

Research Article

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Control of waterhemp (*Amaranthus tuberculatus*) at multiple locations in Illinois with single preemergence applications of VLCFA-inhibiting herbicides

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Abstract

Herbicides that inhibit very-long-chain fatty acids (VLCFAs) have been widely used for pre-emergence control of annual monocot and small-seeded dicot weed species, such as waterhemp, since their discovery in the 1950s. VLCFA-inhibiting herbicides are often applied in combination with active ingredients that possess residual activity on small-seeded broadleaf weeds, which can make their contribution to preemergence waterhemp control difficult to quantify. Bare-ground field experiments were designed to investigate the efficacy of eight VLCFA-inhibiting herbicides applied at their minimum and maximum labeled rates for control of Illinois waterhemp populations. Four different locations were selected, two of which contained previously characterized VLCFA inhibitor-resistant waterhemp populations in Champaign County (CHR) and McLean County (MCR). Two locations with VLCFA inhibitor-sensitive waterhemp populations included the University of Illinois South Farm in Urbana, IL, and the Orr Research Center in Perry, IL. Soils at the CHR, MCR, and Urbana locations contained greater than 3% organic matter, but less than 3% organic matter at Perry. Non-encapsulated acetochlor and alachlor controlled CHR and MCR waterhemp populations 28 d after treatment (DAT), whereas other VLCFA-inhibiting herbicides resulted in 61% and 76% control of the CHR and MCR populations, respectively. In contrast, all VLCFA-inhibiting herbicides resulted in 81% and 88% control of the Perry and Urbana waterhemp populations, respectively, 28 DAT. Waterhemp control decreased by 42 DAT, especially for the VLCFA inhibitor-resistant CHR and MCR populations. Overall, VLCFA-inhibiting herbicides remain effective for controlling sensitive waterhemp, but most are not effective for controlling VLCFA inhibitor-resistant waterhemp populations. Proper herbicide stewardship and integrated weed management practices should be implemented to maintain VLCFA-inhibiting herbicide efficacy for waterhemp management in the future.

Introduction

Waterhemp is a small-seeded, summer annual, dicot weed species that is among the most challenging to control in crops such as corn (*Zea mays* L.) and soybean (*Glycine max* L. Merr) in the midwestern United States (Sauer 1955; Steckel 2007). Waterhemp can reduce corn yield by more than 50% when interference occurs early in the growing season (Steckel and Sprague 2004). Additionally, soybean yield can be reduced by 40% from season-long interference (Hager et al. 2002b). Waterhemp uses the C4 carbon fixation pathway and can produce in excess of one million seeds per female plant (Steckel 2007; Steckel et al. 2003). Seeds are viable within 2 wk of pollination and are frequently dormant (Bell and Tranel 2010). Seed dormancy enables waterhemp to emerge in multiple cohorts throughout the growing season (Buhler and Hartzler 2001; Hartzler et al. 1999). The prolonged emergence makes control difficult, necessitating the use of herbicides with soil-residual activity that are often followed by postemergence herbicides and nonchemical control measures (Steckel et al. 2002).

Waterhemp is dioecious (separate male and female plants) and requires outcrossing for successful pollination (Murray 1940; Sauer 1955). Outcrossing results in high intraspecific genetic variability compared with self-pollinated weed species (Murray 1940). High genetic variability has contributed to the frequent evolution and widespread distribution of herbicide resistance (Adhikary and Pratt 2015; Steckel 2007; Tranel et al. 2011; Tranel 2020). To date, waterhemp has evolved resistance to inhibitors of acetolactate synthase, photosystem

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II, 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS), protoporphyrinogen oxygenase (PPO), 4-hydroxyphenylpyruvate dioxygenase, synthetic auxins, and most recently, very-long-chain fatty acids (VLCFAs; Heap 2021). Multiple resistance to herbicides representing up to six different site of action (SOA) groups has also been reported within waterhemp populations (Evans et al. 2019; Shergill et al. 2018; Strom et al. 2019).

VLCFA-inhibiting herbicides result in preemergence (PRE) control of annual monocot and small-seeded dicot weeds (Fuerst 1987), including waterhemp and Palmer amaranth (*Amaranthus palmeri* S. Watson). VLCFAs consist of acyl chains in excess of 18 carbons and are essential for the formation of cell membranes, cuticle waxes, and lipids (Bach and Faure 2010). VLCFA inhibitors control sensitive weed species by inhibiting the VLCFA-elongase complex in plant cells and subsequent formation of VLCFAs (Böger 2003). Sensitive weed species either fail to emerge or remain in an arrested state of growth after cotyledon expansion (Deal and Hess 1980; Dhillon and Anderson 1972; Fuerst 1987; Pillai et al. 1979). VLCFA-inhibiting herbicides are among the oldest classes of herbicides used in cropping systems and were originally discovered and developed in the 1950s (Hamm 1974).

