Weed community composition in simple and more diverse cropping systems

Abstract

Weed communities in three cropping systems suitable for the Midwestern USA were studied from 2017 through 2020 to examine how diversified cropping systems affected weed community diversity, stand density, and aboveground mass. A baseline 2-year cropping system with corn (*Zea mays* L.) and soybean (*Glycine max* (L.) Merr.) was diversified with cool-season crops, namely oat (*Avena sativa* L.), red clover (*Trifolium pratense* L.), and alfalfa (*Medicago sativa* L.) in 3-year and 4-year systems. Herbicide was not applied in oat, red clover, and alfalfa. The reduction in the mass of herbicide active ingredients applied in the 3-year and 4-year systems was associated with increased weed stand density, aboveground mass, and community diversity, but did not cause crop yield loss. The addition of the cool-season crops into the cropping system did not affect densities of emerged weeds but did affect weed growth. The dominance of aggressive weed species such as common waterhemp (*Amaranthus tuberculatus* (Moq ex DC) J.D. Sauer) and common lambsquarter (*Chenopodium album* L.) tended to be greater in corn and soybean phases of the rotations than in oat, red clover, and alfalfa.

Keywords: weed community, diversity, evenness, richness, cropping system diversification, Midwestern USA

Introduction

Materials and Methods

Empirical measurements of weed community composition were made from 2017 to 2020 at Iowa State University's Marsden Farm in Boone County, Iowa, USA, (42° 01'N, 93° 47'W, 333 m above sea level). All soil types present at the site are Mollisols (Chen et al., 2014). A detailed description of the experiment site and crop management can be found in Liebman et al. (2021) and the field layout and experiment design were provided in Nguyen and Liebman (in review). Briefly, a randomized complete block, split-plot design with four replications were used to study three different crop rotation systems (2-year, 3-year, or 4-year). The crop sequence in each rotation was presented in Nguyen and Liebman (in review). The main-plot factor ('crop identity') was represented by crop species and the rotation system in which it occurred (C2: corn in the 2-year rotation, C3: corn in the 3-year rotation, C4: corn in the 4-year rotation, S2: soybean in the 2-year rotation, S3: soybean in the 3-year rotation, S4: soybean in the 4-year rotation, O3: oat in the 3-year rotation, O4: oat in the 4-year rotation, and A4: alfalfa in the 4-year rotation). The split-plot effect, i.e., weed management regime applied in the corn phase (corn weed management), was represented by herbicide level (conventional - broadcast over the whole corn area, or low - banded 38 cm wide on top of corn rows). Details concerning crop genotypes and weed management regimes are provided in Table 1.

Table 1: Crop variety or hybrid and management from 2017 to 2020

Year	Activity or input	Low herbicide	Conventional herbicide	Low herbicide	Conventional herbicide
		Corn	**Corn**	**Soybean**	**Soybean**
2017	Hybrid or variety	Epley E1420	Epley E1420	Latham L2758 R2	Latham L2758 R2
	Herbicides applied (kg ai./ha)	tembotrione (0.049) applied May 31, interrow cultivated Jun. 7	PRE: thiencarbazone methyl (0.037), isoxaflutole (0.093)	PRE: flumioxazin (0.109); POST: glyphosate as potassium salt (1.249), acifluorfen (0.224)	PRE: flumioxazin (0.109); POST: glyphosate as potassium salt (1.249), acifluorfen (0.224)
	Total (kg a.i./ha)	0.049	0.13	1.581	1.581
	Weed sampling date	Sep. 5 and 6	Sep. 5 and 6	Sep. 6, 7 and 8	Sep. 6, 7 and 8
2018	Hybrid or variety	Epley E1420	Epley E1420	Latham L2758 R2	Latham L2758 R2
	Herbicides applied (kg ai./ha)	POST: tembotrione (0.054)	PRE: thiencarbazone methyl (0.037), isoxaflutole (0.092); POST: mesotrione (0.105), nicosulfuron (0.053)	PRE: flumioxazin (0.096); POST: glyphosate as potassium salt (1.540), lactofen (0.140)	PRE: flumioxazin (0.096); POST: glyphosate as potassium salt (1.540), lactofen (0.140)
	Total (kg a.i./ha)	0.054	0.287	1.776	1.776
	Weed sampling date	Sep. 11, 12, and 13	Sep. 11, 12, and 13	Sep. 17, 19, 20, and 21	Sep. 17, 19, 20, and 21
2019	Hybrid or variety	Epley E1730	Epley E1730	Latham 2684 L (Liberty Link)	Latham 2684 L (Liberty Link)
	Herbicides applied (kg ai./ha)	POST: tembotrione (0.049)	PRE: thiencarbazone methyl (0.037), isoxaflutole (0.092); POST: mesotrione (0.105), nicosulfuron (0.053)	PRE: flumioxazin (0.096); POST: glufosinate ammonium (0.594), clethodim (0.136)	PRE: flumioxazin (0.096); POST: glufosinate ammonium (0.594), clethodim (0.136)
	Total (kg a.i./ha)	0.049	0.287	0.826	0.826
	Weed sampling date	Sep. 17 and 18	Sep. 17 and 18	Sep. 30	Sep. 30
2020	Hybrid or variety	Epley E1730	Epley E1730	Latham 2684 L (Liberty Link)	Latham 2684 L (Liberty Link)
	Herbicides applied (kg ai./ha)	POST: tembotrione (0.051)	PRE: thiencarbazone methyl (0.037), isoxaflutole (0.092); POST: mesotrione (0.105), nicosulfuron (0.053)	PRE: flumioxazin (0.096); POST: glufosinate ammonium (0.594), clethodim (0.136)	PRE: flumioxazin (0.096); POST: glufosinate ammonium (0.594), clethodim (0.136)
	Total (kg a.i./ha)	0.051	0.287	0.826	0.826
	Weed sampling date	Sep. 14 and 15	Sep. 14 and 15	Sep. 16	Sep. 16
		Oat	**Oat**	**Alfalfa**	**Alfalfa**
2017	Hybrid or variety	IN09201	IN09201	Leafguard	Leafguard
	Weed sampling date	Sep. 25, 27, 28 and 29	Sep. 25, 27, 28 and 29	Sep. 25, 27, 28 and 29	Sep. 25, 27, 28 and 29
2018	Hybrid or variety	IN09201	IN09201	Leafguard	Leafguard
	Weed sampling date	Sep. 26, Oct.4, 15, 16, 18, and 19	Sep. 26, Oct.4, 15, 16, 18, and 19	Sep. 26, Oct.4, 15, 16, 18, and 19	Sep. 26, Oct.4, 15, 16, 18, and 19
2019	Hybrid or variety	IN09201	IN09201	Leafguard	Leafguard
	Weed sampling date	Sep. 23, 24, 25, and 26, Oct. 3, 4, 7, and 8	Sep. 23, 24, 25, and 26, Oct. 3, 4, 7, and 8	Sep. 23, 24, 25, and 26, Oct. 3, 4, 7, and 8	Sep. 23, 24, 25, and 26, Oct. 3, 4, 7, and 8
2020	Hybrid or variety	IN09201	IN09201	Leafguard	Leafguard
	Weed sampling date	Sep. 23, 24, and 29, Oct. 2, 6, 7, and 8	Sep. 23, 24, and 29, Oct. 2, 6, 7, and 8	Sep. 23, 24, and 29, Oct. 2, 6, 7, and 8	Sep. 23, 24, and 29, Oct. 2, 6, 7, and 8

Note: Corn was planted at 12950 seeds/ha, soybean at 56656 seeds/ha, oat 80.7 kg/ha, red clover and alfalfa at 19.1 kg/ha. No herbicide was applied in oat, red clover, and alfalfa. 'Belle' (in 2017) or 'Mammoth' (in 2018 - 2020) red clover was intercropped with oat in the 3-year rotation (O3). Alfalfa was intercropped with the

Weeds were not sown experimentally. Volunteer crops from a preceding crop season, such as a volunteer corn plant in a soybean plot or a soybean plant in an oat plot, were excluded from the assessment of weed community composition. Data were collected for individual species aboveground mass and density, total weed biomass and density, and crop yield. Weeds were surveyed four to six weeks before corn and soybean harvests, two to three weeks after oat harvest or the last hay cut of the season. The passage of a few weeks between oat and alfalfa harvest and weed surveys allowed physically damaged plants in those crops to grow back to recognizability per an identification guide developed by Uva et al. (1997).

