

Influence of cover cropping and conservation tillage on weeds during the critical period for weed control in soybean

Research Article

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Abstract

Limited research has been directed at evaluating the ability of single cover crop plantings to suppress weeds in crops beyond the initial field season. Thus, this experiment was conducted to investigate the ability of a second-year self-regenerated annual and second-year perennial cover crop planting to suppress weeds during the critical period for weed control (CPWC) in soybean crops. Whole-plot treatments included 1) conventional till, 2) no-till with cover crop residue, 3) living mulch + cover crop residue, and 4) living mulch + winter-killed residue. Subplot treatments involved weed management intensity: a) no weed management (weedy), b) weeds manually removed through the CPWC (third node soybean stage; V3), and c) weeds manually removed until soybean canopy closure (weed-free). Overall, total annual cover crop biomass during the second field season was comparable to biomass obtained from direct seeded stands during the initial field season. All cover crop treatments reduced total weed biomass through the CPWC compared to conventional till. Soybean yield was low across all treatments in this experiment. Still, yield was similar between cover crop and conventional till treatments at one site-year, however, yields were lower in all cover crop treatments at the other site-year.

Introduction

Cover crop residues and living mulches can suppress agricultural weeds (Creamer et al. 1996, Florence et al. 2019, Mirsky et al. 2011), making cover cropping a viable practice in integrated weed management (IWM) programs. However, most cover crops need to be sown each year, and establishment costs are regarded as a primary economic issue that hinders their adoption (Duke et al. 2022, Dunn et al. 2016, Lemessa and Wakjira 2015) and subsequent incorporation into an IWM plan. Recent policy initiatives, including the Environmental Quality Incentives Program managed by the U.S. Department of Agriculture–Natural Resources Conservation Service, and the Pandemic Cover Crop Program, managed by the U.S. Department of Agriculture–Risk Management Agency, have resulted in an increase in the number of farmers growing cover crops (Wallander et al. 2021). In 2017, U.S. farmers reported planting 6.2 million ha of cover crops, a 50% increase compared with 2012, and in 2018, roughly one-third of the cover crop acreage planted was aided by financial assistance from federal, state, or other programs that foster cover crop adoption (Wallander et al. 2021). Still, prevalence remains low, with cover crop adoption occurring on roughly 5% of the total cropped area in the United States (Deines et al. 2022). In addition to policy incentives, costs associated with cover crop planting may be mitigated by extending single cover crop plantings over several years. This can be accomplished by planting perennial or self-regenerating annual cover crops. Cost savings from self-regenerating annuals may help encourage their adoption (Bergtold et al. 2019). Additionally, lack of time to plant cover crops following fall harvest is frequently stated as another barrier to adoption (Roesch-McNally et al. 2018). Self-regenerating annuals or perennial cover crops would alleviate this concern. Furthermore, if a single cover crop planting can contribute to weed suppression over multiple growing seasons, this will provide farmers an additional incentive for their adoption.

The critical period for weed control (CPWC) is the duration of time during which weeds must be managed to prevent yield loss exceeding a defined threshold (Charles and Taylor 2021). The CPWC contains two weed-crop competition components: the critical time for weed removal (CTWR) and the critical weed-free period (CWFP). The CTWR is the maximum length of time a crop can tolerate early season weed competition, and therefore determines the start of the CPWC. The CWFP is the minimum length of time after planting when a crop must be kept weed free, thus determining the end of the CPWC (Knezevic et al. 2002; Rosset and Gulden 2019). Recent research investigating the influence of cover crops on soybean CPWC determined that the presence of a fall-seeded cereal rye (*Secale cereale* L.) cover crop delayed the onset of the CTWR and shortened the CWFP, thus decreasing the CPWC (Kumari et al. 2023). Similarly,

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Price et al. (2018) found that the presence of a fall-seeded cereal rye cover crop in combination with conservation tillage delayed the CTWR in cotton [*Gossypium hirsutum* L.] by approximately 3 wk after planting, thus shortening the total CPWC. In addition to reducing weed biomass by hindering weed seedling emergence during the CWFP, cover crop residues and living mulches can slow the growth and development of weed seedlings that do successfully emerge by imposing additional competitive pressures (Bhaskar et al. 2021). Several studies have investigated the use of conservation tillage and cover cropping for weed suppression in soybeans (Mirsky et al. 2011; Moore et al. 1994; Rosario-Lebron et al. 2019; Weber et al. 2017). However, limited research has been directed at determining how these practices affect weeds specifically during the CPWC. Yet, this information could assist growers in making more informed weed management decisions.

The duration of the CPWC is influenced by many factors, including cropping environment and management actions. In soybean crops, the average CPWC typically extends until the third trifoliate stage or V3, which typically occurs roughly 30 d after planting (Van Acker et al. 1993). However, in some cases, it may not end until the reproductive stages (Eyherabide and Cendoya 2002). In general, management is often considered beneficial beyond the early vegetative stages only if weed presence will hinder harvest efficiency (Chandler et al. 2001). Notwithstanding, an important goal of IWM is preventing weeds from producing seeds, and subsequently increasing the weed seedbank and contributing to future weed problems (Haring and Flessner 2018). Cover crop mulches may prevent weeds from reaching maturity through the harvest period by delaying weed emergence and slowing their development (Williams et al. 1998). Delayed weed emergence and development can reduce total weed seed production, thereby reducing contributions to the soil seedbank and ultimately lowering the weed pressure in subsequent years (Mennan et al. 2020). Reducing weed seedbank entry is especially important when trying to thwart herbicide-resistant weeds (Norsworthy et al. 2018). Furthermore, cultural weed management practices, including the use of cover crops, if appropriately used as part of an IWM program, can reduce herbicide usage, consequently lowering the selection pressure for herbicide resistant weeds (Bunck et al. 2020).

Research has shown that some annual legumes such as crimson clover (*Trifolium incarnatum* L.) can be used as a cover crop for multiple seasons due to the plant's ability to readily self-regenerate (Myers and Waggoner 1991, Rodrigues et al. 2015). Perennial cover crops may also be used for several production seasons (Sanders et al. 2017). However, limited research has been conducted to evaluate the ability of a single cover crop seeding event to suppress weeds in subsequent growing seasons. Thus, the purpose of this experiment was to investigate the ability of second-year self-regenerated annual and second-year perennial cover crops to suppress weeds through and beyond the CPWC in a soybean crop. For this experiment, we hypothesized that the self-regenerating and perennial cover crop systems would provide greater weed suppression in a subsequent soybean crop than the conventional tillage system. We further investigated treatment impacts on weed maturity and hypothesized that more weeds would reach their reproductive stages in the conventional tillage compared to self-regenerating and perennial systems by the late soybean reproductive stage.