Resistance to VLCFA inhibitors has been relatively infrequent in comparison to herbicides from other SOA groups, and has been reported in only five monocot and two dicot weed species (Heap 2021). We previously reported that waterhemp populations from McLean county (MCR) and Champaign county (CHR) Illinois are resistant to VLCFA-inhibiting herbicides (Strom et al. 2019, 2020). Control of each population in the field was poor with most VLCFA-inhibiting herbicides (Evans et al. 2019; Hausman et al. 2013; Strom et al. 2019). VLCFA-inhibiting herbicides are subject to environmental factors that influence their activity. Precipitation is especially important for VLCFA-inhibiting herbicide incorporation and subsequent weed control (Jhala 2017). Edaphic factors also influence PRE activity of VLCFA-inhibiting herbicides. In general, higher herbicide rates are needed for fine-textured soils with high organic matter. Soils with high organic matter also tend to have high microbial activity, which could reduce residual weed control with VLCFA-inhibiting herbicides (Shaner et al. 2006; Wu et al. 2011).

VLCFA-inhibiting herbicides are commonly applied in preformulated combinations or tank mixtures with herbicides from other SOA groups, and their contribution to weed control can often be difficult to quantify. The objective of our experiments was to investigate the efficacy of VLCFA-inhibiting herbicides applied alone to control waterhemp at locations with differing soil types. The four field locations included two VLCFA inhibitor-resistant and two VLCFA inhibitor-sensitive waterhemp populations.

Materials and Methods

Site Selection

Field experiments were conducted from 2018 to 2020 at four locations in Illinois, with two locations containing VLCFA inhibitor-resistant waterhemp populations: one in Champaign County (Evans et al. 2019; Strom et al. 2019), and one from McLean County (Hausman et al. 2011, 2013; Strom et al. 2019). These locations are designated CHR and MCR, respectively, to coincide with the characterized weed populations. The soil at CHR was a Flanigan silt loam (Fine, smectitic, mesic Aquic Argiudolls), pH 5.5, and 4.8% organic matter. The soil at MCR was a Sable silty clay loam (fine-silty, mixed, superactive, mesic Typic Endoaquolls),

pH 6.4, and 3.2% organic matter. Two additional locations with VLCFA inhibitor-sensitive waterhemp populations were selected. One was at the University of Illinois Agronomy Research and Education Center in Urbana, IL (designated Urbana). The soil at Urbana was a Drummer silty clay loam (fine-silty, mixed, superactive, mesic Typic Endoaquolls), pH 6.7, and 5.5 % organic matter. The other field site was at the University of Illinois Orr Research Center in Perry, IL (designated Perry). The soil at the site was a Downsouth silt loam (Fine-silty, mixed, superactive, mesic Mollic Oxyaquic Hapludalfs), pH 6.2, and 2.5 % organic matter. The Urbana and Perry waterhemp populations were confirmed to be resistant to PPO and EPSPS inhibitors using whole-plant greenhouse assays (Perry) or molecular marker assays (Urbana) by the University of Illinois Plant Clinic (data not shown).

General Field Methods

Eight VLCFA-inhibiting herbicides were applied at their respective minimum and maximum labeled rates based on the manufacturers' recommendation according to soil type (Table 1). Each year the same fields were used at each location, but experiments were conducted in different areas each season. Field experiments were initiated at all locations within 7 d of forecasted rainfall. Cumulative precipitation for each location is presented in Table 2. Experiments were initiated at CHR May 17, June 11, and June 2 in 2018, 2019, and 2020, respectively. MCR experiments were initiated May 2, May 22, and May 13, in 2018, 2019, and 2020, respectively. Experiments were established May 17 and June 19 at Urbana for 2019 and 2020, respectively; and April 30, June 14, and May 12 at Perry for 2018, 2019, and 2020, respectively. Data from 2018 MCR was excluded due to lack of timely initial rainfall and no observable herbicide activity.