Weed total density and aboveground mass Weed aboveground samples were collected from eight quadrats arranged in a 4x2 grid throughout each experimental unit (eu). The total surveyed area was 18.5 m^2/eu in corn and soybean (8 x 3.05 m^2) and 2.2 m^2/eu (8 x 0.25m^2 or 8 x 0.28m^2) in oat and alfalfa. Plants were identified to species.

Weed individual species relative and absolute abundance All the same-species plants from an eu were enumerated, dried, and weighed together to make single data points per eu. Individual species density and aboveground mass were presented for each crop identity to illustrate the community composition in each crop phase.

Ecological indices Aboveground weed mass reflects species competitiveness and density represents population size. Both species aboveground mass and density were used to calculate species diversity, evenness, and richness in each eu. Simpson's diversity, evenness, and richness indices were calculated in terms of stand density and aboveground mass. We evaluated eighteen weed communities, corresponding to nine crop identities crossed with two weed management regimes in corn.

Let:
S represent species richness (i.e., the number of species presented),
n_i represent density of the ith species (plants m^{-2}),
N represent density of all presented species (plants m^{-2}),
b_i represent aboveground mass of the ith species (kg m^{-2}),
B represent aboveground mass of all species, kg m^{-2}, and
p_{i_d} and p_{i_b} represent the proportional of density or aboveground biomass of the ith species.

Community diversity was evaluated with Simpson's index, $Simpson's\ D = \frac{1}{D} = \frac{1}{\sum p_i^2}$, because it is less sensitive to sample size and is useful to describe evenness (Nkoa et al., 2015). Simpson's evenness index was calculated with $\frac{\frac{1}{D}}{S}$. The p_i component in Simpson's diversity and evenness indices here was calculated with stand count ($\frac{n_i}{N}$) or biomass ($\frac{b_i}{B}$). Ideally, only one richness index is needed because it is the number of species presented. However, two ABUTH (*Abutilon theophrasti*) plants that were found in 2019 were too light to register on a scientific scale, resulting in zero weight for the species' aboveground mass. Therefore, the richness index was calculated for both stand and aboveground mass. The evenness index was thus calculated with the relevant richness index with regards to stand count and aboveground mass.

Crop yields Six 84-m long rows of corn and soybean (383 m^2) were harvested from each eu, for oat and alfalfa, whole plots were harvested (two adjacent subplots combined, 1530 m^2). Yields were adjusted to moisture concentrations of 155 g H_2O kg^{-1} for corn, at 130 g H_2O kg^{-1} for soybean, at 140 H_2O kg^{-1} for oat grain and 150 g H_2O kg^{-1} for alfalfa.

Model fitting Block, crop identity, weed management regime applied to the corn phase of a rotation (corn weed management), and the interaction of crop identity and corn weed management were considered fixed factors; year and the interaction between year and the fixed factors were considered random factors; and the residual was random by default. Block was treated as a fixed factor to control for the different field conditions across sections and reduce the variance between eu's (Dixon, 2016).

R version 4.1.2 (R Development Core Team, 2021) was used for all the data organization, manipulation, analysis, models diagnosis, and result presentation. Statistical tests were evaluated at an $\alpha = 0.05$ level of significance. All the response variables were natural logarithm (ln) transformed to meet homogeneity of variance requirement. Type III sums of squared error were calculated with the `emmeans` package's `joint_tests` function to accommodate unbalanced data with interaction (version 1.7.1-1, Lenth et al., 2021). The data were ln-transformed by adding the minimum non-zero value for each response to all values before analysis, but results were back-transformed for presentation. Degree of freedom adjustment was done with Satterthwaite's method. P-values adjustment was done with Tukey's method.

Stand diversity, stand evenness, stand richness, aboveground mass diversity, aboveground mass evenness, aboveground mass richness, total aboveground density, total aboveground mass, single species density, and single species aboveground mass were analyzed separately with a linear mixed-effects model, using the `lmer` function in the `lme4` package (version 1.1-27.1, Bates et al., 2021) according to the following model.

$$R_{ijklm} = \mu + B_i + C_j + H_k + CH_{jk} + Y_l + BY_{il} + YC_{lj} + YH_{lk} + YCH_{ljk} + BYC_{ijl} + \epsilon_{ijkl} \qquad (1)$$

where,

R is one of the aforementioned responses,
μ is the overall mean,
B is the block,
Y is the year,
C is the crop identity,
H is the corn weed management,
CH is the interaction between crop identity and corn weed management,
BY is the block within a year,
YC is interaction between crop identity and year,
YH is the interaction between year and corn herbicide,
YCH is the interaction between year, crop identity, and corn weed management,
BYC is the interaction between block, year, and crop identity, and
ϵ_{ijklm} is the residual.

The crop identity term in the right hand side of the model (Equation (1)) represents the main-plot effect of the experiment, which comprises of the crop species and the rotation to which it belonged. In this present study, "cropping system" is the combination of "rotation system" (2-year, 3-year, and 4-year) and herbicide regime in corn (low or conventional). With this model, we tested the following sets of hypotheses:

1) Weed stand diversity, stand evenness, stand richness, aboveground mass diversity, aboveground mass evenness, aboveground mass richness, total aboveground density, and total aboveground mass increased as cropping system diversity increased.

2) Weed stand diversity, stand evenness, stand richness, aboveground mass diversity, aboveground mass evenness, aboveground mass richness, total aboveground density, and total aboveground mass in the same crop species differed between cropping system .

3) Weed stand diversity, stand evenness, stand richness, aboveground mass diversity, aboveground mass evenness, aboveground mass richness, total aboveground density, and total aboveground mass in the same crop species differed between different crop types within the a given cropping system.

4) Weed single species density and aboveground mass of the most abundant species differed between rotations in the same crop species, differed across rotations, and differed between crop type within a given cropping system. Crop type represents growing condition, so corn and soybean were grouped as warm season crops, whereas oat and alfalfa were grouped as cool season crops.

The first set of hypotheses was tested by contrasting the responses in the 2-year rotation with those in the average of the 3-year and 4-year rotations and the responses in the 3-year rotation with those in the 4-year rotation. The second set of hypotheses was tested by contrasting the responses in the same crop species within different rotations. The third set of hypotheses was tested by contrasting the average responses in the warm season crops across rotations, in the cool season crops across rotations, in the warm season versus cool season crops within the same rotation, and between the warm season crops and the cool season crop(s) averaged over rotations. The same sets of contrasts used to evaluate weed community ecological indices, total weed aboveground mass, and total weed stand density were applied to data concerning the seven most abundant weed species. The fourth set of hypotheses was tested by contrasting individual weed species density and aboveground mass a) in the 2-year rotation versus the average of 3-year and 4-year rotations and in the 3-year versus 4-year rotation, b) in the same crop species or type across rotations, c) in different crop types within the same rotation, and d) in different crop types averaged over rotations.

A different set of linear mixed-effects model was used to analyze corn, soybean, and oat yields (`lme4` version 1.1-27.1, Bates et al., 2021):

$$R_{ijkm} = \mu + B_i + C_j + H_k + CH_{jk} + Y_l + BY_{il} + YC_{lj} + YH_{lk} + YRH_{lij} + BYC_{ilj} + \epsilon_{ijkl} \quad (2)$$

where,

R is the individual crop yield, and
all the terms in the right hand side of the model are as defined in Equation (1).

As each crop species was fitted with a model, the crop identity represents the rotation effect only. With this model (Equation (2)), we tested the hypotheses that the yield of the same crop species (corn, soybean, and oat) did not differ between rotations. Crop yields were then contrasted between rotations to examine the magnitude of any significant difference.

Results

A lack of any obvious bias in plots of residuals versus predicted values suggested that the analysis models fit the data well. Diagnosis plots made with `ggResidpanel` (version 0.3.0 Goode and Rey, 2019) are available in the Data Repository.

How did rotation system and corn weed management affect crop yields? Results of the experiment supported the first hypothesis that "diverse cropping systems, with reduced use of chemical herbicides, would provide weed control equal in effectiveness to the conventional approach in the 2-year rotation". Averaged over four years, soybean was the only crop whose yield was significantly affected by rotation (p-value = 0.0185, Table 2). Soybean yield was 16% higher in the 4-year rotation than in the 2-year rotation (p-value = 0.018). Crop yields in the experiment relative to averages for the state of Iowa and Boone County, where the experiment occurred, are presented in Figure 1. Corn weed management regime did not significantly affect crop yields (Table 2).