Materials and Methods

Experimental Design and Field Operations

Field experiments were conducted during two growing seasons at the Central Maryland Research and Education Center in Upper Marlboro, MD (38.859079°N, 76.778731°W) in 2020 and Beltsville, MD (39.011440°N, 76.833356°W) in 2021 within fields where sweet corn (variety Providence) was the cash crop during the previous growing season. Onsite weather stations indicated the average temperature was 21.3 C and 22.3 C, and total precipitation was 705 mm and 678 mm during 2020 and 2021 soybean growing seasons (June to October), respectively. Soil at the Upper Marlboro site is an Annapolis series (fine-loamy, glauconitic, mesic Typic Hapludults). At the Beltsville site the soil is a Russett-Christiana complex where the Russett surface soil is a fine-loamy, mixed, semiactive, mesic Aquic Hapludults, and the Christiana surface soil is a fine, kaolinitic, mesic Aquic Hapludults.

Treatments were arranged in a Latin square-split-plot design with four replicates. Whole-plot treatments were as follows: conventional till (CT), no-till with self-regenerated cover crop residue (NT), second-year perennial living mulch + self-regenerated forage radish (LMFR) residue, and second-year perennial living mulch + self-regenerated rye residue (LMRye). Each whole plot was subdivided into three subplots that received varying levels of weed management. Subplot treatments included weeds controlled 1) until the end of the CPWC for soybean (hereafter called V3); 2) until soybean canopy closure (weed-free, hereafter termed Wf); and 3) no weed control (weedy, hereafter termed Wd). The main plots measured 82.8 m² (9.1 m × 9.1 m), and each subplot measured 23.6 m² (3.1 m × 9.1 m). Each subplot consisted of four soybean rows planted at an interrow spacing of 0.76 m. Weeds in V3 and Wf subplots were removed weekly by hand pulling and hoeing. While manual weed removal is not the typical weed management practice for this crop, it was the most practical method for weed removal in this experiment.

All cover crops were drilled at an interrow spacing of 0.15 m. During early fall of 2018 (Upper Marlboro) and 2019 (Beltsville), crimson clover (3.36 kg ha⁻¹), forage radish (*Raphanus sativus* 'longipinnatus'; 3.9 kg ha⁻¹), and cereal rye (*Secale cereale* L. 'Aroostook'; 62.8 kg ha⁻¹) were mixed and planted in CT and NT plots. Perennial red clover (*Trifolium pratense* L.; 16.8 kg ha⁻¹) + forage radish (11.2 kg ha⁻¹) and perennial red clover (9 kg ha⁻¹) + rye (75 kg ha⁻¹) were planted in alternating strips in the LMFR and LMRye plots, respectively. The alternating strips arrangement consisted of two rows of red clover followed by three rows of forage radish (LMFR) or rye (LMRye). Sweet corn was planted in all plots and subsequently flail-mowed the following fall to eliminate stalks. Because cover crops in tilled treatments were not expected to self-regenerate, the CT plots were also disked and the cover crop mixture of crimson clover, rye, and forage radish was replanted at the same rates as the previous fall. Crimson clover and rye in the NT and LMRye treatments naturally self-regenerated and the red clover in LMFR and LMRye remained established for the subsequent field season. Thus, the NT, LMFR, and LMRye plots did not require any additional operations following flail mowing during the fall in preparation for the soybean experiment. Photographs showing the arrangement of cover crops within all treatments are provided in Figure 1.

