Received: 23 September 2020 | Accepted: 15 December 2020

DOI: 10.1002/vzj2.20110

Vadose Zone Journal

ORIGINAL RESEARCH ARTICLE

Cover crops and tillage effects on carbon–nitrogen pools: A lysimeter study

Gurbir Singh[1] | **Gurpreet Kaur**[1] | **Karl W. J. Williard**[2] | **Jon E. Schoonover**[2]

[1] Delta Research and Extension Center, Mississippi State University, Stoneville, MS 38776, USA

[2] Dep. of Forestry, College of Agricultural Sciences, Southern Illinois Univ., Carbondale, IL 62901, USA

Correspondence
Gurpreet Kaur, Delta Research and Extension Center, Mississippi State Univ., Stoneville, MS 38776, USA.
Email: gk340@msstate.edu

Assigned to Associate Editor Kathleen Lohse.

Abstract

Cover crops (CCs) and tillage practices influence C and N pools in soil, which can affect dissolved organic C (DOC) and N leaching from agricultural fields. Previous studies on cover crops have focused mostly on nitrate leaching and total C (TC). Therefore, a study was conducted in southern Illinois from 2015 to 2018 to evaluate the effects of tillage systems (conventional till [CT] and no-tillage [NT]) and CCs on C and N pools including water-extractable C (WEC), permanganate oxidizable C (POXC), TC, water-extractable N (WEN), and total N (TN) in soil and on TN and DOC leaching collected with zero-tension lysimeters. Crop rotations included were corn (*Zea mays* L.)–soybean [*Glycine max* (L.) Merr.] rotation without winter CC (C–S), corn–cereal rye (*Secale cereale* L.)–soybean–hairy vetch (*Vicia villosa* Roth) (C–R–S–HV), corn–cereal rye–soybean–oat + radish (*Avena sativa* L.+ *Raphanus sativus* L.) (C–R–S–OR). The WEC decreased over time under CT system at a depth to 0–15 cm in rotation C–R–S–OR having cereal rye and oat + radish CCs. The POXC at depths of 15–30 and 30–45 cm increased significantly over time from fall 2015 to spring 2018 for all rotations under both tillage systems. The cumulative DOC leaching was greater in C–R–S–HV rotation than in C–S rotation in fall 2015 and spring 2018. Inclusion of cereal rye in C–R–S–HV and C–R–S–OR rotations reduced cumulative TN leaching compared with the C–S having no CC in spring 2018. Increased DOC leaching losses with the introduction of CC should be addressed and need further evaluation for its impact on C cycling in surface and subsurface waters.

1 | INTRODUCTION

Soil organic C (SOC) is the dominant chemical constituent of soil organic matter. It is a sensitive indicator of soil quality (Brejda et al., 2000) as it affects soil properties (Weil et al., 2003), that affect crop productivity, and environmental quality. Carbon and N pools in soil play various functional roles in soil organic matter dynamics and nutrient cycling (Doran & Parkin, 1994; Scharpenseel & Neue, 1992; Sherrod et al., 2005). The labile fractions of the SOC or active C pool includes microbial biomass C (Islam & Weil, 2000; Kennedy & Papendick, 1995), soil carbohydrates (Saviozzi, Biasci, Riffaldi, & Levi-Minzi, 1999), and particulate organic matter (Janzen, Campbell, Brandt, Lafond, & Townley-Smith,

Abbreviations: C–R–S–HV, corn–cereal rye–soybean–hairy vetch; C–R–S–OR, corn–cereal rye–soybean–oat + radish; C–S, corn–soybean; CC, cover crop; CT, conventional till; DOC, dissolved organic carbon; NT, no-tillage; POXC, permanganate oxidizable carbon; SOC, soil organic carbon; TC, total carbon; TN, total nitrogen; WEC, water-extractable carbon; WEN, water-extractable nitrogen; ZTL, zero-tension lysimeters.

