2021, 2021-2022 and 2022-2023 was 217, 99, and 130 mm, respectively (Figure 6.1). No difference was recorded in aboveground CC dry biomass at the time of termination across the years and was 1520, 1130, and 1470 kg ha⁻¹ in 2021, 2022, and 2023, respectively, with an average of 1370 ± 123 kg ha⁻¹. During the grain sorghum growing season (June through Oct), the total precipitation amount was 256, 171, and 237 mm in 2021, 2022, and 2023, respectively (Figure 6.1).

Total Weed Density and Weed Dry Biomass

Across three years, four broadleaf weed species were observed, including kochia, Palmer amaranth, Venice mallow (*Hibiscus trionum* L.), and puncturevine (*Tribulus terrestris* L.). Based on the relative abundance, kochia and Palmer amaranth were the dominant weed species across three years (Tables 6.1 to 6.3).

2021 Growing Season

Palmer amaranth was the most dominant weed species with a mean relative abundance of >50% across treatments at all monthly evaluation timings (Table 6.1). Prior to termination, CC reduced the total weed density by 86 to 95% compared to chemical fallow (no herbicide was applied in chemical fallow at this time) in 2021 (Table 6.1). However, an application of GLY plus ACR/ATZ plus dicamba in chemical fallow at the time of CC termination reduced weed density at later evaluation timings. Total weed density at 60 days after termination (DAT) was dominated by Palmer amaranth (relative abundance = 90%) and was significantly greater (approximately 4 times) under the CC terminated with GLY only compared to the CC terminated with GLY plus ACR/ATZ (Table 6.1). This might be due to a lack of residual herbicide and CC residue was not enough to suppress the emerging Palmer amaranth seedlings. At the time of

grain sorghum harvest (120 DAT), CC terminated with GLY plus ACR/ATZ reduced the total weed density by 50% compared to chemical fallow (Table 6.1).

Consistent with total weed density, the total weed dry biomass at CC termination was reduced by 93% compared to chemical fallow (Table 6.4). The CC terminated with GLY only and with GLY plus ACR/ATZ reduced the total weed dry biomass by 50 to 65% and 42 to 68% at 60 to 120 DAT, respectively, compared to chemical fallow. It is interesting to note that no differences were observed in total weed density at 120 DAT between chemical fallow and CC terminated with GLY only; however, the same CC treatment significantly reduced the total weed dry biomass by 42% compared to chemical fallow, indicating the suppressive effect of CC on weed growth that could ultimately reduce the weed seed production.

2022 Growing Season

Mean relative abundance was 46 to 56% for kochia, 0 to 21% for Palmer amaranth, and 29 to 49% for Venice mallow before CC termination (Table 6.2). Similar to 2021, the weed density at CC termination was reduced by 90 to 93% compared to weedy fallow. At 30 DAT, the CC terminated with GLY plus ACR/ATZ had less total weed density by 92% compared to chemical fallow and 96% compared to weedy fallow. At 60 DAT, the mean relative abundance was 41 to 75% for kochia, 11 to 53% for Palmer amaranth, and 12 to 31% for Venice mallow among all treatments. The CC termination with GLY only treatment did not produce enough CC biomass to suppress emerging weed seedlings and had 21 more weed seedlings m⁻² than the CC terminated with GLY plus ACR/ATZ at 60 DAT (Table 6.2). These results indicate the importance of residual herbicide with CC to achieve effective weed suppression. Our findings are consistent with Wiggins et al. (2016), who also concluded that CC alone was not enough for season-long control of GR Palmer amaranth and suggested integrating residual herbicides to

complement the suppressive effect of CC. In the current study, total weed density did not differ between chemical fallow and CC terminated with GLY plus ACR/ATZ treatments at 120 DAT; however, weed density was nearly 95% lower in CC terminated with GLY plus ACR/ATZ compared to weedy fallow and CC terminated with GLY only (Table 6.2). These results are consistent with Obour et al. (2022a), who previously reported 82% reduction in total weed density with spring-planted CC mixture (oat/triticale/pea) compared to weedy fallow. Petrosino et al. (2015) also reported a 78 to 94% reduction in kochia density with fall-planted CC (triticale/triticale-hairy vetch mixture) compared to chemical fallow in winter wheat-fallow rotation. Similar to weed density reduction, CC at the time of termination provided >95% total weed dry biomass reduction compared to weedy fallow (Table 6.4). The presence of CC reduces the sunlight penetration to the ground for weed seed germination and also reduces their competitive ability for other resources, thereby resulting in lower weed biomass (Silva and Bagavathiannan 2023; Webster et al. 2016). The CC terminated with GLY only reduced total weed biomass by 94% at 30 DAT and 63% at 120 DAT compared to weedy fallow. In contrast, the CC terminated with GLY plus ACR/ATZ reduced total weed biomass by >95% as compared to weedy fallow throughout the grain sorghum growing season (Table 6.4). These results indicate the need of residual herbicide in combination with CC for a season-long weed control in grain sorghum. These results are consistent with Whalen et al. (2020), who reported that fall-planted CC, including Italian ryegrass [*Lolium perenne* L. ssp. *multiflorum* (Lam.) Husnot], oat (*Avena sativa* L.), and winter wheat provided 38 to 48% weed control without a residual herbicide; however, the control ranged from 72 to 85% under CC with a residual herbicide (sulfentrazone plus chlorimuron) application.