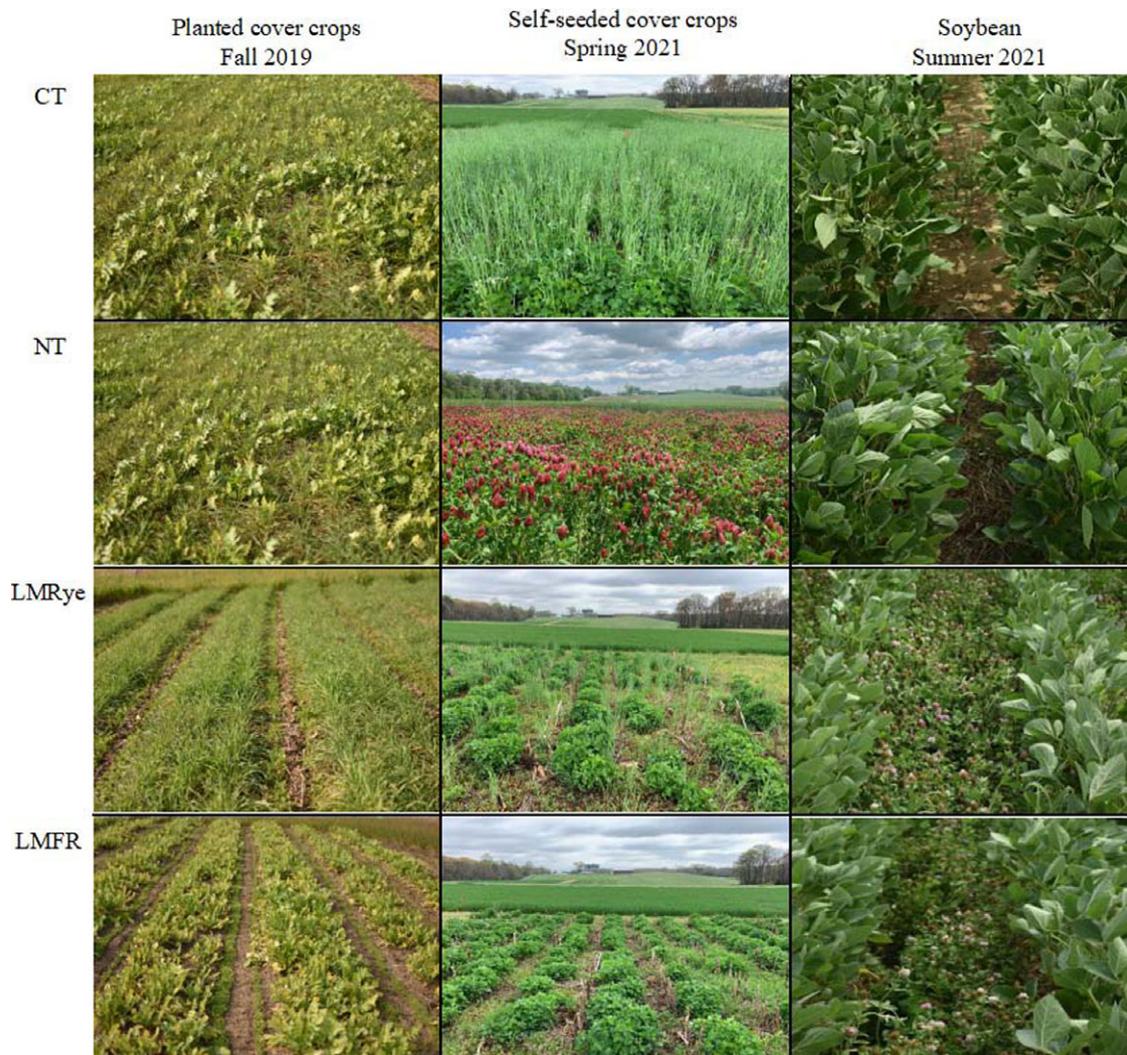


Figure 1. Images showing the cover crop arrangement in planted cover crops following emergence (fall 2019) and self-regenerated cover crops prior to termination (spring 2021), as well as soybean arrangement (summer 2021) at the Beltsville experiment site. Abbreviations: CT, conventional till; NT, no till; LMFR, living mulch + forage radish; LMRye, living mulch + rye. In spring, CT treatments were flail-mowed and tilled; and NT, LMFR and LMRye treatments were roller-crimped. LMFR and LMRye treatments were then strip-tilled, and soybean was planted into bare ground in CT treatment plots, rolled residue in NT treatment plots, and tilled strips in LMFR and LMRye subplots.

In the subsequent spring, when the rye reached anthesis, cover crops in CT plots were mowed and tilled to incorporate the cover crop residue into the soil (green manuring). A 3-m-wide roller crimper was used to terminate the rye in NT and LMRye plots, and temporarily slow red clover growth in LMFR and LMRye plots. To clear encroaching living mulch from the intrarow areas, a two-row strip tiller equipped with cutting disks, a shank, and rolling basket assembly (Bigam Brothers, Inc. Lubbock, TX) with a tillage width of 0.28 m was used in the LMFR and LMRye treatments.

In early June of 2020 and 2021, soybean (cultivar 'Monocacy', maturity group 4.3) was seeded into each plot at an interrow spacing of 0.76 m, resulting in 12 soybean rows per plot. Variation in available equipment at each study location resulted in soybeans being planted using a three-point-hitch mounted, two-row vacuum planter (Monosem; Edwardsville, KS) in Upper Marlboro in 2020, and a six-row no-till planter (John Deere 1750 MaxEmerge; Deere and Company, Moline, IL) in Beltsville in 2021. Soybeans were seeded at a rate of 371,000 seeds ha^{-1} in 2020 and 383,000 seeds ha^{-1} in 2021. In LMFR and LMRye plots, soybean seeds were planted within the center of the tilled strips. In

2021, the grass herbicide fluazifop was applied as a rescue treatment at a rate of 0.84 kg ai ha^{-1} to all Wf subplots to control grass weeds. Timing of field tasks is provided in Table 1. No supplemental irrigation was provided.

Cover Crop and Winter Annual Weed Biomass

Cover crop and winter annual weed biomass were collected from each plot just prior to cover crop termination by clipping shoot tissue at ground level from two 0.3-m \times 0.3-m quadrats. Each quadrat was placed randomly in CT and NT treatments and within one intrarow and interrow area of the LMFR and LMRye plots. Plant material collected within each quadrat was separated by cover crop or weed species, placed in paper bags, dried at 60 C (>1 wk), and weighed to determine dry biomass.

Weed Biomass, Species Assemblages, and Maturity Level

To assess treatment effect on weed emergence and biomass accumulation through the CPWC, two 0.3-m \times 0.3-m quadrats were placed randomly in CT and NT treatments and intrarow

Table 1. Timing of field operations.

Activity	Upper Marlboro	Beltsville
Cover crops planted in all plots ^{1a}	September 14, 2018	September 5, 2019
Annual cover crops terminated	May 23, 2019	May 25, 2020
Cover crops replanted in CT ^{2b}	September 3, 2019	September 16, 2020
Regenerated annual cover crops terminated	May 27, 2020	May 28, 2021
Soybean planted	May 27, 2020	June 2, 2021
Herbicide applied ^{3c}	–	July 7, 2021
Soybean harvested	October 28, 2020	October 28, 2021

^{1a}Cover crops were initially planted in the prior field season for a separate field experiment.

^{2b}Cover crops used in the soybean experiment had to be replanted in conventional till (CT) plots only.

^{3c}Rescue grass herbicide was applied to weed free (Wf) subplots only.

areas of LMFR and LMRye plots. Weeds were clipped at ground level and separated according to species. Weed maturity measurements at the Upper Marlboro site were originally planned to take place at soybean harvest. However, the unexpectedly late maturity of the chosen soybean variety resulted in the senescence of all summer annual weeds prior to soybean harvest. As such, quadrats were similarly used 2 wk after canopy closure at the Beltsville site to measure weed biomass and estimate maturity stages of weeds within all subplots. The maturity stage was categorized as seedling, vegetative, bud, flower, immature seed, or mature seed. Determination of immature versus mature seeds was based on visual characteristics similar to those described by Hill et al. (2016). Samples collected at V3 were taken from the Wd subplots to provide a measurement of weed biomass accumulated through the CPWC in the absence of any weed management intervention. Samples collected 2 wk after canopy closure were taken from all subplots. Dry weight measurements of each species were combined to calculate total weed biomass per treatment, and individual species measurements were used to compare species abundance between treatments. V3 was chosen as the CPWC in this experiment based on previous studies investigating the CPWC for soybean. However, it is known that multiple factors, including tillage operation, weed community, soil type, etc. can influence the soybean CPWC (Halford et al. 2001, Kumari et al. 2023).

Soybean Emergence and Yield

Soybean stand counts were conducted in all treatments less than 10 d after soybean planting. Counts were taken from the center two rows of each subplot and repeated every 3 to 4 d until all viable seedlings had emerged. To estimate yield, all soybean plants within the center 5.3 m of one interior row per subplot were manually harvested, threshed to separate the seeds from the pods, and all seeds were dried to 13% moisture and weighed.