1992; Wander & Bidart, 2000). The permanganate oxidizable C (POXC) test is a fast and affordable soil testing method, which is used for quantifying the labile pool of C in soil (Culman et al., 2012; Weil et al., 2003). The POXC is related to soil quality indicators including microbial biomass C, smaller size and heavier particulate organic C fractions, microbial biomass C, and SOC (Culman et al., 2012; Mirsky et al., 2008; Weil et al., 2003). These studies have also shown that POXC is highly sensitive to changes in management and environments (Culman et al., 2012; Weil et al., 2003). Scharpenseel and Neue (1992) reported that soil factors including temperature, texture, and water content influence the biological transformations among different C pools in soil. The C and N pools in soils can be influenced by topography (Burke, Elliott, & Cole, 1995; Singh, et al., 2018), climate (Burke et al., 1995; Burke et al., 1989), cropping systems (Singh et al., 2018), and management practices such as tillage, cover crops (CCs), N fertilization, and residue management (Doran, 1987; Sainju et al., 2002).

Core Ideas

- Hairy vetch increased cumulative dissolved organic C leaching.
- Cereal rye reduced cumulative total N leaching in spring.
- WEC decreased over time under CT system at depth of 0–15 cm.

Tillage increases aeration, breaks aggregates, incorporates crop residues, and thus accelerates the mineralization of organic C in the soil (Sainju et al., 2002). Tillage places crop residues in the soil, where conditions for the decomposition of residues are more favorable than on the surface of the soil (Christensen, 1986; Douglas et al., 1980; Reicosky & Lindstrom, 1993; Roberts & Chan, 1990). Tillage exposes physically protected SOC while breaking soil aggregates and disrupting soil structure. Tillage also increases losses of SOC via soil erosion and runoff (Paustian et al., 1995). In contrast, conservation tillage practices such as no-till allow retention of crop residues on the soil surface and reduce the loss of C and N by reducing residue decomposition, soil erosion, and runoff (Blanco-Canqui et al., 2013; Varvel & Wilhelm, 2010). Based on a review of the global database of 67 long-term agricultural experiments, West and Post (2002) reported that converting conventional tillage (CT) to no-tillage (NT) can increase C sequestration by 57 ± 14 g C m^{-2} yr^{-1}. Carbon sequestration rates can be expected to reach a peak in 5–10 yr, whereas the SOC will reach a new equilibrium in 15–20 yr (West & Post, 2002). Many previous studies have shown greater organic C and total N (TN) in top soil layers under NT vs. CT systems (Alvarez et al., 1995; Sainju et al., 2010), whereas NT showed mixed results for changes in organic C and N in subsoil layers or whole soil profile (Baker, Ochsner, Venterea, & Griffis, 2007; Blanco-Canqui & Lal, 2008).