The soil at each location was tilled prior to herbicide application to control existing vegetation. Individual bare ground plots measured 3 m × 7.6 m with treatments arranged in a randomized complete block design with four replications at each location. Treatments were applied with a pressurized CO₂ backpack sprayer calibrated to deliver 187 L ha⁻¹ at 276 kPa with a 3-m boom consisting of six AI110025VS nozzles (Teejet Technologies, Wheaton, IL) spaced 51 cm apart. Herbicide efficacy was assessed 28 and 42 d after treatment (DAT) using visual ratings of percent waterhemp control recorded on a scale of 0% (no control) to 100% (complete control). Ratings considered waterhemp density, injury, and biomass reduction compared with a nontreated control. Waterhemp density per square meter was recorded from a consistent quadrat location in the middle of each plot at each evaluation timing, and aboveground biomass was subsequently harvested 42 DAT. Aboveground biomass was then dried in a forced air dryer at 65 C.

Statistical Analysis

Statistical analysis was conducted with SAS software (v9.4, SAS Institute, Inc., Cary, NC). Waterhemp control and density were analyzed by ANOVA using a generalized linear mixed model (GLIMMIX procedure). The assumption of normally distributed residuals was reviewed with the UNIVARIATE procedure and homogeneous variance was checked with the GLM procedure. Waterhemp control was fitted to a Beta distribution with a logit link function (Davis 2018). Waterhemp density and aboveground biomass were fitted to a negative binomial distribution with a log link function (O'Hara and Kotze 2010). We first determined whether interactions of herbicide treatment

Table 1. VLCFA-inhibiting herbicides, rates, and source information for field studies at multiple locations in Illinois with differing soil types (2018–2020).^{d,e}

Common name	Trade name	Organic matter application rate ^a		Manufacturer
		<3%	>3%	
		kg ha ⁻¹		
Acetochlor ^b	Warrant	1.3	2.3	Bayer CropScience, St. Louis, MO 63137
		2.3	2.5	
Acetochlor	Harness	1.7	2.2	Bayer CropScience, St. Louis, MO 63137
		2.2	2.7	
S-metolachlor	Dual Magnum	1.4	1.8	Syngenta Crop Protection, Greensboro, NC 27419
		1.8	2.1	
S-metolachlor ^c	Dual II Magnum	1.4	1.8	Syngenta Crop Protection, Greensboro, NC 27419
		1.8	2.1	
Metolachlor	Stalwart	1.5	1.9	SipcamAgro, Durham, NC 27713
		1.9	2.2	
Dimethenamid- <i>P</i>	Outlook	0.7	1.0	BASF Corporation Agricultural Products, Research Triangle Park, NC 27709
		1.0	1.1	
Alachlor	IntRRo	2.2	2.8	Bayer CropScience, St. Louis, MO 63137
		3.1	3.4	
Pyroxasulfone	Zidua	0.1	0.1	BASF Corporation Agricultural Products, Research Triangle Park, NC 27709
		0.2	0.2	

^aApplication rates were chosen based on manufacturer's labeled recommendations for each soil type and two rate structures were chosen by organic matter content. Application rates for soils less than 3% organic matter were used at the Perry, IL, location. All other locations used rates based on greater than 3% soil organic matter.

^bEncapsulated formulation.

^cContains the safener benoxacor.

^dHerbicides were applied to bare ground prior to weed emergence.

^eAbbreviation: VLCFA, very long-chain fatty acid.

Table 2. Cumulative precipitation at field locations in Illinois during 2018–2020 field experiments.

Location	Days after treatment	Precipitation (cm)		
		2018	2019	2020
Champaign County ^a	7	0.2	3.4	10.6
	14	4.2	7.2	12.0
	42	33.9	11.2	22.7
McLean County ^a	7	0.5	5.8	9.2
	14	0.7	7.6	9.4
	42	5.4	19.6	15.8
Urbana ^b	7	–	4.4	7.1
	14	–	8.1	9.0
	42	–	16.5	21.3
Perry ^c	7	1.8	7.5	1.5
	14	2.2	11.6	7.2
	42	8.5	18.0	25.2

^aData acquired using a Watchdog 2000 Series Weather Station, Spectrum Technologies Inc., 3600 Thayer Ct., Aurora, IL 60504.

^bData acquired from the Illinois State Water Survey, Prairie Research Institute, 2204 Griffith Dr., Champaign, IL 61820.