How did rotation system, crop species, and corn weed management affect community ecological indices? Crop identity (i.e., rotation system x crop phase combination) significantly affected weed aboveground mass diversity (p-value = 0.0007, Table 3A), evenness (p-value = 0.0003, Table 3B), and richness (p-values = 0.013); and stand density evenness (p-value = 0.0064) and richness (p-value = 0.0123, Table 3C). The ecological indices in each crop phrase, averaged over blocks, years and corn weed management are shown in Figure 2. The results of contrasts for the effects of rotation systems, rotation system within individual crops, and crop types on community ecological indices are shown in Tables 4 and 5. For all the significantly differences in ecological indices, crop types were more influential than rotations, with larger differences found across crop types than across rotations.

Table 2: Contrasts of rotation effect (expressed by Crop identity) on crop yields. The abbreviations on the contrast column are crop identities, which are the combinations of the first letter in crop species names and the rotation to which the crops belonged.

ANOVA					Comparison		
Source of variation	df1	df2	F.value	p.value	contrast	ratio	p.value
(A) - Corn							
Crop ID	2	6	3.190	0.1138	C2 vs C3	0.938	0.1882
Corn weed management	1	3	0.324	0.6088	C2 vs C4	0.929	0.1278
Crop ID x Corn weed management	2	6	2.205	0.1914	C3 vs C4	0.990	0.9507
(B) - Soybean							
Crop ID	2	6	8.224	0.0191	S2 vs S3	0.959	0.5499
Corn weed management	1	3	0.178	0.7018	S2 vs S4	0.862	0.0181
Crop ID x Corn weed management	2	6	0.623	0.5677	S3 vs S4	0.898	0.0670
(C) - Oat							
Crop ID	1	2	1.138	0.3979	O3 vs O4	0.913	0.3979

Note: Corn weed management: low herbicide or conventional. Crop ID: crop species and the cropping system to which each belong: C2 - corn in the 2-year rotation, C3 - corn in the 3-year rotation, C4 - corn in the 4-year rotation, S2 - soybean in the 2-year rotation, S3 - soybean in the 3-year rotation, S4 - soybean in the 4-year rotation, O3 - oat in the 3-year rotation, and O4 - oat in the 4-year rotation.

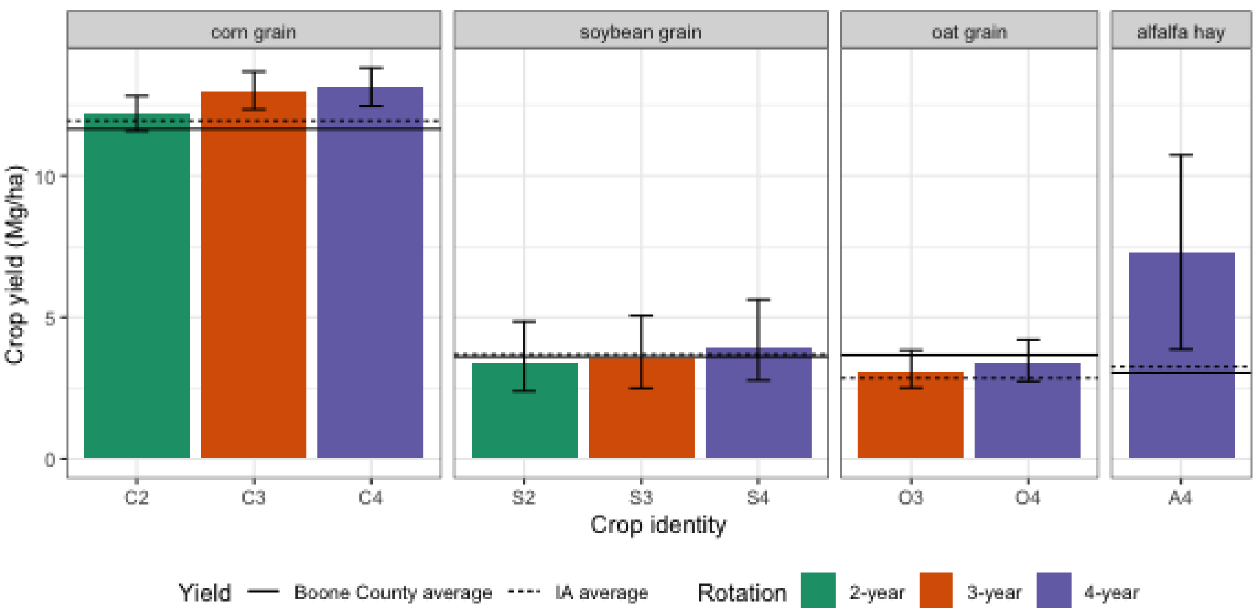

Figure 1: Mean crop yields by rotation from 2017 to 2020. The color-coded bars show crop yields (Mg ha^{-}1) in the experiment plots. The error bars show the 95% confidence intervals. The solid horizontal lines show mean yields for Iowa and dashed lines show mean yields for Boone County. Corn, soybean, and alfalfa yields in the experiment were averaged over four years, oat grain yields in the experiment were averaged over 2017, 2019, and 2020 because in 2018 oat was harvested for hay. Boone County and Iowa hay yields were averaged over 2017 and 2018 because 2019 and 2020 yields were not available at this writing.

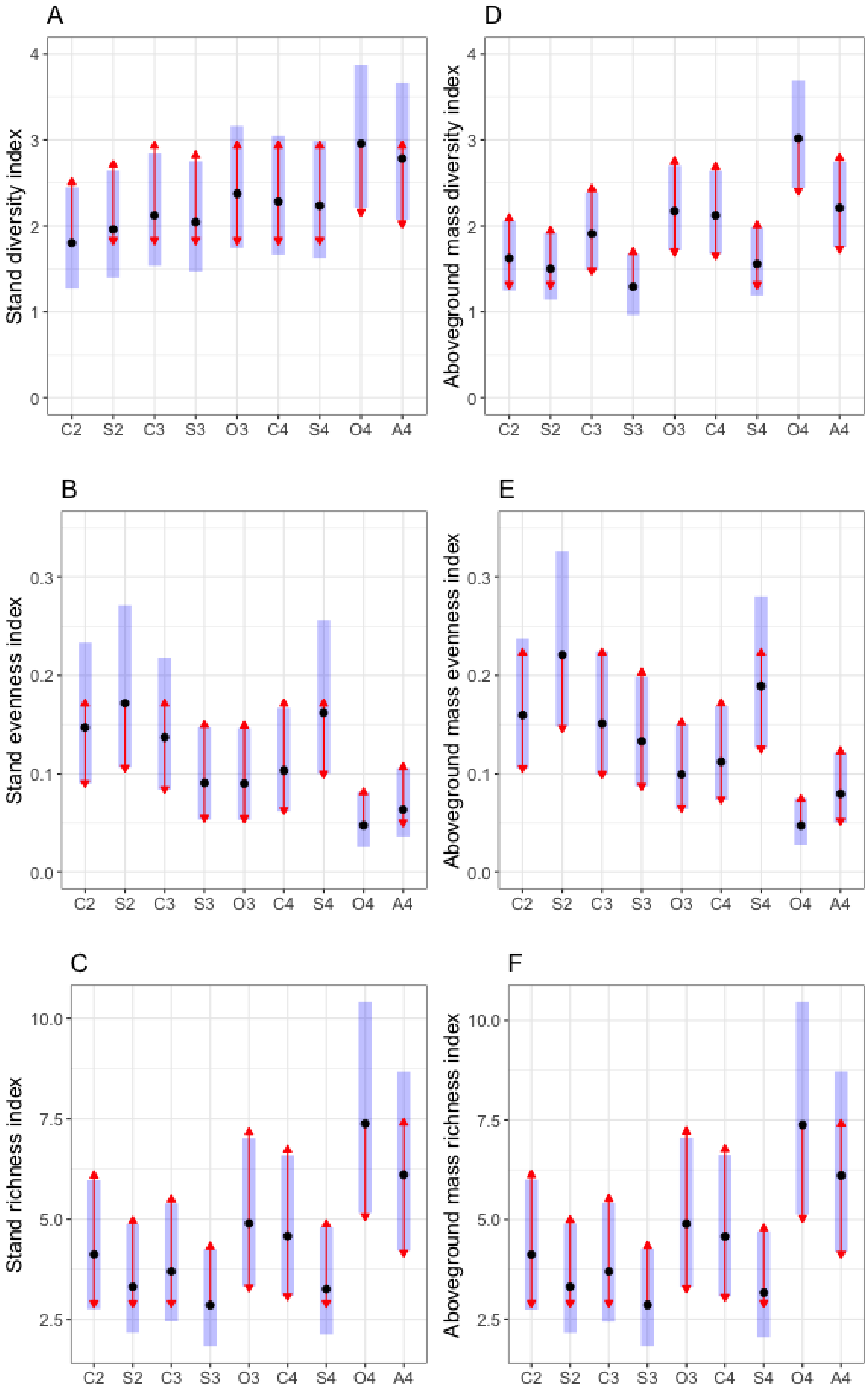

Figure 2: Weed community stand diversity (A), evenness (B), and richness (C) and community aboveground diversity (D), evenness (E), and richness (F). The abbreviations on the x-axis are crop identities, which are the combinations of the first letter in crop species names and the rotation to which the crops belonged (C2 - corn in the 2-year rotation, C3 - corn in the 3-year rotation, C4 - corn in the 4-year rotation, S2 - soybean in the 2-year rotation, S3 - soybean in the 3-year rotation, S4 - soybean in the 4-year rotation, O3 - oat in the