Statistical Analysis

Biomass of each cover crop species was not analyzed statistically, but means and standard errors of each treatment were calculated for reference. Linear models were used to determine differences in total regenerated cover crop biomass and winter annual weed biomass among treatments and between years. No statistical comparisons of first-year (direct seeded) versus second-year (regenerated) cover crop biomass were performed because CT treatments were replanted the following year, providing a better comparison between direct seeded and regenerated biomass in NT treatments. Furthermore, red clover biomass was not taken during the first experiment year following planting at both locations. Linear mixed models were performed on weed abundance, weed biomass, and soybean yield data to test for differences among the

fixed effects of cover crop treatment (whole plot), weeding intensity (subplot treatment), and experiment year. When year was significant, the two site-years were analyzed separately. Plot identity was included as a random effect to account for the split-plot design. When there was a significant effect of cover crop treatment, preplanned orthogonal contrasts were performed to test for treatment differences between 1) CT and pooled cover crop treatments (NT, LMFR, and LMRye); 2) NT and pooled living mulch treatments (LMFR and LMRye); and 3) living mulch treatments (LMFR vs. LMRye). When there was a significant difference between subplot means, all pairwise comparisons were performed using Tukey-adjusted P-values (Lenth 2020). Weed biomass was log transformed to meet assumptions of normally distributed residuals. An alpha level of 0.05 was used throughout. All statistical analyses were performed using R software (version 4.1.2; R Core Team 2021). Linear models were built using the LME4 package (Bates et al. 2015). Preplanned contrasts and post hoc means comparisons were performed using the EMMEANS package (Lenth 2020). All figures were made using the GGLOT2 package (Wickham 2009).

Results and Discussion

Cover Crop Biomass

Statistical comparisons of direct-seeded versus regenerated cover crops were not practical due to the influence of variable environmental factors between experiment years. Generally, spring biomass of fall-planted cover crops was mostly similar between experimental sites. Likewise, total biomass of second-year self-regenerated annual and second-year perennial cover crops was similar between sites (Table 2). In NT plots, the total biomass of the self-regenerated cover crop measured in the spring of Season 2 was similar to the biomass collected during the spring of Season 1 when the cover crop was directed seeded during the fall. However, the ratio of legume to grass cover crop in the self-regenerated NT treatment was higher than in the direct-seeded cover crops. In NT plots, the dry biomass of crimson clover was 2,400 kg ha⁻¹ in Upper Marlboro and 4,040 kg ha⁻¹ in Beltsville, and the dry biomass of rye was 4,070 kg ha⁻¹ in Upper Marlboro and 2,820 kg ha⁻¹ in Beltsville during the initial planting. However, following self-regeneration, there was a greater percentage of crimson clover in NT plots, which was 7,040 kg ha⁻¹ in Upper Marlboro and 8,810 kg ha⁻¹ in Beltsville. In contrast, there was no rye biomass in NT plots at the Upper Marlboro site and only 54 kg ha⁻¹ at the Beltsville site (Table 2). Biomass measurements of red clover were not taken from LMRye or LMFR plots during the initial spring following cover crop planting (2019 in Upper Marlboro and 2020 in Beltsville). In those years, biomass data were taken only from the

Table 2. Total spring biomass of all cover crop species and winter annual weeds during the initial and subsequent field season.^{a,b,c}

Treatment	Crimson clover	Rye	Radish	Red clover ^d	Total cover crop	Weed
kg ha ⁻¹						
Upper Marlboro – direct seeded cover crop (biomass sampled in spring 2019)						
CT	2,967.6 ± 673	4,342 ± 711	0 ± 0	N/A	7,310 ± 662	42 ± 12
NT	2,396 ± 699	4,073 ± 332	0 ± 0	N/A	6,470 ± 591	26 ± 10
LMFR	N/A	N/A	0 ± 0	–	0 ± 0	1,020 ± 91
LMRye	N/A	7,222 ± 364	N/A	–	7,222 ± 634	34 ± 14
Upper Marlboro – self-regenerated cover crop (biomass sampled in spring 2020)						
CT	2,356 ± 1,154	3,311 ± 1,120	0 ± 0	N/A	5,667 ± 598	69 ± 31
NT	7,043 ± 983	0 ± 0	0 ± 0	N/A	7,043 ± 983	6 ± 6
LMFR	N/A	N/A	0 ± 0	3,581 ± 1,010	3,581 ± 1,010	236 ± 104
LMRye	N/A	1,071 ± 1,037	N/A	1,465 ± 470	3,536 ± 655	126 ± 79
Beltsville – direct seeded cover crop (biomass sampled in spring 2020)						
CT	2,826 ± 1,089	5,534 ± 1,204	391 ± 373	N/A	6,571 ± 742	130 ± 117
NT	4,041 ± 1,111	2,823 ± 866	221 ± 221	N/A	7,085 ± 1,097	91 ± 66
LMFR	N/A	N/A	1,323 ± 668	–	1,323 ± 668	513 ± 73
LMRye	N/A	6,992 ± 2,254	N/A	–	6,993 ± 2,254	75 ± 38
Beltsville – self-regenerated cover crop (biomass sampled in spring 2021)						
CT	3,732 ± 1,235	4,371 ± 888	82 ± 71	N/A	8,485 ± 1,466	93 ± 53
NT	8,812 ± 1,196	54 ± 41	0 ± 0	N/A	8,866 ± 1,203	85 ± 57
LMFR	N/A	N/A	0 ± 0	6,098 ± 910	6,098 ± 910	534 ± 222
LMRye	N/A	1,449 ± 843	N/A	4,767 ± 1,326	6,217 ± 1,092	272 ± 132

^aAbbreviations: CT, conventional till; LMFR, living mulch + forest radish; LMRye, living mulch + rye; NT, no till.

^bNumbers represent total spring biomass in kilograms per hectare (kg ha⁻¹) ± SEM.

^cAll cover crops were initially planted in Upper Marlboro in fall 2018 and in Beltsville in fall 2019. Cover crops in the CT treatment were replanted each year.

^dRed clover living mulch was not sampled in LMFR and LMRye in spring following planting.