^cData acquired from the U.S. Department of Commerce National Oceanic & Atmospheric Administration, Savoy 0.9 N, IL US US1LCP0082, National Centers for Environmental Information 151 Patton Ave, Asheville, NC 28801.

and location existed. Fixed effects were location, VLCFA-inhibiting herbicide, rate, and their interactions. Random effects included year and block nested within year. Initial analysis revealed location and location by VLCFA-inhibiting herbicide interaction effects were significant. The range of waterhemp control with each VLCFA-inhibiting herbicide is presented in Figure 1, and each location was then analyzed separately to explore control at each location. Analysis did not indicate a significant interaction of VLCFA-inhibiting herbicide and rate. Mean estimates were then separated by Fisher's LSD ($\alpha = 0.05$), and treatment means were

pooled between rates. Listed means represent the data scale and were back-transformed following analysis.

Results and Discussion

Champaign County Resistant Location

At 28 DAT, only non-encapsulated acetochlor and alachlor resulted in 91% or greater control of waterhemp at CHR, whereas control with all other VLCFA-inhibiting herbicides was 61% or less (Table 3). Waterhemp control at the CHR location was the least with metolachlor and S-metolachlor and did not exceed 37%. Non-encapsulated acetochlor and alachlor reduced waterhemp densities 92% to 95% 28 DAT. Dimethenamid-*P*, encapsulated acetochlor, and pyroxasulfone reduced waterhemp density 72% to 77% 28 DAT while metolachlor or S-metolachlor reduced waterhemp density 42% to 62%.

Waterhemp control at the CHR location with all VLCFA-inhibiting herbicides decreased by 42 DAT (Table 3). Non-encapsulated acetochlor and alachlor resulted in 85% to 88% control, respectively, whereas control was 40% or less for the other VLCFA-inhibiting herbicides. These results with non-encapsulated acetochlor are consistent with previous reports in which non-encapsulated acetochlor controlled waterhemp at CHR by more than 70%, whereas control with other VLCFA-inhibiting products was less (Evans et al. 2019; Strom et al. 2019). Waterhemp densities in non-treated plots decreased by 42 DAT, potentially due to intraspecific competition. Non-encapsulated acetochlor and alachlor reduced waterhemp density 88% and 84%, respectively, at 42 DAT. Other VLCFA-inhibiting herbicides reduced waterhemp density less than 66% at 42 DAT, while metolachlor and S-metolachlor reduced density by only 26% to 45%. Average biomass was 336 g m⁻² for nontreated plots 42 DAT, and non-encapsulated acetochlor reduced biomass by 81%. Alachlor reduced biomass by 74%, but biomass reduction did not exceed 41% for all other