2023 Growing Season

No weed emergence was observed under both CC treatments at the time of termination (Table 6.3). In addition, no weed emergence was observed in the CC terminated with GLY plus ACR/ATZ treatment at 30 DAT. In contrast, a greater weed density of 91 plants m⁻² with a relative abundance of 70% for Palmer amaranth and 30% for kochia was recorded in the CC terminated with GLY only treatment at 30 DAT (Table 6.3). This increase in Palmer amaranth and kochia densities under CC terminated with GLY only was probably due to more precipitation within 30 DAT (78 mm, 33% of total precipitation received during the entire sorghum growing season), and lack of any residual herbicide applied at CC termination. Treatments, including the CC terminated with GLY plus ACR/ATZ and chemical fallow reduced the total weed density by 70% compared to weedy fallow at 90 DAT. Chemical fallow and CC terminated with GLY plus ACR/ATZ had much fewer weeds (7 to 8 plants m⁻²) compared to weedy fallow (54 plants m⁻² with 73% relative abundance of Palmer amaranth) at 120 DAT (Table 6.3). Similar to weed density reduction, CC terminated with GLY plus ACR/ATZ resulted in 98%, 46%, and 57% total weed biomass reduction compared to weedy fallow, chemical fallow, and CC terminated with GLY only, respectively, at 60 DAT (Table 6.4). At 120 DAT, the CC terminated with GLY plus ACR/ATZ and chemical fallow reduced the total weed biomass by 83% and 65% compared to weedy fallow. Our results are consistent with Petrosino et al. (2015), who also reported that fall-planted triticale and a triticale-hairy vetch mixture reduced kochia density by 78% and 94%, respectively, and biomass up to 98% compared to chemical fallow. Compared to chemical fallow, the CC terminated with GLY plus ACR/ATZ provided 5 to 18% more reductions in total weed dry biomass during the entire grain sorghum growing season (Table 6.4). Wiggins et al. (2016) reported that CC including cereal rye, winter vetch,

crimson clover, or winter wheat had less than 65% control of Palmer amaranth, whereas the same CC in combination with PRE-applied acetochlor or fluometuron resulted in >87% control of Palmer amaranth.

Volumetric Water Content and Grain Sorghum Yield

The soil water content at sorghum planting is directly related to grain yields in the dryland region (Holman et al. 2023; Obour et al. 2022b). In 2021, fall-planted CC did not affect volumetric water content at grain sorghum planting compared to chemical fallow across all soil depths (Figure 6.2). This might be due to greater precipitation at pre- and post-CC termination time (Figure 6.1). The precipitation received closer to termination or post-termination of CC may have recharged the soil profile thereby diminishing the effects of the growing CC on water use. The additional CC residue might have also helped decrease soil water evaporation, increasing moisture storage (Holman et al. 2020, 2021). In contrast, fall-planted CC did reduce the volumetric water content at grain sorghum planting compared to chemical fallow at 0-30 cm and 30-60 cm depths in the 2023 growing season (Figure 6.2). No differences in the water content were observed between CC and chemical fallow treatments at deeper soil depths. Similar to CC, the weedy fallow treatment also had relatively low volumetric water content (0.16 to $0.20\text{ cm}^3\text{ cm}^{-3}$) up to 60 cm soil depth compared to chemical fallow (0.20 to $0.22\text{ cm}^3\text{ cm}^{-3}$) indicating the water use by weeds. Holman et al. (2021) reported no difference in the available soil moisture at time of wheat planting between fallow and spring-planted CC left standing during the fallow phase of W-S-F rotation.

No differences in grain sorghum yields were observed between chemical fallow (1876 kg ha^{-1}) and CC terminated with GLY plus ACR/ATZ (2072 kg ha^{-1}) in 2021 and the least grain yield (1456 kg ha^{-1}) was recorded under CC terminated with GLY only (Figure 6.3). During the

2022 growing season, the overall grain sorghum yield was low due to lower season precipitation (171 mm) compared to 2021 (256 mm). Effective weed suppression (both density and total weed dry biomass) achieved with the CC terminated with GLY plus ACR/ATZ resulted in a greater grain sorghum yield (1319 kg ha^{-1}) compared to chemical fallow (912 kg ha^{-1}). The CC termination with GLY only was able to suppress weeds up to 30 DAT (Table 6.2) and Palmer amaranth emerged after 30 DAT reduced the grain sorghum yield (472 kg ha^{-1}) (Figure 6.3). The total precipitation during the 2023 sorghum growing season was 237 mm, but the majority of this precipitation occurred in May and June. There was moisture stress at the boot stage of grain sorghum in September (only 17 mm of rainfall) that resulted in reduced grain yield (432 to 1323 kg ha^{-1}) (Figures 6.1 and 6.3). No difference in grain sorghum yield was observed between chemical fallow and CC terminated with GLY plus ACR/ATZ. The precipitation events in May and June resulted in the emergence of several cohorts of Palmer amaranth under the CC terminated with GLY only that resulted in competition with grain sorghum and reduced yield (432 kg ha^{-1}) compared to chemical fallow and CC terminated with GLY plus ACR/ATZ (Figure 6.3). Based on the precipitation amount and frequency, fall-planted CC had a variable response to impact the grain sorghum yield. Nielsen et al. (2016) reported a 10% reduction in winter wheat yield following a CC compared to fallow in the W-S-F rotation under the semiarid CGP. Holman et al. (2022) reported that spring-planted CC (oat grain) after sorghum harvesting in the fallow phase of W-S-F rotation resulted in 29% less available soil moisture at sorghum planting and did not affect the wheat and sorghum yield compared to fallow. In south-central Kansas, Janke et al. (2002) reported no differences in grain sorghum yield following CC (hairy vetch/winter pea) in winter wheat-grain sorghum rotation compared to no CC in years with good rainfall; however, during years with low rainfall, CC establishment was poor, and grain sorghum

yield was reduced because of water use by the CC compared to no CC treatment. Eash et al. (2021) also reported that CC had no impact on subsequent crop yields in a very low-yielding environment in Colorado, mainly due to low CC biomass.