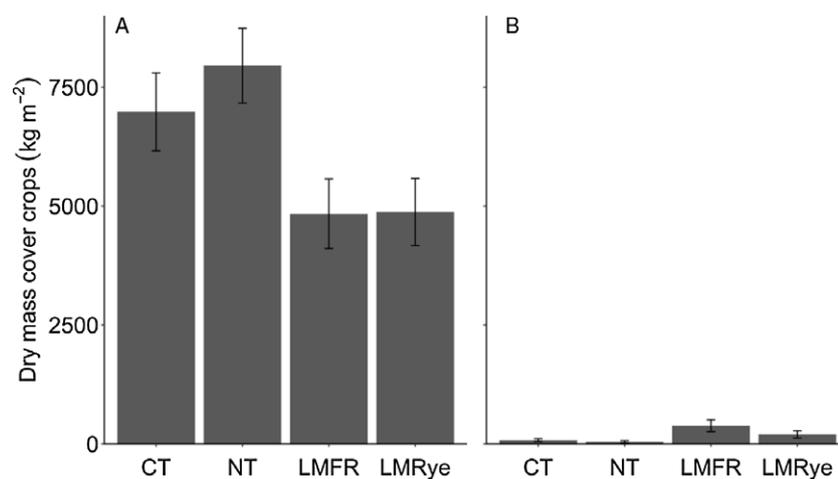


Figure 2. Total dry mass of cover crops (A) and winter annual weeds (B) in 2020 (Upper Marlboro) and 20201 (Beltsville) in replanted conventional till (CT), and self-regenerated no-till (NT), living mulch + forage radish residue (LMFR), and living mulch + rye residue (LMRye) treatment plots.

intrarow area of these treatments, which consisted of rye or forage radish. The red clover was used as a living mulch as opposed to organic residue and was restricted to the interrow area during the first field season following planting. However, by spring of the second growing season, the larger red clover plants extended into the intrarow area of LMFR and LMRye treatments. Strips of self-regenerated cereal rye were mixed in with the red clover in the intrarow area of LMRye plots. However, self-regenerated forage radish plants were not perceptible in any treatments during the second field season.

A significant effect of treatment ($F_{3,24} = 4.33$, $P = 0.01$) and experiment year ($F_{1,24} = 11.11$, $P < 0.01$) were detected for total self-regenerated cover crop biomass. However, P-value corrections

resulting from Tukey pairwise comparisons did not detect any significant treatment differences, although P-values were close for LMFR ($P = 0.0502$) and LMRye ($P = 0.0538$) when compared to NT (Figure 2A). Total regenerated cover crop biomass was greater in Beltsville in 2021 than in Upper Marlboro in 2020. No significant interaction of treatment and experiment year was detected ($F_{3,24} = 0.22$, $P = 0.08$). For winter annual weed biomass, no significant effect of treatment ($F_{3,24} = 2.50$, $P = 0.08$), experiment year ($F_{1,24} = 1.86$, $P = 0.19$), or their interaction ($F_{3,24} = 0.38$, $P = 0.77$) was detected (Figure 2B).

This experiment was designed, in part, to test the capacity for fall-planted annual cover crops to naturally re-establish through self-regeneration and a perennial cover crop to remain established

the subsequent growing season. While statistical comparison of direct-seeded versus regenerated biomass between years was not performed, comparisons between replanted CT and regenerated NT biomass indicated that total cover crop biomass in plots with self-regenerated cover crops was comparable to that obtained from direct-seeded stands. However, the proportion of individual species by weight changed. Limited regeneration of cereal rye and forage radish in NT plots resulted in second-year annual cover crop biomass that was predominantly crimson clover. In general, crimson clover comprised 40% to 46% of the total replanted biomass in CT; however, biomass in the regenerated NT treatment, originally planted the previous year with the same cover crop mixture, was 99% to 100% crimson clover at both sites. This was expected because the crimson clover senesced prior to termination, while the rye was terminated before all plants had reached full anthesis, and most of the forage radish was killed in winter while still vegetative. In living-mulch treatments, the red clover, which was mostly restricted to the interrow areas in Season 1, overwintered and the larger regenerated plants had spread throughout the entire plot prior to soybean planting. The high cover crop biomass across all treatments hindered winter annual weed establishment, which resulted in low weed biomass at cover crop termination in all plots. The successful self-regeneration and continued growth of perennial cover crops may prove useful to growers who are unable to direct-seed a cover crop following fall harvest or who desire to avoid the additional labor or cost associated with replanting, and it may be useful to farmers who prefer that their cover crops are established earlier in the growing season. Roesch-McNally et al. (2018) identified difficulty in timing of cover crop establishment as a specific challenge associated with cover crop usage. There is often insufficient time to establish a cover crop following late-harvested crops such as corn and soybean (Roesch-McNally et al. 2018; Wallace et al. 2017). Furthermore, if a cover crop is planted too late, this could result in insufficient biomass needed to suppress weed establishment during the subsequent cropping season (Akbari et al. 2019). Lawson et al. (2015) found that delaying the planting of winter cover crops by 2 to 3 wk can have a marked effect on a farmer's ability to protect soil and produce biomass. Those researchers also found that a delay in cover crop planting by 2.5 wk reduced average winter ground cover by 65% and biomass by 50%. As such, using self-regenerated annual or established perennial cover crops provides an additional cover cropping strategy that may ameliorate some of the issues associated with planting cover crops annually.

Weed Biomass, Species Assemblages, and Maturity

In 2020, greater weed biomass was collected from Wd subplots at the end of the CPWC (31 d after soybean planting) from CT plots, where cover crops were terminated in spring prior to tillage and soybean planting, compared to all cover crop treatments (LMFR, LMRye, and NT; $t_{12} = 12.65$, $P < 0.0001$) (Figure 3). Greater weed biomass was also collected in Wd subplots in the NT plots compared to the living mulch treatments (LMFR and LMRye; $t_{12} = 8.86$, $P < 0.0001$), and in LMFR compared to LMRye ($t_{12} = 2.00$, $P = 0.02$) at the end of the CPWC (Figure 2). In 2021, weed biomass was again greater in CT plots compared to the cover crop treatments ($t_{11} = 5.10$, $P = 0.03$). In contrast, weed biomass in NT plots was less than in the living-mulch treatments ($t_{11} = 3.64$, $P = 0.05$) and no difference was detected between LMFR and LMRye ($t_{11} = 0.01$, $P = 0.98$) at the end of the CPWC (38 d after planting; Figure 3). Substantial variation in weed species

abundance occurred between experiment site-years and treatments at the conclusion of the CPWC. Dominant species in 2020 at the Upper Marlboro site were primarily carpetweed (*Mollugo verticillata* L., 22%), white clover (*Trifolium repens* L., 31%), and goosegrass [*Eleusine indica* (L.) Gaertn., 33%]. In 2021 at the Beltsville site, dominant weed species were yellow nutsedge (*Cyperus esculentus* L., 14%), goosegrass (15%), and giant foxtail (*Setaria faberii* R.A.W. Herrm., 37%).