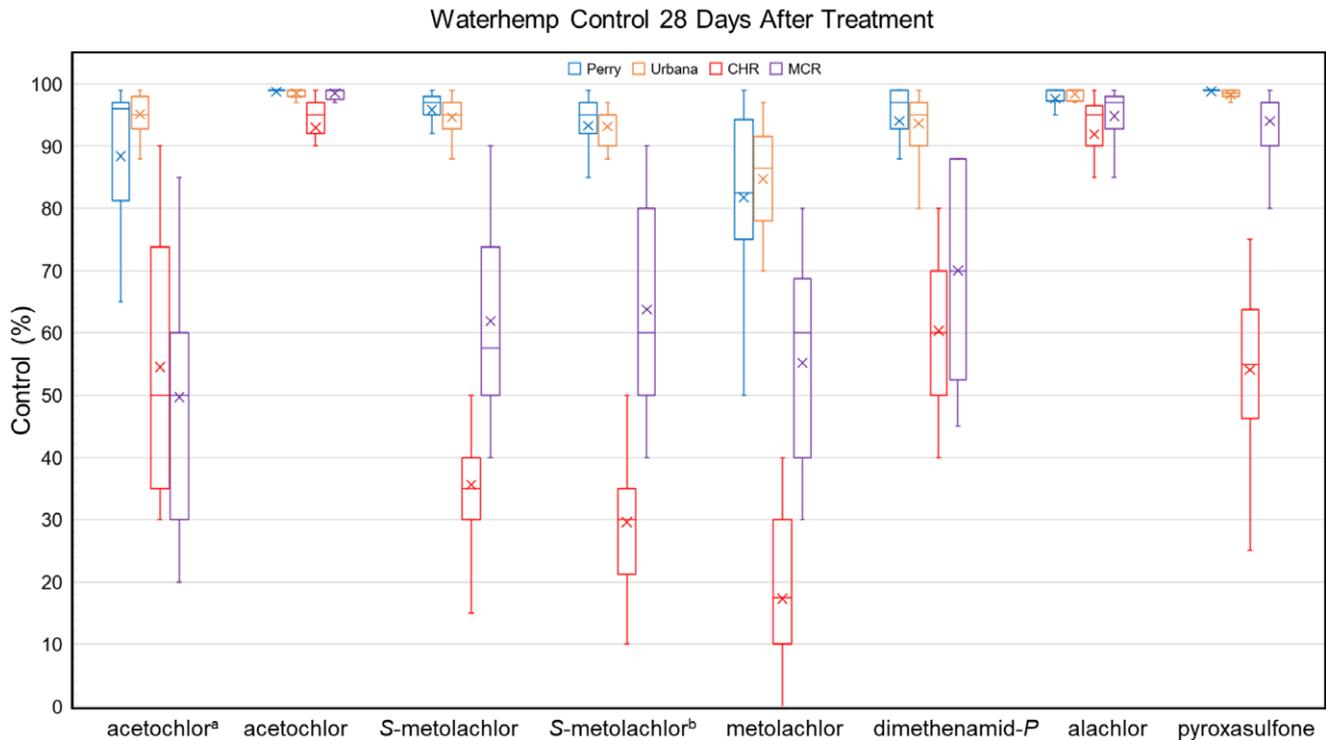


Figure 1. Waterhemp control at each location with eight different very-long-chain fatty acid (VLCFA)-inhibiting herbicides at 28 d after treatment. Blue, orange, red, and purple boxplots correspond to the Perry, Urbana, CHR, and MCR locations, respectively. ^aEncapsulated formulation. ^bContains the herbicide safener benoxacor.

VLCFA-inhibiting active ingredients. Biomass from plots treated with either *S*-metolachlor formulation, metolachlor, or pyroxasulfone was not different from the nontreated control.

McLean County Resistant Location

Control of waterhemp at the MCR location with VLCFA-inhibiting herbicides 28 DAT was greater than previously reported by Hausman et al. (2013). VLCFA-inhibiting herbicide was significant ($P = 0.007$): non-encapsulated acetochlor, alachlor, and pyroxasulfone controlled waterhemp at MCR by 94% to 98% 28 DAT (Table 4). Control with other treatments was 62% to 76%. Metolachlor and *S*-metolachlor controlled waterhemp at MCR by 62% to 73%, whereas control 30 DAT was 17% to 53% in previous research (Hausman et al. 2013). All VLCFA-inhibiting herbicides reduced waterhemp density 28 DAT. Non-encapsulated acetochlor and alachlor reduced waterhemp density by 94% and 91%, respectively, whereas metolachlor and *S*-metolachlor reduced waterhemp density by 64% to 72%. Encapsulated acetochlor reduced waterhemp density by only 48%.

At 42 DAT non-encapsulated acetochlor, alachlor, and pyroxasulfone resulted in the greatest control (85% to 94%), whereas control among the other VLCFA-inhibiting herbicides was similar and ranged from 39% to 54% (Table 4). Hausman et al. (2013) reported 7% to 55% control of waterhemp at MCR 60 DAT. Waterhemp density in nontreated plots was 69 plants m^{-2} and all VLCFA-inhibiting herbicides reduced waterhemp density. Encapsulated acetochlor reduced waterhemp density by only 32%. Waterhemp densities in plots treated with metolachlor, *S*-metolachlor, and dimethenamid-*P* were similar and were 45% to 55% less than the nontreated. Non-encapsulated acetochlor reduced waterhemp density the most (90%) 42 DAT. All

VLCFA-inhibiting herbicides reduced waterhemp biomass 42 DAT. Biomass from nontreated plots was 83 g m^{-2} , and non-encapsulated acetochlor resulted in the greatest reduction (96%). Biomass from plots treated with encapsulated acetochlor, *S*-metolachlor, metolachlor, and dimethenamid-*P* were similar and were 48% to 65% less than the nontreated.