Table 3: ANOVAs of crop identity, corn weed management, and their interactive effects on weed community ecological indices

			Stand density		Aboveground mass	
Source of variation	df1	df2	F.value	p.value	F.value	p.value
(A) - Community diversity						
Crop ID	8	24	1.25	0.3116	5.22	0.0007
Corn weed management	1	3	0.21	0.6804	0.47	0.5439
Crop ID x Corn weed management	8	24	0.54	0.8182	1.35	0.2659
(B) - Community evenness						
Crop ID	8	24	3.66	0.0064	5.87	0.0003
Corn weed management	1	3	0.24	0.6589	0.01	0.9414
Crop ID x Corn weed management	8	24	0.74	0.6547	0.47	0.8632
(C) - Community richness						
Crop ID	8	24	3.23	0.0123	3.19	0.0130
Corn weed management	1	3	1.32	0.3330	1.59	0.2959
Crop ID x Corn weed management	8	24	0.71	0.6803	0.86	0.5635

Note: Corn weed management: low herbicide or conventional. Crop ID: crop species and the cropping system to which each belong: C2 - corn in the 2-year rotation, C3 - corn in the 3-year rotation, C4 - corn in the 4-year rotation, S2 - soybean in the 2-year rotation, S3 - soybean in the 3-year rotation, S4 - soybean in the 4-year rotation, O3 - oat in the 3-year rotation, and O4 - oat in the 4-year rotation, and A4 - alfalfa in the 4-year rotation.

In general, the hypothesis that "weed communities in the more diverse cropping systems are more diverse" was supported.

Averaged over crop phases within each rotation system (Table 4A), the weed stand diversity index for the 3-year and 4-year rotation systems was comparable with that in the 2-year rotation (p-values = 0.0535 and 0.1575). For the individual crops (Table 4B), the weed stand density diversity index was comparable among rotations (p-values > 0.05). For different crop types (Table 4C), the weed stand density diversity index was significantly different between the average for the cool season crops (O3, O4, and A4) and the average for the warm season crops (C2, S2, C3, S3, C4, and S4) (p-value = 0.0145), but similar between the warm season and cool season crops in the same rotations (p-values = 0.4666 and 0.0987). The weed stand density diversity index was similar between oat and alfalfa (p-value = 0.7762).

Averaged over crop phases within the same rotation (Table 5A), the weed aboveground mass diversity index was significantly different between the 2-year rotation and the average of the 3-year and 4-year rotations (p-value = 0.0148), and between the 3-year and 4-year rotations (p-value = 0.0209). Averaged over the corn and soybean phases within the same rotation (Table 5A), the weed aboveground mass diversity index was similar between rotations (p-values = 0.4217 and 0.2426). For the individual crops (Table 4B), the weed aboveground mass diversity index was comparable across rotations, except for oat (p-value = 0.0351). For different crop types (Table 4C), the weed aboveground mass diversity index was significantly different between the cool season crops and warm season crops averages (p-values < 0.0001) and between the cool season and warm season crops within the same rotation (p-values = 0.034 and 0.0037). The weed aboveground mass diversity index was comparable between oat and alfalfa (p-value = 0.2583).

The hypothesis that "weed communities in the more diverse cropping systems are more even" was partially supported (Figure 2B and E). However, a lower community evenness index can occur because the presence of rarer species decreases the overall evenness index (Stirling and Wilsey, 2001). More details to support this concept are presented later (Figure 3C and D).

Averaged over crop phases within the same rotation (Table 4A), the weed stand density evenness index was significantly different between the 2-year rotation and the average of the 3-year and 4-year rotations (p-value

= 0.006), but comparable between the 3-year and 4-year rotations (p-value = 0.2802). Averaged over the corn and soybean phases within the same rotation (Table 4A), the weed stand density evenness index was comparable between rotations (p-values = 0.1539 and 0.5031). For the individual crops (Table 4B), the weed stand density evenness index was comparable between rotations (p-values > 0.05). For different crop types (Table 4C), the weed stand density evenness index was significantly different between the cool season crops average and the warm season crops average (p-value = 0.0002) and between the cool season and warm season crop in the 4-year rotation (p-value = 0.0012), but similar between the warm season and cool season crops in the 3-year rotation (p-values = 0.4418). The weed stand density evenness index was comparable between oat and alfalfa (p-value = 0.8986).

Averaged over crop phases within the same rotation (Table 5A), the weed aboveground mass evenness index was significantly different between the 2-year rotation and the average of 3-year and 4-year rotations (p-value = 0.0012), but similar between the 3-year and 4-year rotations (p-value = 0.0802). Averaged over the corn and soybean phases within the same rotation (Table 5A), weed aboveground mass evenness index was comparable between rotations (p-values = 0.1081 and 0.8682). For the individual crops (Table 4B), the weed aboveground mass evenness index was comparable across rotations (p-values > 0.05), except for oat (p-value = 0.0189). For different crop types (Table 5C), the weed aboveground mass evenness index was significantly different between the cool season crops average and the warm season crops average (p-value < 0.0001) and between the cool season and warm season crops in the 4-year rotation (p-value = 0.0002), but comparable between the warm season and cool season crops in the 3-year rotation (p-values = 0.141). The weed aboveground mass evenness index was comparable between oat and alfalfa (p-value = 0.5911).

The hypothesis that "the weed communities in the more diverse cropping systems are more species-rich" was supported.

Averaged over crop phases within the same rotation (Table 4A), the weed stand density richness index was comparable in the 2-year rotation and in the average of the 3-year and 4-year rotations (p-values = 0.1819), but significantly different between the 3-year and 4-year rotation (p-value = 0.0257). Averaged over the corn and soybean phases within the same rotation (Table 4A), weed aboveground mass richness index was comparable between the 2-year rotation and the 3-year and 4-year rotations average (p-value = 0.7996) and between the 3-year and 4-year rotations (p-value = 0.3469). For individual crops (Table 4B), the weed stand density richness index was comparable between rotations (p-values > 0.05). For different crop types (Table 4C), the weed stand density richness index was significantly different between the cool season crops average and the warm season crops average (p-value = 0.0003) and between the cool season and warm season crops in the 4-year rotation (p-value = 0.0034), but comparable between the warm season and cool season crops in the 3-year rotation (p-values = 0.0725). The weed stand density richness index was comparable between oat and alfalfa (p-value = 0.9499).

Averaged over crop phases within the same rotation (Table 5A), the weed aboveground mass richness index was comparable in the 2-year rotation and in the average of the 3-year and 4-year rotations (p-values = 0.1967), but significantly different between the 3-year and 4-year rotations (p-value = 0.0309). Averaged over the corn and soybean phases within the same rotation (Table 5A), the weed aboveground mass richness index was comparable between the 2-year rotation and the 3-year and 4-year rotations average (p-value = 0.7694) and between the 3-year and 4-year rotations (p-value = 0.393). For the same crop types, (Table 5B), the weed aboveground mass richness index was comparable across rotations (p-values > 0.05). For different crop types (Table 5C), the weed aboveground richness index was significantly different between the cool season and warm season crop averages (p-value = 0.0003) and between the cool season and warm season crops in the 4-year rotation (p-value = 0.0766), but comparable between the cool season and warm season crops in the 3-year rotation (p-value = 0.0766). The weed aboveground mass richness index was comparable between oat and alfalfa (p-value = 0.9506).

General description of the weed flora Overall, 34 weed species were identified during the four years of data collection (Table 6). Combined over four years of data, seven weed species, SETFA (*Setaria faberi*), AMATA (*Amaranthus tuberculatus*), CHEAL (*Chenopodium album*), DIGSA (*Digitaria sanguinalis*), ECHCG (*Echinochloa crus-galli*), SETLU (*Setaria glauca*), and TAROF (*Taraxacum officinale*) made up 94.4% of the

Table 4: Weed stand density ecological indices contrast significance. The abbreviations on the contrast column are crop identities, which are the combinations of the first letter in crop species names and the rotation to which the crops belonged.