Economic Analyses

During 2021, the CC terminated with GLY plus ACR/ATZ resulted in greater gross returns of \$497 ha⁻¹ compared to \$450 ha⁻¹ under chemical fallow (Table 6.5). This was because of better weed control and greater sorghum yield with CC terminated with GLY plus ACR/ATZ compared to chemical fallow. However, CC seed and planting costs decreased net returns. Chemical fallow had more net returns by \$55 ha⁻¹ compared to CC terminated with GLY plus ACR/ATZ, although both treatments did not differ for grain yields. Net returns were negative under CC terminated with GLY only because of lower grain sorghum yield than chemical fallow and CC terminated with GLY plus ACR/ATZ.

Similar to 2021, CC terminated with GLY plus ACR/ATZ had greater gross returns (\$317 ha⁻¹) followed by chemical fallow (\$219 ha⁻¹) and CC terminated with GLY only (\$113 ha⁻¹) in 2022 (Table 6.6). Weedy fallow had the lowest gross returns of \$24 ha⁻¹. The net returns for 2022 were negative for all treatments because of low grain sorghum yields, which suggests that gross return from grain sorghum was not enough to cover the variable input costs. However, net returns for CC terminated with GLY plus ACR/ATZ and chemical fallow were not different (Table 6.6).

In 2023, the lower grain sorghum yield in CC terminated with GLY plus ACR/ATZ resulted in a lower gross return of \$181 ha⁻¹ compared to \$265 ha⁻¹ in chemical fallow (Table 6.7). Similar to 2022, the net returns in 2023 were also negative for all treatments. Chemical fallow had less negative net returns (-\$65 ha⁻¹) compared to other treatments (-\$138 to -\$275 ha⁻¹).

¹). The cost of CC seed and planting increased the variable cost for both CC treatments and thus resulted in more negative net returns. Janke et al. (2002) also reported lower net returns with fall-planted CC (hairy vetch/winter pea) before grain sorghum because of lower grain sorghum yield in the years with low rainfall. Obour et al. (2022a) also reported that integrating CC in the dryland cropping system resulted in negative net returns compared to fallow. These results indicate that growing CC only for weed suppression was not profitable compared to chemical fallow.

Based on the sensitivity analysis, a minimum grain sorghum yield of 2000 kg ha⁻¹ was needed to obtain a positive net return (\$8 ha⁻¹) with CC terminated with GLY plus ACR/ATZ at average grain sorghum price (\$0.22 kg⁻¹) during the 3-year study period (Table 6.8). In the present study, the average grain sorghum yield was low (786 to 1432 kg ha⁻¹). The estimated average grain sorghum yield in western Kansas typically ranges from 3800 to 5000 kg ha⁻¹ (Ciampitti and Carcedo, 2022); therefore, based on this yield scenario, the expected net returns would be \$404 to \$668 ha⁻¹ with CC terminated with GLY plus ACR/ATZ and \$506 to \$770 ha⁻¹ with chemical fallow at a grain sorghum price of \$0.22 kg⁻¹ (Table 6.8). At the lowest grain sorghum price (\$0.09 ha⁻¹), a grain yield of 5000 kg ha⁻¹ was expected to cover the cost of CC seed and planting and obtain positive net returns (\$18 ha⁻¹) with CC terminated with GLY plus ACR/ATZ. However, with chemical fallow, a lower yield of 3800 kg ha⁻¹ was sufficient to obtain positive net returns (\$12 ha⁻¹) (Table 6.8). It is important to note that in the present study, greater weed control was observed under CC terminated with GLY plus ACR/ATZ over the grain sorghum growing season compared to chemical fallow. POST herbicides were not applied in chemical fallow treatment but farmers generally apply POST herbicides to control later emerged weeds, which could increase the cost of production and decrease the net returns with

chemical fallow as compared to CC treatment. At the highest sorghum price (\$0.24 ha⁻¹), CC terminated with GLY plus ACR/ATZ would result in \$48 ha⁻¹ net returns at only 2000 kg ha⁻¹ sorghum yield (Table 6.8). These results indicate the cost of integrating fall-planted CC in the W-S-F exceeded the benefits of improved weed control.

Conclusions

Results from this 3-year study indicate that integrating a fall-planted CC mixture after winter wheat harvest and terminating it with GLY in combination with residual herbicide before grain sorghum planting in the W-S-F rotation can effectively suppress weeds in grain sorghum. Fall-planted CC terminated with GLY plus ACR/ATZ reduced total weed density by 34 to 81% and total weed biomass by 45 to 73% compared to chemical fallow over the grain sorghum growing season. No yield penalty was observed after CC terminated with GLY plus ACR/ATZ. However, due to costs of CC seed and planting, lower net returns were recorded in all three years compared to chemical fallow. These results suggest that growing CC for only weed suppression in the semiarid CGP would not be profitable. If the CC were used for hay or forage then net returns could be increased (Holman et al. 2018, 2021, 2022, 2023; Obour et al. 2022a). However, weed control during the sorghum growing season may be affected by the removal of CC residue from the field. Therefore, future studies should focus on understanding the timing for the removal of CC residue and its interaction with weed control during the grain sorghum growing period.

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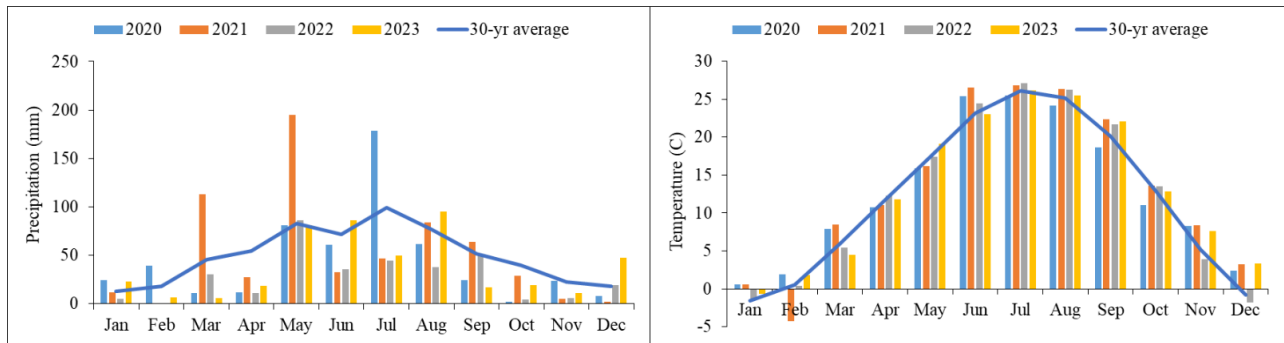


Figure 6.1. Total monthly precipitation and average monthly temperature from 2020 to 2023 growing seasons.