Urbana, IL, Location

VLCFA-inhibiting herbicides were effective for control of the Urbana waterhemp populations 28 DAT (Figure 2). VLCFA-inhibiting herbicide was significant ($P = 0.0013$); control with each VLCFA-inhibiting herbicide was 88% or greater (Table 5). Control with non-encapsulated acetochlor, alachlor, and pyroxasulfone was 98%. *S*-metolachlor, encapsulated acetochlor, and dimethenamid-*P* resulted in up to 96% control, which was greater than that reported at the VLCFA-inhibiting herbicide-resistant locations (Tables 3 and 4). Results from the Urbana location are consistent with previous reports in which non-encapsulated acetochlor, *S*-metolachlor, dimethenamid-*P*, and pyroxasulfone resulted in greater than 85% control of waterhemp 28 DAT (Hedges et al. 2019; Schryver et al. 2017; Steckel et al. 2002). Waterhemp density in nontreated plots averaged 122 plants m^{-2} and was reduced at least 95% with non-encapsulated acetochlor, alachlor, and pyroxasulfone. Encapsulated acetochlor reduced waterhemp density 89%, while *S*-metolachlor and dimethenamid-*P* reduced density 80% to 84%. Metolachlor reduced waterhemp density by only 67%.

At 42 DAT waterhemp control with all VLCFA-inhibiting herbicides decreased (Table 5), but control with all treatments, with the exception of metolachlor, remained 80% or greater. Control with non-encapsulated acetochlor, alachlor, and pyroxasulfone was at least 95% 42 DAT. In general, waterhemp densities

Table 3. Mean estimates^a of waterhemp control 28 and 42 DAT, density 28 and 42 DAT, and recovered biomass 42 DAT for VLCFA-inhibiting herbicides at the Champaign County, IL, location (2018–2020).^d

Herbicide	Control		Density		Biomass
	28 DAT	42 DAT	28 DAT	42 DAT	42 DAT
	———— % ————		— plants m ⁻² —		g m ⁻²
Acetochlor ^b	56 b	39 b	49 d	46 d	219 b
Acetochlor	93 a	88 a	10 f	15 e	64 e
S-metolachlor	37 c	20 bc	72 c	67 c	291 ab
S-metolachlor ^c	30 cd	20 bc	79 c	66 c	262 abc
Metolachlor	20 d	13 c	111 b	89 b	300 a
Dimethenamid- <i>P</i>	61 b	28 bc	44 d	42 d	198 c
Alachlor	91 a	85 a	15 e	19 e	89 d
Pyroxasulfone	57 b	40 b	52 d	43 d	253 abc
Nontreated	–	–	190 a	121 a	336 a

^aMean estimates with the same letter within a column are not significantly different at $\alpha = 0.05$ separated by LSD.

^bEncapsulated formulation.

^cContains the herbicide safener benoxacor.

^dAbbreviations: DAT, days after treatment; VLCFA, very long-chain fatty acid.

Table 4. Mean estimates^a of waterhemp control 28 and 42 DAT, density 28 and 42 DAT, and recovered biomass 42 DAT for VLCFA-inhibiting herbicides at the McLean County, IL, location (2018–2020).^{b,e}

Herbicide	Control		Density		Biomass
	28 DAT	42 DAT	28 DAT	42 DAT	42 DAT
	———— % ————		— plants m ⁻² —		g m ⁻²
Acetochlor ^c	65 c	39 b	45 b	47 b	43 b
Acetochlor	98 a	94 a	5 f	7 e	3 e
S-metolachlor	73 c	52 b	24 cd	31 c	38 bc
S-metolachlor ^d	71 c	48 b	25 cd	33 c	34 bc
Metolachlor	62 c	40 b	31 c	38 bc	35 bc
Dimethenamid- <i>P</i>	76 bc	54 b	21 d	32 c	29 c
Alachlor	97 a	90 a	8 ef	13 d	6 d
Pyroxasulfone	94 ab	85 a	10 e	13 d	7 d
Acetochlor ^c	65 c	39 b	45 b	47 b	43 b
Nontreated	–	–	86 a	69 a	83 a

^aMean estimates with the same letter within a column are not significantly different at $\alpha = 0.05$ separated by LSD.

^bData from 2018 were excluded due to lack of initial rainfall.

^cEncapsulated formulation.

^dContains the herbicide safener benoxacor.

^eAbbreviations: DAT, days after treatment; VLCFA, very long-chain fatty acid.

in the treated plots increased by 42 DAT, whereas densities in the nontreated decreased slightly. Non-encapsulated acetochlor, alachlor, and pyroxasulfone decreased waterhemp density by 93%, 94%, and 93%, respectively, whereas density reduction with other treatments was 83% or less. All treatments reduced waterhemp biomass 42 DAT when compared with the nontreated, with pyroxasulfone reducing biomass 99%.