	Diversity index		Evenness index		Richness index	
Contrast	ratio	p.value	ratio	p.value	ratio	p.value
(A) - Rotation system effects						
[(C2+S2)/2] vs [(C3+S3+O3+C4+S4+O4+A4)/7]	0.85	0.0535	1.60	0.0060	0.86	0.1819
[(C3+S3+O3)/3] vs [(C4+S4+O4+A4)/4]	0.90	0.1575	1.18	0.2802	0.77	0.0257
[(C2+S2)/2] vs [(C3+S3+C4+S4)/4]	0.91	0.2749	1.28	0.1539	1.03	0.7996
[(C3+S3)/2] vs [(C4+S4)/2]	0.95	0.5824	0.88	0.5031	0.87	0.3469
(B) - Rotation system effects within individual crops						
C2 vs [(C3+C4)/2]	0.88	0.2836	1.20	0.4406	1.00	0.9985
C3 vs C4	0.95	0.7231	1.28	0.3757	0.84	0.3966
S2 vs [(S3+S4)/2]	0.94	0.6331	1.36	0.2065	1.06	0.7212
S3 vs S4	0.94	0.6711	0.60	0.0746	0.91	0.6260
O3 vs O4	0.85	0.2716	1.66	0.0757	0.70	0.0912
(C) - Crop type effects						
[(O3+O4+A4)/3] vs [(C2+S2+C3+S3+C4+S4)/6]	1.20	0.0145	0.55	0.0002	1.53	0.0003
O3 vs [(C3+S3)/2]	1.09	0.4666	0.83	0.4418	1.38	0.0725
[(O4+A4)/2] vs [(C4+S4)/2]	1.19	0.0987	0.49	0.0012	1.58	0.0034
[(O3+O4)/2] vs A4	0.97	0.7762	1.03	0.8986	0.99	0.9499

Note: C2 - corn in the 2-year rotation, C3 - corn in the 3-year rotation, C4 - corn in the 4-year rotation, S2 - soybean in the 2-year rotation, S3 - soybean in the 3-year rotation, S4 - soybean in the 4-year rotation, O3 - oat in the 3-year rotation, O4: oat in the 4-year rotation, and A4 - alfalfa in the 4-year rotation

Table 5: Weed aboveground mass ecological indices contrast significance. The abbreviations on the contrast column are crop identities, which are the combinations of the first letter in crop species names and the rotation to which the crops belonged.

	Diversity index		Evenness index		Richness index	
Contrast	ratio	p.value	ratio	p.value	ratio	p.value
(A) - Rotation system effects						
[(C2+S2)/2] vs [(C3+S3+O3+C4+S4+O4+A4)/7]	0.85	0.0148	1.65	0.0012	0.86	0.1967
[(C3+S3+O3)/3] vs [(C4+S4+O4+A4)/4]	0.87	0.0209	1.27	0.0802	0.78	0.0309
[(C2+S2)/2] vs [(C3+S3+C4+S4)/4]	0.95	0.4217	1.28	0.1081	1.04	0.7694
[(C3+S3)/2] vs [(C4+S4)/2]	0.91	0.2426	0.97	0.8682	0.88	0.3930
(B) - Rotation system effects within individual crops						
C2 vs [(C3+C4)/2]	0.87	0.1425	1.20	0.3825	1.00	0.9985
C3 vs C4	0.93	0.5084	1.31	0.2780	0.84	0.4035
S2 vs [(S3+S4)/2]	1.03	0.7219	1.36	0.1543	1.08	0.6801
S3 vs S4	0.90	0.3166	0.72	0.1905	0.93	0.7075
O3 vs O4	0.79	0.0351	1.83	0.0189	0.70	0.0957
(C) - Crop type effects						
[(O3+O4+A4)/3] vs [(C2+S2+C3+S3+C4+S4)/6]	1.30	<.0001	0.51	<.0001	1.54	0.0003
O3 vs [(C3+S3)/2]	1.23	0.0340	0.73	0.1410	1.38	0.0766
[(O4+A4)/2] vs [(C4+S4)/2]	1.27	0.0037	0.48	0.0002	1.60	0.0032
[(O3+O4)/2] vs A4	1.11	0.2583	0.89	0.5911	0.99	0.9506

Note: C2 - corn in the 2-year rotation, C3 - corn in the 3-year rotation, C4 - corn in the 4-year rotation, S2 - soybean in the 2-year rotation, S3 - soybean in the 3-year rotation, S4 - soybean in the 4-year rotation, O3 - oat in the 3-year rotation, O4 - oat in the 4-year rotation, and A4 - alfalfa in the 4-year rotation total weed density and 94.0% of the total weed biomass (Figure 3C and D).

How did rotation, crop species, and corn weed management affect weed community density and growth? Crop identity had a significant effect on weed community stand density (p-value < 0.0001) and weed aboveground mass (p-value $= 0.0057$), but corn weed management and its interaction with crop identity did not have a significant effect on weed community stand density or biomass (p-values > 0.05) (Table 4 and 5). Weed total stand density and aboveground mass in each crop identity category, averaged over blocks, years, and corn weed management regimes, are presented in Figure 3A and B. Contribution by the dominant species are presented in Figure 3C and D. Contrasts for the effects of rotation systems, rotation system within individual crops, and crop types on community stand density and aboveground mass are shown in Table 7C.

Weed community density and aboveground mass of the 3-year and 4-year systems averages were comparable to those of the 2-year system (p-values $= 0.058$ and 0.9451, Table 7B1). The weed density in the 4-year rotation was 2.5 fold greater than in the 3-year rotation (p-value $= 0.0368$), but the aboveground mass was comparable between the 3-year and 4-year rotations.

For the individual crops (Table 7B2), increased rotation diversity tended to decrease weed abundance in corn and soybean and increase weed abundance in oat, but these trends were not significant (p-values $= 0.6354$ and 0.4041 for corn, 0.1834 and 0.0739 for soybean, and 0.3955 and 0.335 for oat). The patchiness of weeds, which was reflected in the high standard error values, might have caused the lack of significance for these inconclusive trends.

For different crop types (Table 7B3), weed community density and aboveground mass were comparable between the warm season crops (corn and soybean, p-values $= 0.2032$, 0.3426, 0.065, and 0.1274) and between the cool season crops (oat and alfalfa, p-values $= 0.774$ and 0.687). Overall, the average weed community density in the cool season crops was 26-fold greater than that in the warm season crops (p-value < 0.0001), and the average weed aboveground mass in cool season crops was 16-fold greater than that in warm season crops (p-value $= 0.0001$). In the 3-year rotation, the weed stand community stand in oat (O3) was 11.5-fold greater than the average in corn and soybean (C3 and S3) (p-value $= 0.0012$), but the weed community total aboveground mass was comparable between O3 and the average of the C3 and S3 phases (p-value $= 0.1502$). In the 4-year rotation, the weed community stand density in the average of oat and alfalfa (O4 and A4) was 36-fold greater than the average of the corn (C4) and soybean (S4) phases (p-value < 0.0001), and the average weed biomass for the O4 and A4 phases was 29-fold greater than for the C4 and S4 phases (p-value < 0.0001).

How did rotation, crop species, and corn weed management affect individual weed species abundance? The stand density and aboveground mass of the seven most abundant weed species are shown in Figure 3. The effects of crop identity (i.e., rotation system crossed with crop species), corn weed management, and their interaction on the seven most abundant weeds in the present study are shown in Table 8. No interactive effects between crop identity and corn weed management were seen in any of those seven species' density or aboveground mass. The main effects of crop identity and corn weed management on stand density and aboveground mass differed by species.

The hypothesis that "including oat and alfalfa in rotations with corn and soybean will reduce the density and aboveground mass of noxious weed species in corn and soybean" was partially supported. Among the seven most abundant weed species, the stand densities were all affected by crop identity, but the aboveground mass was affected by crop identity for only four out of seven species (Table 8).

Since increased weed stand density and aboveground mass were not correlated with increased crop yield loss, the magnitude of differences in individual weed density and aboveground mass are not presented here. Significance of differences in individual species density and aboveground mass (p-values) are presented to illustrate community composition shift. Individual species stand density and aboveground mass data were combined over four years and four blocks.

Table 6: List of weed species (in alphabetical order) found from 2017 to 2020 field seasons.