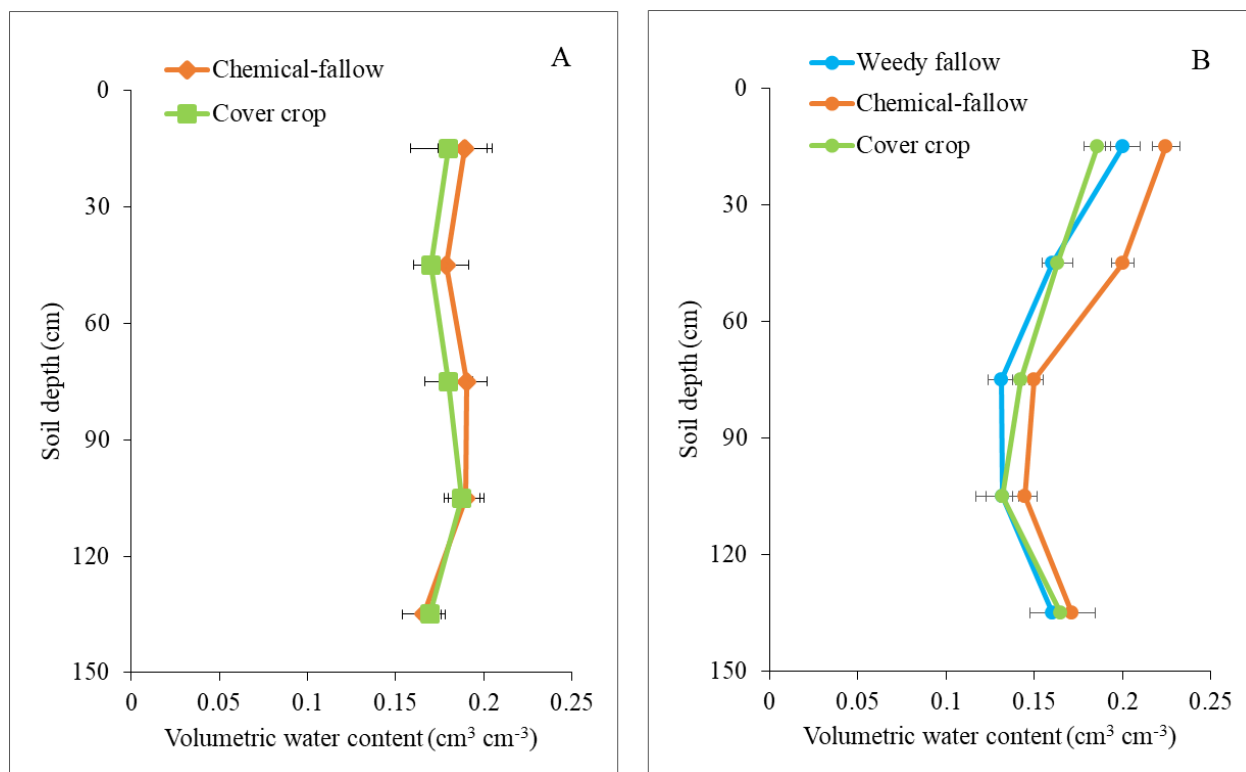


Figure 6.2. Fall-planted cover crop effect on volumetric water content at grain sorghum planting in 2021 (A) and 2023 (B) growing seasons. Error bars represent standard error of the mean.

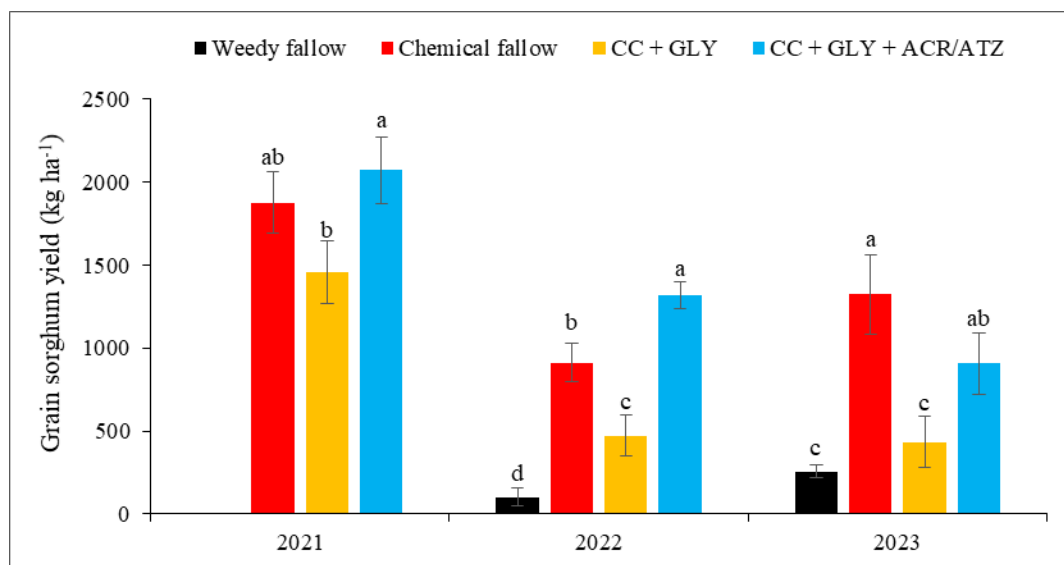


Figure 6.3. Fall-planted cover crop effect on grain sorghum yield over three growing seasons. CC + GLY indicates cover crop terminated with glyphosate only and CC + GLY + ACR/ATZ indicates cover crop terminated with glyphosate plus a premix of acetochlor/atrazine. Means followed by the same letter within the year are not significantly different according to Fisher's protected LSD test at $P < 0.05$. Error bars represent standard error of the mean.