Several studies have investigated how various cultural practices, including cover cropping, impact the timing of the CPWC in soybeans (Halford et al. 2001; Kumari et al. 2023; Rosset and Gulden 2019) and other crops (Price et al. 2018; Tursun et al. 2016). However, no studies have specifically linked the influence of cover crop residues or living mulches on weed growth and development with a defined CPWC. Relevant to this, a meta-analysis conducted by Osipitan et al. (2018) found that cover crop residues provided early-season weed suppression comparable to that provided by chemical and mechanical weed control methods. However, it is unclear whether the weed suppression provided by cover crop residues occurred during the CPWC. During the current experiment, variable levels of weed suppression occurred among treatments and years through the V3 soybean stage. Still, as hypothesized, all cover crop treatments consistently reduced weed biomass compared to the CT treatment. In this treatment, tillage likely mixed the weed seedbank, exposing seeds to sunlight and subsequently stimulating germination. However, the NT treatment contained greater weed biomass than the living-mulch treatments at the end of the CPWC in 2020 and less weed biomass in 2021. In 2020, perennial white clover made up much of the weed biomass in the NT treatment, which suggests that the cover crop residue could not adequately suppress white clover. Previous research has demonstrated that the weed-suppressive effects of cover crop residues are species specific and that perennial weed species are not adequately suppressed by cover crop residue (Liebman and Davis 2000; Mirsky et al. 2011; Mohler and Teasdale 1993). In contrast, low white clover biomass was found among LMFR and LMRye in 2020, which suggests that a living mulch may be more successful in preventing some perennial weeds from establishing in crop fields than cover crop residue. Similarly, Hiltbrunner et al. (2007) found that living mulches were more effective at preventing the germination and establishment of perennial weeds than cover crop residues that have been killed. Although treatment differences were detected between LMFR and LMRye during 2020 in Upper Marlboro, this difference may not be agronomically important in most production scenarios because weed levels remained low in both treatments. In 2021 at the Beltsville site, the red clover did not regenerate well, and open gaps were exploited by weeds, which likely contributed to the greater weed biomass collected from living mulch than from the NT treatment. When using cover crop residue for weed suppression, the amount of biomass is critical (Nichols et al. 2020). However, for a clover living mulch, having the ground completely covered is presumably more important than the overall biomass because gaps in stand can be exploited by weeds (Basinger and Hill 2021). Still, weed biomass was low across all treatments at the Beltsville site during 2021.

In Upper Marlboro during 2020, weed maturity measurements were not taken because soybean plants matured much later than anticipated. As such, it was noted at the time of soybean harvest that weeds in all treatment plots had senesced. In the data presented here, all weed maturity results pertain to Beltsville in 2021. Variation in species abundance between treatments and subplots precluded the ability to conduct maturity comparisons

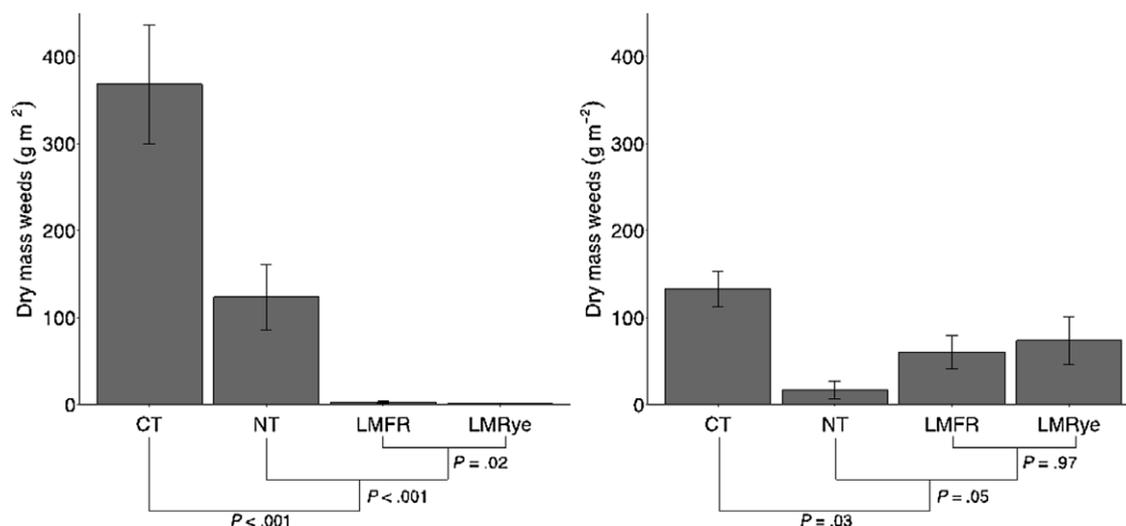


Figure 3. Dry mass of weeds accumulated through the V3 soybean stage in 2020 in Upper Marlboro, MD (left) and 2021 in Beltsville, MD (right). Abbreviations: CT, conventional till; NT, no-till; LMFR, living mulch + forage radish residue; LMRye, living mulch + rye residue. P-values represent significance levels for contrasts of groups intersecting at those nodes.

Table 3. Total biomass of weed species in reproductive stages 2 wk following soybean canopy closure in 2021, in Beltsville, MD.^{a,b}

Treatment	Weed biomass		
	Wd	V3	Wf
	g m ⁻²		
CT	92.5 ± 36.9	2.3 ± 1.5	0.0 ± 0.0
NT	19.5 ± 10.0	3.7 ± 2.6	0.0 ± 0.0
LMFR	50.7 ± 29.1	0.0 ± 0.0	0.0 ± 0.0
LMRye	49.8 ± 27.0	0.9 ± 0.4	0.0 ± 0.0

^aAbbreviations: CT, conventional till; LMFR, living mulch + forage radish; LMRye, living mulch + rye; NT, no till; V3, weeds controlled through the soybean critical period for weed control (V3 stage); Wd, unweeded; Wf, weeds controlled through soybean canopy closure.