Bayer code	Scientific name	Life cycle
(A) - Dicotyledon species		
ABUTH	*Abutilon theophrasti* Medicus	annual
AMARE	*Amaranthus retrofelxus* L.	summer annual
AMATA	*Amaranthus tuberculatus* (Moq.) Sauer var. *rudis*	summer annual
AMBEL	*Ambrosia artemissifolia* L.	erect, branching, summer annual
ARFMI	*Arctium minus* (Hill) Bernh.	biennial
CHEAL	*Chenopodium album* L.	erect summer annual
CIRAR	*Cirsium arvense* (L.) Scop.	rhizomatous perennial
CIRVU	*Cirsium vulgare* (Savi) Tenore	biennial
EPHHT	*Euphorbia humistrata* Engelm. ex Gray	mat-forming summer annual
EPHMA	*Euphorbia maculata* L.	mat-forming summer annual
EUPHY	*Eupatorium hyssopifolium* L.	summer annual
MORAL	*Morus alba* L.	perennial shrub
PHYSU	*Physalis subglabrata* Mackenz. and Bush	rhizomatous perennial
PLAMA	*Plantago major* L.	rosette-forming perennial
POLPY	*Polygonum pensylvanicum* L.	ascending much-branched summer annual
POPDE	*Polygonum perfoliatum* L.	spiny summer annual vine
POROL	*Portulaca oleracea* L.	prostrate mat-forming summer annual
SOLPT	*Solanum ptycanthum* Dun.	erect branching summer annual
SONAR	*Sonchus arvensis* L.	rhizomatous perennial
TAROF	*Taraxacum officinale* Weberin Wiggers	tap-rooted perennial
(B) - Monocotyledon species		
AGRRE	*Elytrigia repens* (L.) Nevski	rhizomatous perennial
BROTE	*Bromus tectorum* L.	summer or winter annual
CCHPA	*Cenchrus longispinus* (Hack.) Fern.	summer annual
CONAR	*Convolvulus arvensis* L.	rhizomatous perennial
CYPES	*Cyperus esculentus* L.	rhizomatous perennial
DACGL	*Dactylis glomerata* L.	chump-forming perennial
DIGSA	*Digitaria sanguinalis* (L.) Scop.	summer annual
ECHCG	*Echinochloa crus – galli* (L.) Beauv.	summer annual
ERBVI	*Eriochloa villosa* (Thunb.) Kunth	erect summer annual
FESSP	*Festuca* spp.	clump-forming perennial
PANCA	*Panicum capillare* L.	summer annual
PANDI	*Panicum dichotomiflorum* Michx.	summer annual
SETFA	*Setaria faberi* Herrm.	clump-forming, erect summer annual
SETLU	*Setaria glauca* (L.) Beauv.	clump-forming, erect summer annual

Table 7: Community density and aboveground mass ANOVA and contrasts. The abbreviations in the contrast column are crop identities, which are the combinations of the first letter in crop species names and the rotation to which the crops belonged.

			Stand density		Aboveground mass	
Source of variation	df1	df2	F.value	p.value	F.value	p.value
(A) - ANOVA						
Crop ID	8	24	12.22	<.0001	3.74	0.0057
Corn weed management	1	3	2.13	0.2402	0.02	0.8900
Crop ID x Corn weed management	8	24	1.66	0.1613	0.99	0.4660
Contrasts ratio p.value ratio p.value						
(B1) - Rotation system effects						
[(C2+S2)/2] vs [(C3+S3+O3+C4+S4+O4+A4)/7]			0.42	0.0580	0.96	0.9451
[(C3+S3+O3)/3] vs [(C4+S4+O4+A4)/4]			0.40	0.0368	0.42	0.1712
(B2) - Rotation system effects within individual crops						
C2 vs [(C3+C4)/2]			1.38	0.6354	2.30	0.4041
C3 vs C4			0.59	0.4969	0.73	0.7853
S2 vs [(S3+S4)/2]			2.49	0.1834	6.25	0.0739
S3 vs S4			1.19	0.8248	1.04	0.9731
O3 vs O4			0.51	0.3955	0.33	0.3350
(B3) - Crop type effects						
[(C2+S2)/2] vs [(C3+S3+C4+S4)/4]			1.85	0.2032	3.79	0.0665
[(C3+S3)/2] vs [(C4+S4)/2]			1.69	0.3426	3.54	0.1274
[(O3+O4+A4)/3] vs [(C2+S2+C3+S3+C4+S4)/6]			26.10	<.0001	16.00	0.0001
O3 vs [(C3+S3)/2]			11.50	0.0012	4.29	0.1502
[(O4+A4)/2] vs [(C4+S4)/2]			35.90	<.0001	28.70	0.0003
[(O3+O4)/2] vs A4			0.80	0.7440	1.49	0.6870

Note: C2 - corn in the 2-year rotation, C3 - corn in the 3-year rotation, C4 - corn in the 4-year rotation, S2 - soybean in the 2-year rotation, S3 - soybean in the 3-year rotation, S4 - soybean in the 4-year rotation, O3 - oat in the 3-year rotation, and O4 - oat in the 4-year rotation.