Table 6.1. Total weed density and mean relative abundance of weed species observed in the cover crop (CC) treatments at 0 to 120 days after CC termination (DAT) in 2021.^a

Treatments	Total weed density	Mean relative abundance			
		Kochia	Palmer amaranth	Venice mallow	Puncture vine
				%	
	plants m ⁻²				
At 0 DAT					
Chemical fallow	43 a	43	41	16	0
CC + GLY	6 b	0	100	0	0
CC + GLY + ACR/ATZ	4 b	0	100	0	0
At 30 DAT					
Chemical fallow	10 a	38	50	4	8
CC + GLY	10 a	12	69	0	19
CC + GLY + ACR/ATZ	6 a	11	53	8	28
At 60 DAT					
Chemical fallow	9 a	24	70	0	6
CC + GLY	12 a	10	90	0	0
CC + GLY + ACR/ATZ	3 b	10	90	0	0
At 120 DAT					
Chemical fallow	4 a	50	50	0	0
CC + GLY	5 a	29	71	0	0
CC + GLY + ACR/ATZ	2 b	39	61	0	0

^aCC + GLY indicates cover crop terminated with glyphosate only and CC + GLY + ACR/ATZ indicates cover crop terminated with glyphosate plus a premix of acetochlor/atrazine. Means followed by the same letter within a column at each timing are not different according to Fisher's protected LSD at $P < 0.05$.

Table 6.2. Total weed density and mean relative abundance of weed species observed in the cover crop (CC) treatments at 0 to 120 days after CC termination (DAT) in 2022.^a

Treatments	Total weed density		Mean relative abundance			
			Kochia	Palmer amaranth	Venice mallow	Puncture vine
	plants m ⁻²		%			
At 0 DAT						
Weedy fallow	58	a	46	21	33	0
Chemical fallow	47	a	53	18	29	0
CC + GLY	4	b	51	0	49	0
CC + GLY + ACR/ATZ	6	b	56	6	38	0
At 30 DAT						
Weedy fallow	27	ab	56	36	6	2
Chemical fallow	13	b	62	32	6	0
CC + GLY	37	a	41	54	5	0
CC + GLY + ACR/ATZ	1	c	62	38	0	0
At 60 DAT						
Weedy fallow	28	a	58	11	31	0
Chemical fallow	6	bc	47	53	0	0
CC + GLY	22	ab	41	47	12	0
CC + GLY + ACR/ATZ	1	c	75	25	0	0
At 90 DAT						
Weedy fallow	27	a	80	20	0	0
Chemical fallow	1	b	100	0	0	0
CC + GLY	13	a	43	57	0	0
CC + GLY + ACR/ATZ	1	b	87	13	0	0
At 120 DAT						
Weedy fallow	19	a	93	7	0	0
Chemical fallow	2	b	91	9	0	0
CC + GLY	21	a	46	54	0	0
CC + GLY + ACR/ATZ	1	b	89	11	0	0

^aCC + GLY indicates cover crop terminated with glyphosate only and CC + GLY + ACR/ATZ indicates cover crop terminated with glyphosate plus a premix of acetochlor/atrazine. Means followed by the same letter within a column at each timing are not different according to Fisher's protected LSD at P < 0.05.

Table 6.3. Total weed density and mean relative abundance of weed species observed in the cover crop (CC) treatments at 0 to 120 days after CC termination (DAT) in 2023.^a

Treatments	Total weed density		Mean relative abundance			
			Kochia	Palmer amaranth	Venice mallow	Puncture vine
	plants m ⁻²		%			
At 0 DAT						
Weedy fallow	32	a	100	0	0	0
Chemical fallow	28	a	100	0	0	0
CC + GLY	0	b	0	0	0	0
CC + GLY + ACR/ATZ	0	b	0	0	0	0
At 30 DAT						
Weedy fallow	142	a	37	61	2	1
Chemical fallow	12	b	0	100	0	0
CC + GLY	91	a	30	70	0	0
CC + GLY + ACR/ATZ	0	b	0	0	0	0
At 60 DAT						
Weedy fallow	22	a	13	87	0	0
Chemical fallow	7	b	0	100	0	0
CC + GLY	15	ab	25	75	0	0
CC + GLY + ACR/ATZ	1	b	0	100	0	0
At 90 DAT						
Weedy fallow	30	a	16	84	0	0
Chemical fallow	9	b	10	90	0	0
CC + GLY	20	ab	15	85	0	0
CC + GLY + ACR/ATZ	9	b	0	100	0	0
At 120 DAT						
Weedy fallow	54	a	27	73	0	0
Chemical fallow	8	b	19	81	0	0
CC + GLY	22	ab	27	73	0	0
CC + GLY + ACR/ATZ	7	b	0	100	0	0

^aCC + GLY indicates cover crop terminated with glyphosate only and CC + GLY + ACR/ATZ indicates cover crop terminated with glyphosate plus a premix of acetochlor/atrazine. Means followed by the same letter within a column at each timing are not different according to Fisher's protected LSD at P < 0.05.