Perry, IL, location

The Perry, IL, location was the only location with soils containing less than 3% organic matter. Results from Perry were similar to those from the Urbana location, and VLCFA-inhibiting herbicide was significant ($P = 0.0016$). At 28 DAT all treatments controlled waterhemp by 81% or greater (Table 6). Control among treatments was similar, with the exceptions of encapsulated acetochlor and metolachlor, which resulted in 88% and 81% control, respectively.

Table 5. Mean estimates^a of waterhemp control 28 and 42 DAT, density 28 and 42 DAT, and recovered biomass 42 DAT for VLCFA-inhibiting herbicides at the Urbana, IL, location (2019–2020).^d

Herbicide	Control		Density		Biomass
	28 DAT	42 DAT	28 DAT	42 DAT	42 DAT
	———— % ————		— plants m ⁻² —		g m ⁻²
Acetochlor ^b	96 b	88 abc	14 d	18 d	58 c
Acetochlor	98 a	95 ab	5 e	8 e	24 d
S-metolachlor	95 b	84 abc	19 cd	24 cd	74 bc
S-metolachlor ^c	93 b	80 bc	24 c	26 bc	117 b
Metolachlor	88 c	68 c	40 b	35 b	126 b
Dimethenamid- <i>P</i>	94 b	87 abc	19 cd	20 b	86 bc
Alachlor	98 a	96 a	5 e	7 e	6 e
Pyroxasulfone	98 a	95 ab	6 e	8 e	2 f
Nontreated	–	–	122 a	108 a	345 a

^aMean estimates with the same letter within a column are not significantly different at $\alpha = 0.05$ separated by LSD.

^bEncapsulated formulation.

^cContains the herbicide safener benoxacor.

^dAbbreviations: DAT, days after treatment; VLCFA, very long-chain fatty acid.

Table 6. Mean estimates^a of waterhemp control 28 and 42 DAT, density 28 and 42 DAT, and recovered biomass 42 DAT for VLCFA-inhibiting herbicides at the Perry, IL, location (2018–2020).^d

Herbicide	Control		Density		Biomass
	28 DAT	42 DAT	28 DAT	42 DAT	42 DAT
	———— % ————		— plants m ⁻² —		g m ⁻²
Acetochlor ^b	88 bc	79 c	21 bc	22 bcd	9 cd
Acetochlor	98 a	95 a	5 e	6 fg	2 ef
S-metolachlor	96 a	86 bc	12 d	15 de	7 d
S-metolachlor ^c	93 ab	80 c	19 cd	26 bc	18 bc
Metolachlor	81 c	60 d	32 b	33 b	21 b
Dimethenamid- <i>P</i>	94 ab	85 bc	14 cd	18 cd	7 d
Alachlor	98 a	92 ab	6 e	9 ef	3 e
Pyroxasulfone	98 a	94 a	4 e	5 g	1 f
Nontreated	–	–	91 a	81 a	102 a

^aMean estimates with the same letter within a column are not significantly different at $\alpha = 0.05$ separated by LSD.

^bEncapsulated formulation

^cContains the herbicide safener benoxacor.

^dAbbreviations: DAT, days after treatment; VLCFA, very long-chain fatty acid.

All treatments reduced waterhemp density, with non-encapsulated acetochlor, alachlor, and pyroxasulfone reducing density by 93% to 96%. Other treatments reduced waterhemp density up to 65% to 87%; encapsulated acetochlor and metolachlor reduced densities the least.

Waterhemp control at Perry decreased by 42 DAT, but all VLCFA-inhibiting herbicides resulted in greater than 80% control with the exception of encapsulated acetochlor and metolachlor (Table 6). Control at Perry 42 DAT with racemic metolachlor was greater than that observed at the CHR and MCR locations (Tables 3 and 4), but less than previous reports where racemic metolachlor resulted in more than 80% control up to 60 DAT (Hager et al. 2002a; Oliveira et al. 2017). Control was greatest with non-encapsulated acetochlor, alachlor, and pyroxasulfone (92% to 95%). Waterhemp densities in treated plots numerically increased at 42 DAT, but remained less than the nontreated. Encapsulated acetochlor, S-metolachlor, and metolachlor reduced waterhemp density by 59% to 81%. All herbicides reduced aboveground



Figure 2. Representative images of very-long-chain fatty acid (VLCFA)-inhibiting herbicide efficacy for control of VLCFA inhibitor-resistant waterhemp population (CHR) and a sensitive (Urbana) population, 28 d after treatment.

biomass 42 DAT compared with the nontreated. Nontreated biomass averaged 102 g m^{-2} , and biomass reduction ranged from 79% to 99% across treatments. Non-encapsulated acetochlor and pyroxasulfone reduced waterhemp biomass by 98% to 99%.