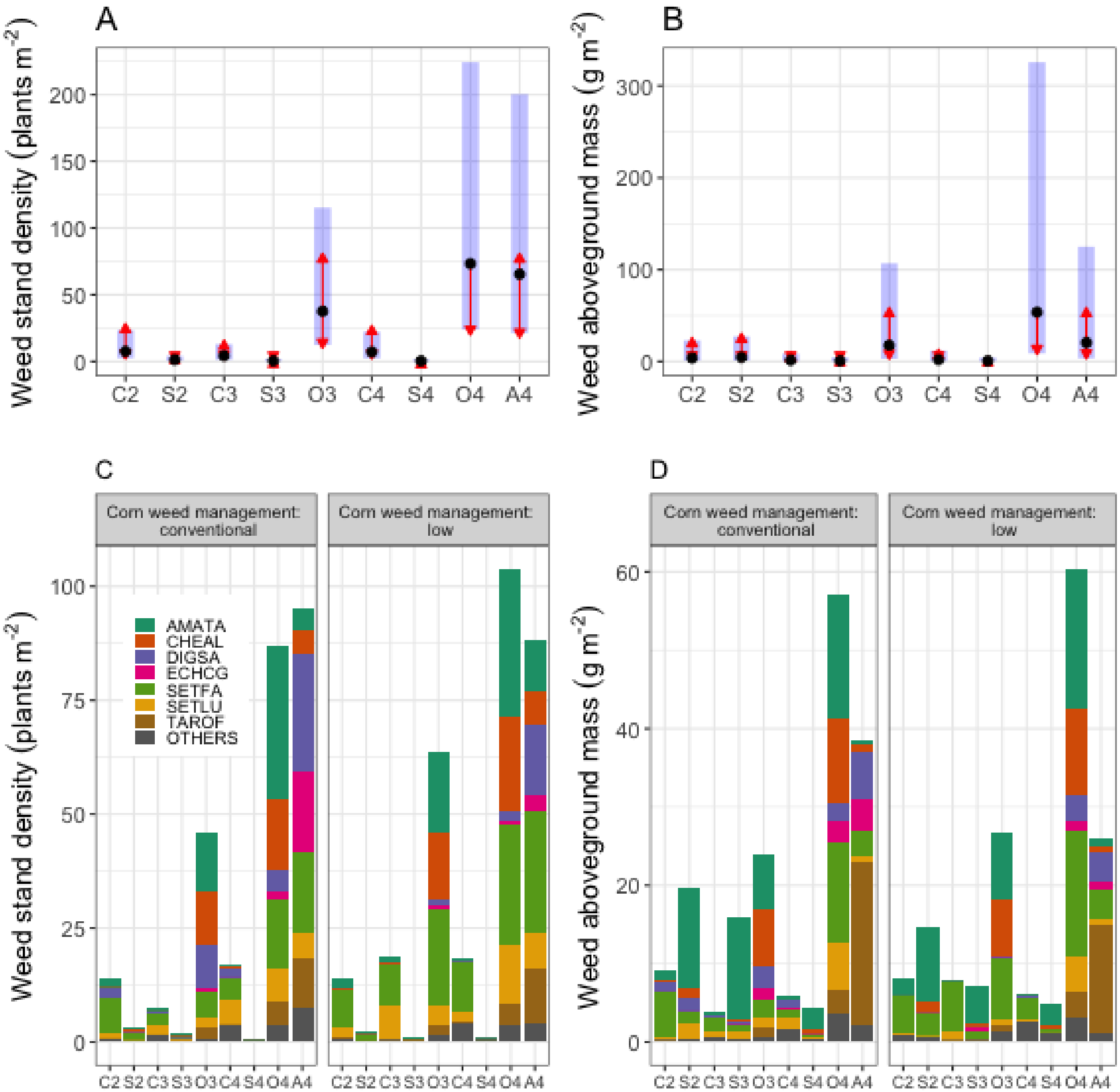

Figure 3: In panels A and B: weed community stand density and aboveground mass were averaged over four blocks, four years, and two corn weed management regimes; the black dots are estimated marginal means; the blue bars are 95% confidence intervals; the red arrows reflect the comparisons among means; overlapping arrows indicate non-significant differences. In panels C and D: the contribution of the seven most abundant weed species and the rarer species (species ordered eighth and above grouped in OTHERS) in each crop identity, averaged over four blocks and four years, are ordered alphabetically. The abbreviations on the x-axis are crop identities, which are the combinations of the first letter in crop species names and the rotation to which the crops belonged (C2 - corn in the 2-year rotation, C3 - corn in the 3-year rotation, C4 - corn in the 4-year rotation, S2 - soybean in the 2-year rotation, S3 - soybean in the 3-year rotation, S4 - soybean in the 4-year rotation, O3 - oat in the 3-year rotation, O4 - oat in the 4-year rotation, and A4 - alfalfa in the 4-year rotation.) The less abundant weed species which made up 6% of the whole community are grouped in OTHERS. The means displayed on panels A and B were estimated marginal means, calculated based on the analysis model (with `emmip` function) but the means displayed on panels C and D were arithmetic means, calculated from the data so they are slightly different.

Table 8: Treatment effects on the abundance of the most population and vigorous weed species, listed alphabetically. All the other weeds species were grouped into OTHERS

			Stand density		Aboveground mass	
Source of variation	df1	df2	F.value	p.value	F.value	p.value
(A) - AMATA						
Crop ID	8	24	3.72	0.0058	1.52	0.2016
Corn weed management	1	3	0.73	0.4566	4.19	0.1333
Crop ID x Corn weed management	8	24	0.96	0.4886	1.09	0.4052
(B) - CHEAL						
Crop ID	8	24	22.06	<.0001	15.53	<.0001
Corn weed management	1	3	2.10	0.2430	0.56	0.5097
Crop ID x Corn weed management	8	24	1.59	0.1808	1.07	0.4180
(C) - DIGSA						
Crop ID	8	24	15.52	<.0001	8.14	<.0001
Corn weed management	1	3	21.52	0.0189	16.44	0.0270
Crop ID x Corn weed management	8	24	1.25	0.3126	0.78	0.6237
(D) - ECHCG						
Crop ID	8	24	2.61	0.0328	2.20	0.0645
Corn weed management	1	3	5.80	0.0952	4.84	0.1150
Crop ID x Corn weed management	8	24	1.16	0.3615	1.04	0.4348
(E) - SETFA						
Crop ID	8	24	8.78	<.0001	4.22	0.0028
Corn weed management	1	3	20.91	0.0196	13.96	0.0334
Crop ID x Corn weed management	8	24	0.70	0.6892	1.04	0.4371
(F) - SETLU						
Crop ID	8	24	3.09	0.0154	1.33	0.2774
Corn weed management	1	3	4.44	0.1257	3.28	0.1681
Crop ID x Corn weed management	8	24	1.11	0.3930	0.83	0.5875
(G) - TAROF						
Crop ID	8	24	49.63	<.0001	35.81	<.0001
Corn weed management	1	3	0.61	0.4914	0.33	0.6067
Crop ID x Corn weed management	8	24	0.74	0.6553	1.20	0.3382
(H) - OTHERS						
Crop ID	8	24	4.76	0.0014	2.35	0.0503
Corn weed management	1	3	1.99	0.2533	2.27	0.2288
Crop ID x Corn weed management	8	24	0.07	0.9997	0.43	0.8939

Note: Corn weed management: low herbicide or conventional. C2 - corn in the 2-year rotation, C3 - corn in the 3-year rotation, C4 - corn in the 4-year rotation, S2 - soybean in the 2-year rotation, S3 - soybean in the 3-year rotation, S4 - soybean in the 4-year rotation, O3 - oat in the 3-year rotation, O4 - oat in the 4-year rotation, and A4 - alfalfa in the 4-year rotation.

Averaged over crop identity, DIGSA and SETFA stand density and aboveground mass were affected by corn weed management (p-values = 0.0189 and 0.0196, Table 8). Averaged over corn weed management regimes, the differences in weed species stand density and aboveground mass were observed more often between crop types (Tables 9B and C, and Tables @ref(tab:10B and C)) than for individual crops across rotations (Tables 9A and 10A). The main-plot effects concerning crop identity on individual species responses are elaborated below.

The cool season crops were responsible for AMATA stand density differences, but those differences were not strong enough to be apparent between rotation averages. AMATA stand density and aboveground mass were comparable among all rotation systems averaged over crop phases (p-values > 0.05), among rotations for the same crop species (p-values > 0.05), and within the same crop type across rotations (p-values > 0.05). Averaged over the same crop types (warm season or cool season), AMATA stand density was significantly different in cool season versus warm season crops (p-value = 0.0001), but AMATA aboveground mass was comparable (p-value = 0.0906) in cool season and warm season crops. Within the same rotation, AMATA stand density was greater in the cool season than in the warm season crops (p-values 0.0143, and 0.0003), but AMATA aboveground mass was comparable in these crop environments (p-values = 0.2355, and 0.0493).

The cool season crops, especially oat were responsible for CHEAL stand density and aboveground mass differences between rotation averages. CHEAL stand density and aboveground mass were 11-fold (p-value = 0.0001) and 96-fold (p-value = 0.0001) greater in oat than in alfalfa. CHEAL stand density and aboveground mass were significantly different between the 2-year rotation and the average of the 3-year and 4-year rotations, but comparable between the 3-year and 4-year rotations (p-values = 0.9195 and 0.6114). CHEAL stand density and aboveground mass were comparable across rotations for the same crop species (p-values > 0.05) and within the warm season crops (p-values > 0.05), but significantly different across crop types overall (p-values < 0.0001), between the warm season and cool season crops of the same rotation (p-values = 0.0001), and within the cool season crops (oat versus alfalfa).

The cool season crops, especially alfalfa were responsible for DIGSA stand density and aboveground mass differences between rotation averages. DIGSA stand density and aboveground mass were 14-fold (p-value = 0.0001) and 33-fold (p-value = 0.0001) greater in alfalfa than in oat. DIGSA stand density significantly was different between the 2-year rotation and the average of the 3-year and 4-year rotations (p-value = 0.0072) and between the 3-year and 4-year rotation (p-value < 0.0001). DIGSA aboveground mass was comparable between the 2-year and the average of the 3-year and 4-year rotations (p-value = 0.1098), but significantly different between the 3-year and 4-year rotations (p-value = 0.0001). DIGSA stand density and aboveground mass were comparable across rotations for the same crop species (p-values > 0.05), except for oat (p-values = 0.0062 and 0.0032). Within the 3-year rotation, DIGSA stand density was comparable among crop phases (p-value = 0.0603), but DIGSA aboveground mass was significantly different between oat and the average of corn and soybean phases (p-value < 0.0001). DIGSA stand density and aboveground mass were significantly different across crop types overall, between the warm season and cool season crops of the 4-year rotation (p-values = 0.0001), and within the cool season crops (oat versus alfalfa) (p-values < 0.0001).

ECHCG responses generally were similar to those of AMATA. ECHCG stand density and aboveground mass were comparable between all rotation averages (p-values > 0.05), across rotations for the same crop species (p-values > 0.05), within the same crop type across rotations (p-values > 0.05), and within the 3-year rotation (p-values > 0.05). Averaged over the same crop types, ECHCG stand density and aboveground mass were significantly different in cool season versus warm season crops (p-value = 0.0003 and 0.0012). Within the 4-year rotation, ECHCG stand density and aboveground mass were greater in the cool season than in the warm season crops (p-values 0.0014, and 0.0031).

The cool season crops were responsible for SETFA stand density and aboveground mass differences, but those differences were not strong enough be apparent between rotation averages. SETFA stand density and aboveground mass were comparable across all rotation averages (p-values > 0.05), across rotations for the same crop species (p-values > 0.05), within the warm season crops across rotations (p-values > 0.05), and within the cool season crops (p-values > 0.05). Averaged over the same crop types, SETFA stand density and aboveground mass were significantly different in cool season versus warm season crops (p-value < 0.0001 and p-value = 0.0008). Within the same rotation, SETFA stand density and aboveground mass were greater in the cool season than in the warm season crops (p-values = 0.001, 0.018, 0.0001, and 0.0045).