Table 6.4. Total weed biomass in the cover crop treatments at 0, 30, 60, 90, and 120 days after cover crop termination during 2021 to 2023 growing seasons.^a

Treatments	Total weed biomass																											
	2021				2022				2023																			
	0	30	60	120	0	30	60	90	120	0	30	60	90	120														
	g m ⁻²																											
Weedy fallow	-	-	-	-	34	a	107	a	118	a	193	a	263	a	12	a	115	a	388	a	1430	a	1624	a				
Chemical fallow	15	a	72	a	139	a	147	a	30	a	2	c	3	bc	6	c	12	c	9	a	5	c	13	c	345	b	560	bc
CC + GLY	1	b	41	a	69	b	85	b	1	b	6	b	15	b	41	b	97	b	0	b	44	b	167	b	513	b	1258	ab
CC + GLY + ACR/ATZ	1	b	24	a	48	b	47	b	1	b	1	c	1	c	3	c	10	c	0	b	0	d	7	d	236	c	278	c

^aCC + GLY indicates cover crop terminated with glyphosate only and CC + GLY + ACR/ATZ indicates cover crop terminated with glyphosate plus a premix of acetochlor/atrazine. Means followed by the same letter within a column are not different according to Fisher's protected LSD at $P < 0.05$.

Table 6.5. Economic analyses of grain sorghum after fall-planted cover crop in 2021 growing season.^a

Variables	Treatments		
	Chemical fallow	CC + GLY	CC + GLY + ACR/ATZ
<u>Gross returns</u>			
Grain sorghum yield, kg ha ⁻¹	1877.0	1456.0	2072.0
Grain sorghum price, \$ kg ⁻¹	0.24	0.24	0.24
Revenue from grain sorghum, \$ ha ⁻¹	450.5	349.4	497.3
Gross returns, \$ ha ⁻¹	450.5	349.4	497.3
<u>Variable input costs</u>			
Cover crop seed, \$ ha ⁻¹	0	95.7	95.7
Cover crop planting, \$ ha ⁻¹	0	37.1	37.1
Grain sorghum seed, \$ ha ⁻¹	30.4	30.4	30.4
Grain sorghum planting, \$ ha ⁻¹	32.1	32.1	32.1
Fertilizer with application cost, \$ ha ⁻¹	66.7	66.7	66.7
Herbicide, \$ ha ⁻¹	126.9	25.6	95.7
Herbicide application cost, \$ ha ⁻¹	13.6	13.6	13.6
Grain sorghum harvesting, \$ ha ⁻¹	60.3	60.3	60.3
Total variable cost, \$ ha ⁻¹	330.0	361.5	431.6
Net returns, \$ ha ⁻¹	120.5 a	-12.1 b	65.7 a

^aCC + GLY indicates cover crop terminated with glyphosate only and CC + GLY + ACR/ATZ indicates cover crop terminated with glyphosate plus a premix of acetochlor/atrazine. Means followed by the same letter among treatments are not different according to Fisher's protected LSD at $P < 0.05$.

Table 6.6. Economic analyses of grain sorghum after fall-planted cover crop in 2022 growing season.^a

Variables	Treatments			
	Weedy fallow	Chemical fallow	CC + GLY	CC + GLY + ACR/ATZ
<u>Gross returns</u>				
Grain sorghum yield, kg ha ⁻¹	101.0	912.0	472.0	1319.0
Grain sorghum price, \$ kg ⁻¹	0.24	0.24	0.24	0.24
Revenue from grain sorghum, \$ ha ⁻¹	24.2	218.9	113.3	316.6
Gross returns, \$ ha ⁻¹	24.2	218.9	113.3	316.6
<u>Variable input costs</u>				
Cover crop seed, \$ ha ⁻¹	0	0	95.7	95.7
Cover crop planting, \$ ha ⁻¹	0	0	37.1	37.1
Grain sorghum seed, \$ ha ⁻¹	30.4	30.4	30.4	30.4
Grain sorghum planting, \$ ha ⁻¹	32.1	32.1	32.1	32.1
Fertilizer with application cost, \$ ha ⁻¹	66.7	66.7	66.7	66.7
Herbicide, \$ ha ⁻¹	0	126.9	25.6	95.7
Herbicide application cost, \$ ha ⁻¹	0	13.6	13.6	13.6
Grain sorghum harvesting, \$ ha ⁻¹	60.3	60.3	60.3	60.3
Total variable cost, \$ ha ⁻¹	189.5	330.0	361.5	431.6
Net returns, \$ ha ⁻¹	-165.3 b	-111.1 a	-248.2 b	-115.0 a

^aCC + GLY indicates cover crop terminated with glyphosate only and CC + GLY + ACR/ATZ indicates cover crop terminated with glyphosate plus a premix of acetochlor/atrazine. Means followed by the same letter among treatments are not different according to Fisher's protected LSD at $P < 0.05$.

Table 6.7. Economic analyses of grain sorghum after fall-planted cover crop in 2023 growing season.^a

Variables	Treatments			
	Weedy fallow	Chemical fallow	CC + GLY	CC + GLY + ACR/ATZ
<u>Gross returns</u>				
Grain sorghum yield, kg ha ⁻¹	257.0	1323.0	432.0	905.0
Grain sorghum price, \$ kg ⁻¹	0.20	0.20	0.20	0.20
Revenue from grain sorghum, \$ ha ⁻¹	51.4	264.6	86.4	181.0
Gross returns, \$ ha ⁻¹	51.4	264.6	86.4	181.0
<u>Variable input costs</u>				
Cover crop seed, \$ ha ⁻¹	0	0	95.7	95.7
Cover crop planting, \$ ha ⁻¹	0	0	37.1	37.1
Grain sorghum seed, \$ ha ⁻¹	30.4	30.4	30.4	30.4
Grain sorghum planting, \$ ha ⁻¹	32.1	32.1	32.1	32.1
Fertilizer with application cost, \$ ha ⁻¹	66.7	66.7	66.7	66.7
Herbicide, \$ ha ⁻¹	0	126.9	25.6	95.7
Herbicide application cost, \$ ha ⁻¹	0	13.6	13.6	13.6
Grain sorghum harvesting, \$ ha ⁻¹	60.3	60.3	60.3	60.3
Total variable cost, \$ ha ⁻¹	189.5	330.0	361.5	431.6
Net returns, \$ ha ⁻¹	-138.1 b	-65.4 a	-275.1 b	-250.6 b

^aCC + GLY indicates cover crop terminated with glyphosate only and CC + GLY + ACR/ATZ indicates cover crop terminated with glyphosate plus a premix of acetochlor/atrazine. Means followed by the same letter among treatments are not different according to Fisher's protected LSD at $P < 0.05$.