^bTotal biomass is presented in grams per square meter (g m⁻²) ± SEM.

among treatments for individual species. As such, all weed species were pooled for comparisons. Overall, a greater amount of reproductive-stage weeds (flower, immature seed, and mature seed) was present 2 wk after soybean canopy closure in Wd subplots compared to V3 or Wf subplots ($\chi^2 = 90.33$, $df = 1$, $P < 0.001$; Table 3). No treatment differences were detected within any subplots. Weeds in the reproductive stage were present in low numbers at V3 in CT, NT, and LMRye subplots, and their total biomass was not significantly different from zero. Wf subplot treatments did not contain any reproductive-stage weeds and were therefore not included in the analysis (Table 3). These findings contrast our original supposition that more reproductive-stage weeds would be present in conventional tillage (CT) than in conservation tillage cover crop treatments. It was expected that the cover crops would provide an additional source of competition, resulting in reduced weed emergence and delayed weed maturation compared to the bare-ground CT treatment. However, in 2021, the red clover in the LMFR and LMRye plots was patchy, which allowed open areas for early weed establishment, notably grass species.

The low biomass of reproductive-stage weeds in V3 subplots during one site-year may suggest that restricting weed management to only to the CPWC period may be sufficient in some

production situations. However, if highly prolific weed species are present, even low numbers of reproductive-stage weeds can result in large numbers of unwanted seeds entering the seedbank (Schwartz et al. 2016). Furthermore, if herbicide-resistant weeds are present, low numbers of these seeds entering the seedbank is undesirable.

Soybean Emergence and Yield

In 2020 at the Upper Marlboro field site, there were reduced ($F_{3,12} = 7.71$, $P < 0.001$) soybean stand counts in NT treatment plots compared to living mulch treatments ($t_{12} = 0.58$, $P < 0.01$; Figure 4). In contrast, greater soybean yields were detected from NT treatments compared to the living mulch treatments ($t_{44} = 0.39$, $P < 0.05$; Figure 4). However, yields from CT treatments were similar to those from the cover crop treatments. There was also a significant subplot treatment effect ($F_{2,45} = 15.94$, $P < 0.001$) with greater yields occurring in Wf and V3 subplots compared to Wd subplots (Figure 5). In 2021 at the Beltsville site, final stand counts were also lower from NT treatments compared to CT and living mulch treatments ($F_{3,12} = 9.49$, $P < 0.01$; Figure 4). Overall, soybean yield was low across all treatments in 2021; however, greater yields were detected from CT treatments compared to all cover crop treatments ($t_{44} = 0.99$, $P < 0.05$; Figure 5). Similar to 2020, a subplot treatment effect ($F_{2,45} = 9.18$, $P < 0.001$) indicated that yields were greater from Wf subplots compared to Wd subplots (Figure 6).

Soybean yield was low across all treatments in both experiment years compared with regional averages (USDA-NASS 2023). Regional soybean yield measured 2,804 kg ha⁻¹ in 2020 and 3,396 kg ha⁻¹ in 2021. Measured yields in this experiment were likely reduced by the harvesting equipment we used, which was old and unable to collect all of the soybean pods on the plants. However, any effect on total yield was consistent between treatments, and therefore, is considered inconsequential to the interpretation of treatment impacts on soybean yield. In 2020, greater yield and lower stand counts were detected from NT treatments. This suggests that reduced stands likely resulted from large amounts of residue interfering with seed placement by the planter. However,

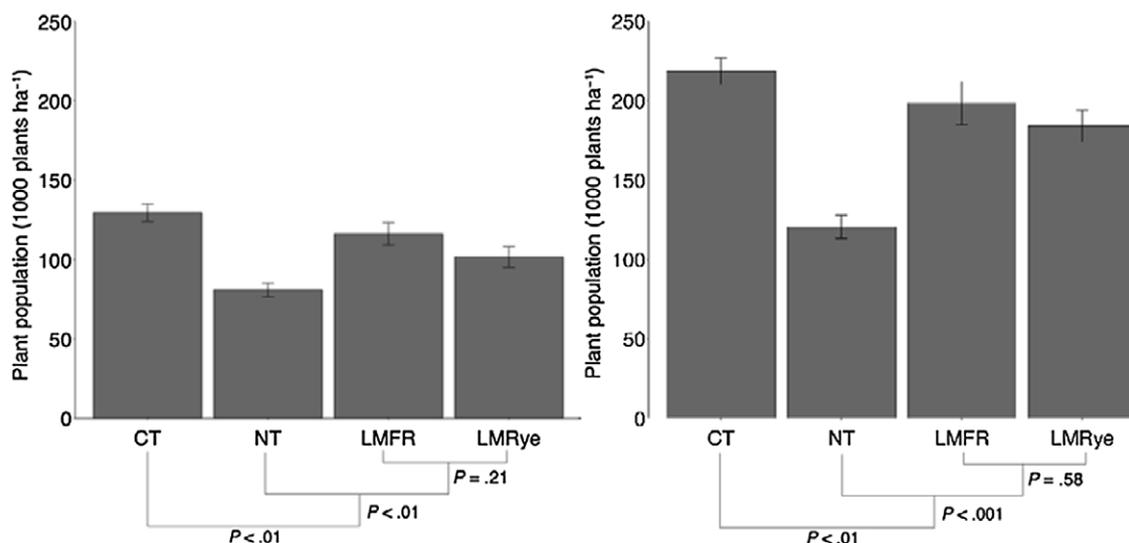


Figure 4. Final soybean stand counts scaled to plants per hectare in 2020 in Upper Marlboro, MD (left) and 2021 in Beltsville, MD (right). Abbreviations: CT, conventional till; NT, no-till; LMFR, living mulch + forage radish residue; LMRye, living mulch + rye residue. P-values represent significance levels for contrasts of groups intersecting at those nodes.