Implications

Only non-encapsulated acetochlor and alachlor controlled waterhemp at the CHR location by 91% or greater 28 DAT; these two VLCFA-inhibiting herbicides along with pyroxasulfone controlled waterhemp at the MCR location by 94% or greater 28 DAT. Control of these VLCFA inhibitor-resistant populations with other VLCFA-inhibiting herbicides ranged from 20% to 76% 28 DAT. All treatments reduced waterhemp density at CHR and MCR 28 DAT compared with a nontreated plot; non-encapsulated acetochlor and alachlor consistently reduced waterhemp density the most among all VLCFA-inhibiting herbicides. Previous greenhouse research has demonstrated that resistance ratios calculated from dose-response experiments for CHR and MCR are variable and tend to be greatest with *S*-metolachlor and least with acetochlor (Strom et al. 2019). In contrast, control of VLCFA inhibitor-sensitive waterhemp at Urbana was 93% or greater with all treatments except metolachlor, while the VLCFA inhibitor-sensitive waterhemp at Perry was controlled by at least 93% with all treatments except metolachlor and encapsulated acetochlor 28 DAT. Even though most VLCFA-inhibiting herbicides controlled sensitive waterhemp, resistance in the CHR and MCR populations should serve as a reminder that resistance to VLCFA-inhibiting herbicides has evolved and, if present, will reduce efficacy under field conditions.

Strom et al. (2020) demonstrated CHR and MCR seedlings rapidly metabolized *S*-metolachlor. Resistance ratios to pyroxasulfone, dimethenamid-*P*, and acetochlor for waterhemp from CHR and MCR were also calculated in greenhouse dose-response experiments (Strom et al. 2019). Mechanisms for reduced sensitivity to these other VLCFA-inhibiting herbicides have not been elucidated, but they might be similar to those conferring resistance to *S*-metolachlor. Resistance to soil-applied herbicides under field

conditions can be difficult to accurately identify. In general, resistance to soil-applied herbicides is manifest as a reduced duration of residual weed control (Hager 2019).

Poor weed control with soil-residual herbicides, including VLCFA-inhibiting herbicides, is not always due to resistance. Many climatic and edaphic factors influence herbicide activity and duration of residual control (Stewart et al. 2010). The timing and amount of precipitation in relation to herbicide application are essential for incorporation of surface-applied VLCFA-inhibiting herbicides (Jhala 2017). In addition, photodegradation can occur if excess time elapses between herbicide application and movement into the soil (Shaner, 2014). In general, fine soil textures with high organic matter require increased herbicide rates. Soils with higher organic matter also tend to have elevated microbial communities that hasten herbicide degradation, and thus, decrease residual control (Beestman and Deming 1974; Long et al. 2014; Shaner et al. 2006; Wu et al. 2011).

The magnitude and distribution of resistance to VLCFA-inhibiting herbicides is currently not well understood. A recent 5-yr study of PRE herbicides in Iowa discovered that most VLCFA-inhibiting herbicides resulted in greater than 80% control of waterhemp throughout the season, but there were instances when control was much less (Jha 2020). We have identified only two populations in Illinois that are resistant to VLCFA-inhibiting herbicides at this time (Strom et al. 2019, 2020). The dioecious biology of waterhemp contributes to the rapid spread of resistance traits if proper management and chemical stewardship practices are not implemented (Liu et al. 2012; Sarangi et al. 2017). Growers, applicators, and crop protection professionals should understand the effective weed control methods available for each field. The continued use of best management practices such as applications of herbicides from multiple, effective sites of action should be used (Evans et al. 2016), including those with soil-residual activity. In addition, overlapping residual herbicides can enhance the probability of an effective postemergence program (Chahal et al. 2018; Steckel et al. 2002).

Nonchemical control methods should also be considered in integrated management strategies. Cover crops such as cereal

rye have demonstrated promise in reducing weed densities and are compatible with many herbicide programs (Cornelius and Bradley 2017; Jha et al. 2020; Loux et al. 2017). Additionally, postharvest seed destruction and manual removal of weeds can be incorporated into sustainable weed management programs with the goal of limiting the number of seeds reintroduced into the soil seed bank each year (Schwartz-Lazaro and Copes 2019; Shergill et al. 2020; Walsh et al. 2012).

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