Table 6.8. Net returns to possible grain sorghum yield (700 to 7400 kg ha⁻¹) and prices (\$0.09 to \$0.24 kg⁻¹) in the region.^a

Grain sorghum yield	Net returns													
	\$ ha ⁻¹													
	Chemical fallow							CC + GLY +ACR/ATZ						
	Grain sorghum price													
kg ha ⁻¹	\$ kg ⁻¹													
	0.09	0.12	0.16	0.18	0.20	0.22	0.24	0.09	0.12	0.16	0.18	0.20	0.22	0.24
700	-267	-246	-218	-204	-190	-176	-162	-369	-348	-320	-306	-292	-278	-264
1400	-204	-162	-106	-78	-50	-22	6	-306	-264	-208	-180	-152	-124	-96
2000	-150	-90	-10	30	70	110	150	-252	-192	-112	-72	-32	8	48
2600	-96	-18	86	138	190	242	294	-198	-120	-16	36	88	140	192
3200	-42	54	182	246	310	374	438	-144	-48	80	144	208	272	336
3800	12	126	278	354	430	506	582	-90	24	176	252	328	404	480
4400	66	198	374	462	550	638	726	-36	96	272	360	448	536	624
5000	120	270	470	570	670	770	870	18	168	368	468	568	668	768
5600	174	342	566	678	790	902	1014	72	240	464	576	688	800	912
6200	228	414	662	786	910	1034	1158	126	312	560	684	808	932	1056
6800	282	486	758	894	1030	1166	1302	180	384	656	792	928	1064	1200
7400	336	558	854	1002	1150	1298	1446	234	456	752	900	1048	1196	1344

^aCC + GLY + ACR/ATZ indicates cover crop terminated with glyphosate plus a premix of acetochlor/atrazine

Chapter 7 - Conclusions and Future Directions

Field surveys and screening of kochia populations from Kansas, Oklahoma and Texas confirmed multiple herbicide resistance to glyphosate, chlorsulfuron, dicamba, atrazine, and fluroxypyr. Dose-response studies further revealed low to moderate level resistance to glyphosate (3- to 5-fold), dicamba (5- to 13-fold), and fluroxypyr (3- to 6-fold) in selected kochia populations from Kansas and Oklahoma and resistance to glyphosate (5-fold) and dicamba (2- to 4-fold) in selected kochia populations from Texas. Future studies should investigate the underlying mechanism(s) of resistance in these multiple herbicide resistant (MHR) kochia populations.

Results from greenhouse and field studies 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. In a separate field and greenhouse study, tank-mixing of pyridate with atrazine, dicamba, dichlorprop-p, fluroxypyr, glyphosate, or halauxifen/fluroxypyr also resulted in synergistic or additive interactions and provided greater control of MHR kochia than without adding pyridate in mixtures. Considering the rapid spread of MHR kochia, the synergistic interactions of auxinic herbicides in two- or three-way mixtures or pyridate tank-mixed with atrazine, dicamba, dichlorprop-p, fluroxypyr, glyphosate, or halauxifen/fluroxypyr can play a crucial role in managing MHR kochia populations in fallow-fields or burndown (prior to crop planting or after crop harvesting) after careful considerations of the rotational crops. However, it is also important to note that overreliance on these mixtures should be avoided to further prevent

evolution of cross-resistance to auxinic herbicides or pyridate-based mixtures among kochia populations. Along with these herbicide tank-mixtures, growers should also adopt integrated weed management approach, 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 for kochia control. Future studies will investigate the possible underlying mechanism(s) of synergistic interactions among herbicides in various mixtures for MHR kochia control.

Results from the 3-year field study indicated that fall-planted cover crops (CC) after winter wheat harvest and terminated in the following spring with glyphosate plus acetochlor/atrazine could delay and reduce the cumulative emergence of glyphosate-resistant (GR) Palmer amaranth in grain sorghum. Different Palmer amaranth populations within central Great Plains could exhibit a differential emergence pattern as previously observed by Liu et al. (2022). Therefore, the findings of the current study will further help to improve the prediction models for Palmer amaranth emergence under CC plus residual herbicide strategy to build decision support tools for cost-effective and sustainable management of GR Palmer amaranth in the region. These combined practices of CC and residual herbicides would also help growers to target smaller GR Palmer amaranth seedlings with post-emergence herbicides for effective in-season control. Future studies should investigate weed seedbank dynamics with CC plus residual herbicides under winter wheat-grain sorghum-fallow rotation.

Results also showed that terminating fall-planted CC with glyphosate in combination with residual herbicide before grain sorghum planting in the winter wheat-grain sorghum-fallow rotation can provide an effective weed suppression in grain sorghum. Fall-planted CC terminated with glyphosate plus acetochlor/atrazine reduced total weed density by 34 to 81% and total weed

biomass by 45 to 73% compared to chemical fallow over the grain sorghum growing season. No sorghum yield penalty was observed after CC terminated with glyphosate plus acetochlor/atrazine. However, due to lower grain sorghum yield in all three years, results suggested that growing CC for only weed suppression in the semiarid central Great Plains would not be profitable. Therefore, CC could be used for hay or forage to increase the net returns (Holman et al. 2018, 2021, 2022, 2023; Obour et al. 2022). However, weed control during the sorghum growing season may be affected after the CC residue removal from the field. Therefore, future studies should focus on understanding the CC grazing timing and its interaction with weed control during the grain sorghum growing period. Furthermore, based on the available soil moisture, the fallow phase after grain sorghum harvest in the wheat-grain sorghum-fallow rotation could also be replaced with the spring-planted CC such as barley, oat, or pea for summer annual weed suppression.