SETLU stand density and aboveground mass were comparable in most pairs of comparison (p-values > 0.05), with the exception in the warm season versus cool season density (p-value = 0.0404).

The cool season crops, especially oat were responsible for TAROF stand density and aboveground mass differences across rotation averages. TAROF stand density and aboveground mass were 6-fold (p-value < 0.0001) and 20-fold (p-value = 0.0001) greater in oat than in alfalfa. TAROF stand density and aboveground mass were significantly different in the 2-year versus the average of the 3-year and 4-year rotations, and between the 3-year and 4-year rotations (p-values < 0.0001). TAROF stand density and aboveground mass were comparable among the warm season crops across rotations and within the same crops across rotations (p-values > 0.05), except in oat (p-values < 0.0001). TAROF stand density and aboveground mass were significantly different across crop types overall (p-values < 0.0001), across crop types within the same rotations (p-values = 0.0001, 0.0002 and < 0.0001), and between oat versus alfalfa (p-values $\leq$ 0.0001).

Table 9: Significance of difference in abundance of the top seven weed species. Weed species are listed alphabetically. The abbreviations on the contrast column are crop identities, which are the combinations of the first letter in crop species names and the rotation to which the crops belonged.

	p-values													
	Stand density							Aboveground mass						
Contrast of the main-plot effect	AMATA	CHEAL	DIGSA	ECHCG	SETFA	SETLU	TAROF	AMATA	CHEAL	DIGSA	ECHCG	SETFA	SETLU	TAROF
(A) - Rotation system effects														
[(C2+S2)/2] vs [(C3+S3+O3+C4+S4+O4+A4)/7]	0.6105	0.0008	0.0072	0.1170	0.3011	0.1569	<.0001	0.3402	0.0199	0.1098	0.1417	0.9245	0.3588	<.0001
[(C3+S3+O3)/3] vs [(C4+S4+O4+A4)/4]	0.7077	0.9195	<.0001	0.0834	0.0927	0.0827	<.0001	0.8168	0.6414	0.0001	0.1040	0.4497	0.2420	<.0001
[(C2+S2)/2] vs [(C3+S3+C4+S4)/4]	0.1746	0.3889	0.6798	0.9584	0.1906	0.4944	0.8129	0.0893	0.2315	0.4852	0.8841	0.1566	0.5502	0.7608
[(C3+S3)/2] vs [(C4+S4)/2]	0.4533	0.3823	0.3213	0.9384	0.5877	0.6234	0.5105	0.4799	0.2676	0.4264	0.9958	0.9537	0.9148	0.4810
(B) - Rotation system effects within individual crops														
C2 vs [(C3+C4)/2]	0.3598	0.4995	0.8818	0.9497	0.5010	0.4277	0.9547	0.2696	0.4167	0.9499	0.9882	0.4070	0.5668	0.9237
C3 vs C4	0.6368	0.6510	0.2466	0.8579	0.3501	0.3990	0.6923	0.7802	0.6372	0.3994	0.7630	0.5131	0.6404	0.8309
S2 vs [(S3+S4)/2]	0.3065	0.5837	0.4658	0.9915	0.2337	0.8628	0.6958	0.1821	0.3720	0.3571	0.8252	0.2329	0.7847	0.7378
S3 vs S4	0.5543	0.4312	0.8088	0.9444	0.8620	0.8780	0.5914	0.4709	0.2708	0.7772	0.7687	0.5667	0.7516	0.4336
O3 vs O4	0.2890	0.6212	0.0062	0.2130	0.4848	0.2006	<.0001	0.3486	0.5666	0.0032	0.0768	0.3941	0.1539	<.0001
(C) - Crop type effects														
[(O3+O4+A4)/3] vs [(C2+S2+C3+S3+C4+S4)/6]	0.0001	<.0001	<.0001	0.0003	<.0001	0.0404	<.0001	0.0906	<.0001	<.0001	0.0012	0.0008	0.3316	<.0001
O3 vs [(C3+S3)/2]	0.0143	<.0001	0.0630	0.2248	0.0010	0.9435	0.0001	0.2355	<.0001	0.3924	0.3920	0.0180	0.5554	0.0002
[(O4+A4)/2] vs [(C4+S4)/2]	0.0003	<.0001	<.0001	0.0014	0.0001	0.0798	<.0001	0.0493	<.0001	<.0001	0.0031	0.0045	0.2706	<.0001
[(O3+O4)/2] vs A4	0.1606	0.0001	<.0001	0.1954	0.8068	0.1812	<.0001	0.0724	0.0001	0.0008	0.6762	0.1818	0.5132	0.0001

Note: C2 - corn in the 2-year rotation, C3 - corn in the 3-year rotation, C4 - corn in the 4-year rotation, S2 - soybean in the 2-year rotation, S3: soybean in the 3-year rotation, S4 - soybean in the 4-year rotation, O3 - oat in the 3-year rotation, O4 - oat in the 4-year rotation, and A4 - alfalfa in the 4-year rotation.

Table 10: Means of difference in abundance of the top seven weed species. Weed species are listed alphabetically. The abbreviations on the contrast column are crop identities, which are the combinations of the first letter in crop species names and the rotation to which the crops belonged.

	Contrast ratio													
	Stand density							Aboveground mass						
Contrast of the main-plot effect	AMATA	CHEAL	DIGSA	ECHCG	SETFA	SETLU	TAROF	AMATA	CHEAL	DIGSA	ECHCG	SETFA	SETLU	TAROF
(A) - Rotation system effects														
[(C2+S2)/2] vs [(C3+S3+O3+C4+S4+O4+A4)/7]	0.74	0.28	0.42	0.57	0.64	0.50	0.24	3.10	0.21	0.36	0.35	0.93	0.46	0.07
[(C3+S3+O3)/3] vs [(C4+S4+O4+A4)/4]	0.81	0.97	0.21	0.55	0.49	0.44	0.19	1.30	1.33	0.07	0.32	0.56	0.39	0.05
[(C2+S2)/2] vs [(C3+S3+C4+S4)/4]	2.45	1.37	1.14	0.98	1.86	0.70	0.95	9.26	2.30	1.60	0.89	3.54	0.58	0.86
[(C3+S3)/2] vs [(C4+S4)/2]	1.76	1.45	0.69	0.97	0.75	0.74	0.84	2.83	2.43	0.54	1.00	0.94	0.89	0.67
(B) - Rotation system effects within individual crops														
C2 vs [(C3+C4)/2]	2.33	1.42	0.93	0.97	1.56	0.56	1.02	7.45	2.21	1.06	1.02	2.81	0.48	0.94
C3 vs C4	1.65	1.31	0.54	0.89	0.49	0.49	0.87	1.78	1.70	0.40	0.69	0.39	0.50	0.85
S2 vs [(S3+S4)/2]	2.58	1.33	1.40	0.99	2.21	0.88	0.88	11.50	2.39	2.40	0.79	4.47	0.71	0.80
S3 vs S4	1.87	1.60	0.88	1.04	1.14	1.14	0.82	4.50	3.49	0.73	1.44	2.27	1.59	0.54
O3 vs O4	0.32	0.74	0.21	0.46	0.59	0.33	0.09	0.14	0.53	0.03	0.10	0.29	0.12	0.01
(C) - Crop type effects														
[(O3+O4+A4)/3] vs [(C2+S2+C3+S3+C4+S4)/6]	12.25	38.15	10.11	3.60	9.85	2.48	24.33	6.11	204.44	27.29	9.56	15.00	2.05	389.81
O3 vs [(C3+S3)/2]	10.94	67.07	2.43	1.94	11.32	1.05	4.33	8.70	571.14	2.26	2.54	22.34	0.47	19.10
[(O4+A4)/2] vs [(C4+S4)/2]	23.36	36.99	20.08	4.82	11.63	2.96	53.81	20.20	231.64	102.80	17.54	22.79	3.18	1482.81
[(O3+O4)/2] vs A4	3.71	10.75	0.07	0.49	1.17	0.37	0.17	28.24	94.46	0.03	0.64	5.38	0.43	0.05

Note: C2 - corn in the 2-year rotation, C3 - corn in the 3-year rotation, C4 - corn in the 4-year rotation, S2 - soybean in the 2-year rotation, S3: soybean in the 3-year rotation, S4 - soybean in the 4-year rotation, O3 - oat in the 3-year rotation, O4 - oat in the 4-year rotation, and A4 - alfalfa in the 4-year rotation.

Discussion

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