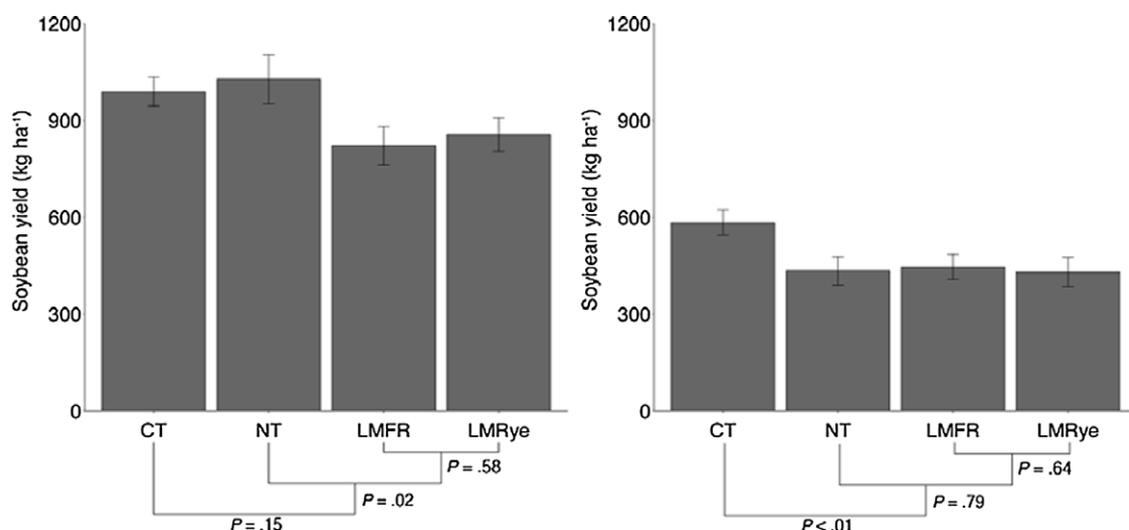


Figure 5. Soybean yield within whole-plot treatments for 2020 in Upper Marlboro, MD (left) and 2021 in Beltsville, MD (right) field seasons. Abbreviations: CT, conventional till; NT, no-till; LMFR, living mulch + forage radish residue; LMRye, living mulch + rye residue. P-values represent significance levels for contrasts of groups intersecting at those nodes.

this reduced stand did not result in yield reductions. In another experiment investigating the impact of conventional tillage and cover crop residue on weed emergence and yield in soybean, Weber et al. (2017) found reduced weed density in NT compared to CT treatments; however, soybean yield was lower from NT treatments than from CT treatments due to poorer soybean stands. However, Weber et al. (2017) credited poorer stand establishment to the seeding equipment not being adequate for planting where cover crop residue was high. Lower yield obtained from living-mulch treatments during the current experiment in 2020 suggests that competition between soybean plants and red clover may have occurred. Yield reductions in soybean–living-mulch systems have been documented previously (Uchino et al. 2009). In 2021, the red clover was not completely terminated by the strip-tiller in the intrarow areas. Still, similar yields between NT and living mulch

treatments indicates that no additional competition occurred between soybean and red clover compared to terminated cover crop residue, possibly as a result of the patchy clover stands. Overall similar yields from Wd and V3 subplots in 2021 suggests that cover crops alone may provide sufficient weed control through the CPWC, while greater yields from V3 compared to Wd in 2020 suggests that additional weed control efforts may be necessary through the CPWC.

Practical Implications

The current experiment highlights the potential for annual cover crops and perennial clovers to be used over multiple growing seasons as part of an IWM program. Self-regenerating annual cover crops and continued establishment of perennial cover crops

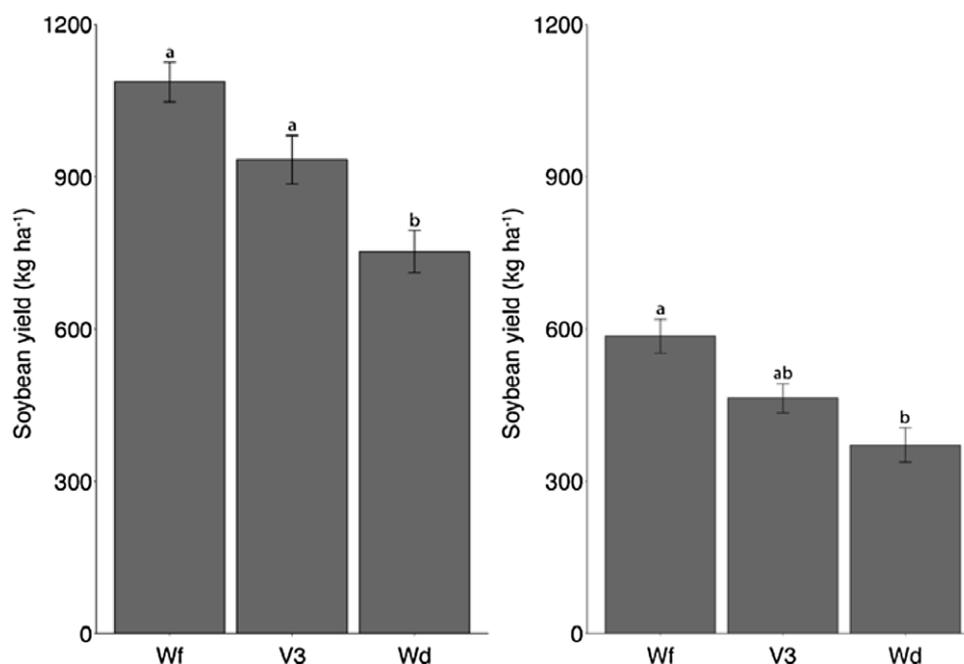


Figure 6. Soybean yield within subplot treatments for 2020 (left) and 2021 (right) field seasons. Abbreviations: V3, weeds controlled through the V3 soybean stage; Wf, weeds controlled through soybean canopy closure; Wd, weedy all season. Bars show means and standard errors with different letters indicating differences between subplot treatments according to Tukey's honestly significant difference test ($P \leq 0.05$).

may be beneficial for growers experiencing challenges such as limited time available for planting following fall harvest. Natural reestablishment may also be beneficial for growers who desire to avoid the added seed and labor expenses associated with replanting. It may also be beneficial to growers who prefer that their cover crops are established earlier in the growing season.

During this experiment, variation in weed species among treatments complicated the findings. Still, taken together, these results highlight the potential of cover crop residues and/or perennial clovers to contribute to an IWM program for the suppression of weed species during the soybean CPWC. The no-till and living mulch operations deployed during this experiment may be especially useful for organic soybean producers who lack good herbicide options and want to reduce in-season tillage. These operations may similarly be useful to conventional producers interested in reducing their herbicide applications. Reductions in weeds resulting from cover crop residues or living mulches may result in fewer spray applications, thereby placing less selection pressure on weeds, and subsequently reducing the likelihood for the development of herbicide resistance. Additional research is needed to better understand the influence of self-regenerated cover crop residues and living mulches on weed maturity and weed seed production, as well as the level of weed suppression provided by other cover crop species. Farmers can then be better informed regarding the benefits and risks of using cover crops to manage weeds under varying conditions.

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