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Appendix A - Supplementary Material

Appendix Table A.1. Information on kochia populations collected from Kansas, Oklahoma, and Texas during 2018 and 2019.

Population	Longitude	Latitude	Comments
KS 18-1	-99.3064	38.3199	Wheat Stubble
KS 18-2	-99.3087	38.3199	Wheat Stubble
KS 18-3	-99.3270	38.4663	Wheat Stubble
KS 18-4	-100.0704	38.4501	Wheat Stubble
KS 18-5	-100.3176	38.4802	Wheat Stubble
KS 18-6	-100.5242	38.6102	Wheat Stubble
KS 18-6	-100.5433	38.5116	Wheat Stubble
KS 19-1	-101.0500	39.4200	Wheat Stubble
KS 19-2	-101.6250	39.7570	Wheat Stubble
KS 19-3	-101.7070	39.5100	Wheat Stubble
KS 19-4	-101.7410	38.7300	Wheat Stubble
KS 19-5	-101.7700	38.4700	Wheat Stubble
KS 19-6	-101.7100	38.4400	Wheat Stubble
KS 19-7	-101.7200	38.4300	Wheat Stubble
KS 19-8	-101.6500	38.4370	Wheat Stubble
KS 19-9	-101.6000	38.4220	Wheat Stubble
KS 19-10	-101.5901	38.4431	Wheat Stubble
KS 19-11	-101.5601	38.4044	Wheat Stubble
KS 19-CI	-99.3256	38.8537	Sorghum Stubble
OK-1	-97.3628	36.8586	Wheat Stubble
OK-2	-97.4603	36.7127	Wheat Stubble
OK-3	-97.6243	36.7413	Wheat Stubble
OK-10	-97.6039	36.7508	Wheat Stubble
OK-11	-97.9238	36.6787	Wheat Stubble
OK-12	-98.3633	36.6976	Wheat Stubble
OK-13	-98.5781	36.8465	Wheat Stubble
OK-15	-99.4646	36.8592	Wheat Stubble
OK-17	-99.8351	36.8365	Wheat Stubble
OK-19	-100.6511	36.9671	Wheat Stubble
OK-20	-100.8032	36.9518	Wheat Stubble
OK-21	-101.0475	36.8646	Wheat Stubble
OK-23	-102.4604	36.7800	Wheat Stubble
TX 18-52	-101.5936	35.2661	Cotton Field
TX 18-53	-101.5567	35.2739	Sorghum Field Edge
TX 19-54	-101.5006	35.3536	Cotton Field Edge
TX 19-55	-101.3817	35.2658	Wheat Field Edge
TX 19-56	-101.4228	35.2072	Wheat Field Edge
TX 19-57	-101.5322	35.2206	Sorghum Field Edge
TX 19-58	-102.0400	35.1800	Sorghum Field Edge
TX 19-59	-102.1494	35.2081	Fallow Field Edge

TX 19-60	-102.2025	35.1750	Cotton Field Edge
TX 19-61	-102.2042	35.0444	Cotton Field Edge
TX 19-62	-102.2653	34.9861	Cotton Field Edge
TX 19-63	-102.4025	35.0097	Cotton Field Edge
TX 19-64	-102.4383	35.0572	Wheat Field Edge
TX 19-65	-102.4100	34.9100	Sorghum Field Edge
TX 19-66	-102.5600	34.8100	Wheat Field Edge
TX 19-67	-102.5725	34.7590	Cotton Field Edge
TX 19-68	-102.7100	34.6600	Wheat Field Edge
TX 19-69	-102.6899	34.5261	Fallow Field Edge
TX 19-71	-102.5900	34.3800	Fallow Field Edge
TX 19-72	-102.4600	34.3600	Corn Field Edge
TX 19-73	-102.4600	34.4700	Corn Field Edge
TX 19-74	-102.3396	34.4250	Wheat Field Edge
TX 19-77	-102.0376	34.3918	Cotton Field Edge
TX 19-78	-101.7200	34.2900	Fallow Field Edge
TX 19-80	-102.0200	34.2600	Cotton Field Edge
TX 19-81	-102.0400	34.1900	Fallow Field Edge
TX 19-82	-101.8400	34.1100	Cotton Field Edge
TX 19-83	-102.0369	34.0361	Fallow Field Edge
TX 19-84	-101.9400	33.9700	Fallow Field Edge
TX 19-86	-101.8000	34.0700	Fallow Field Edge
TX 19-88	-101.8294	34.1997	Cotton Field Edge
TX 19-89	-101.6600	34.1700	Corn Field Edge
TX 19-90	-101.5022	34.1561	Wheat Field Edge
TX 19-91	-101.4753	34.0517	Cotton Field Edge
TX 19-92	-101.5700	34.0300	Wheat Field Edge
TX 19-93	-101.5500	33.9200	Cotton Field Edge
TX 19-94	-101.5133	33.8847	Cotton Field Edge
TX 19-95	-101.6608	33.8656	Corn Field Edge
TX 19-96	-101.7653	33.4214	Cotton Field Edge
TX 19-97	-101.5100	33.7700	Wheat Field Edge
TX 19-99	-101.6500	33.7400	Fallow Field Edge
TX 19-100	-101.7500	33.6900	Cotton Field Edge
TX 19-101	-101.8700	33.7700	Pasture Edge
TX 19-102	-102.0567	33.7664	Cotton Field Edge
TX 19-103	-102.1100	33.6900	Cotton Field Edge
TX 19-104	-102.0400	33.6500	Cotton Field Edge
TX 19-105	-102.6010	33.6300	Cotton Field Edge
TX 19-106	-102.1400	33.6200	Fallow/Pasture Edge
TX 19-107	-102.2100	33.5400	Cotton Field Edge
TX 19-108	-102.1200	33.4500	Cotton Field Edge