Among conservation practices other than conservation tillage, CCs increase SOC content by the addition of biomass inputs to soil (Blanco-Canqui et al., 2013). Cover cropping provides multiple benefits including reduction of weed populations, soil erosion and nutrient leaching, and addition of organic matter (Blanco-Canqui et al., 2015; Meisinger & Ricigliano, 2017; Singh, Williard, & Schoonover, 2018; Singh, Thilakarathne, et al., 2020). However, the benefits of CCs on soil properties and crop yields depend on CC species, amount of biomass produced, length of CC growing season, cropping system, and precipitation input (Blanco-Canqui et al., 2013; Singh, Williard, & Schoonover, 2018; Singh et al., 2019). For example, multiple studies have shown that cereal rye (*Secale cereale* L.) CC reduces nitrate leaching to groundwater and surface waters by scavenging residual N from the soil after harvest of cash crop (Brandi-Dohrn et al., 1997; Meisinger & Ricigliano, 2017; Staver & Brinsfield, 1998). However, mixed results have been reported by previous studies on the efficacy of CCs in reducing nitrate leaching (Lewan, 1994; Meisinger & Ricigliano, 2017; Meisinger, Hargrove et al., 1991; Meisinger et al., 1990). Multiple factors affect N scavenging by CCs including the amount of rainfall received during the CC growing season (Meisinger & Ricigliano, 2017), amount of residual N after harvest of cash crop (Kuo & Jellum, 2002), type of CC species (legume vs grasses), and CCs planting and termination timing (Mirsky, Ackroyd, et al., 2017; Mirsky, Spargo, et al., 2017). Similarly, CCs have shown mixed results for their impact on soil C and N pools. Rorick and Kladivko (2017) and Beehler, Fry, Negassa, and Kravchenko (2017) reported that cereal rye did not cause any significant changes in SOC and TN. However, Sainju et al. (2002) found that rye increased organic C and N by 3–4%, whereas the hairy vetch (*Vicia villosa* Roth) reduced organic C and N by 1% in a 7-yr period from 1994 to 2000 in a study conducted in Georgia, USA. In their study, Sainju et al. (2002) reported that the use of hairy vetch and cereal rye CCs increased organic C and N concentrations as compared with treatments without CCs. In California, increasing the frequency of legume–rye CC mix (annual vs. quadrennial) in a rotation led to an increase in SOC by 3.4 Mg ha^{-1} and POXC by 26% after 8 yr of study (White, Brennan, Cavigelli, & Smith, 2020). Tautges, Chiartas, Gaudin, O'Green, Herrera, and Scow (2019) found that winter CC mix including hairy vetch, field pea (*Pisum sativum* L.), faba bean (*Vicia faba* L.), and cereal oat (*Avena sativa* L.) increased the SOC stocks in the top 30-cm soil profile by 3.5% but reduced SOC stocks by 10.8% in the soil profile at 30-to-200-cm depth in a period from 1993 to 2012. In a global meta-analysis of 30 studies, Poeplau and Don (2015) found that soil C stocks increase by

0.32 Mg C ha^{-1} yr^{-1} by CCs in the top 22-cm soil depth. Ye and Hall (2020) reported that decomposition of slow-and fast-cycling C pools was increased by rye CC, which reduced the size of the pools across all soil depths compared with treatments without any CC. The authors concluded that CC did not result in faster C sequestration than the conventional annual bioenergy systems in their study (Ye & Hall, 2020). It is difficult to detect the impact of CCs on SOC, with reports of positive, negative, and neutral effects on SOC stocks (et al., 2010; Poeplau & Don, 2015).

Previous studies have mostly focused on individual effects of CCs or tillage on SOC content or POXC. However, little is known about the effects of CCs and tillage on dissolved organic C (DOC) and water-extractable C (WEC). Organic matter usually consists of <1% WEC and DOC (Chantigny, 2003; Zsolnay, 1996). Water-extractable C is the most labile pool of soil organic matter and is readily decomposable material available to soil microbes (Gregorich et al., 1994). In northeastern Illinois on Drummer (fine-silty, mixed, superactive, mesic Typic Endoaquolls) and Elpaso (fine-silty, mixed, superactive, mesic Typic Endoaquolls) silt clay loam, cereal rye–daikon radish (*Raphanus sativus* L.) CC in continuous corn (*Zea mays* L.) cropping system showed no significant differences in WEC compared with no-CC treatments at soil depths of 0–5 and 5–20 cm (Grebliunas, Armstrong, & Perry, 2016). However, the authors observed greater water-extractable organic C after the termination of CCs as compared with the sampling date before the CC termination. Cereal rye–Daikon radish did not alter the quantity and quality of water-extractable organic C after 3 yr of study (Grebliunas et al., 2016). White et al. (2020) found that increasing the frequency of CCs (annual vs. quadrennial) altered the SOC composition through an increase in labile C proportions.

Excess labile C can be detrimental to N in soil as it promotes bacterial activity and can lead to increased denitrification rates in soil as well as in receiving waterways (Grebliunas et al., 2016; Vinther, Hansen, & Eriksen, 2006). In addition, leaching of DOC from agricultural landscapes due to changes in management practices can have wide impacts on surface and groundwater quality, aquatic ecosystems, and upland C balances (Evans, Monteith, & Cooper, 2005). Greater DOC leaching under the CCs as compared with bare soil or treatments with no CCs has been reported by Vinther et al. (2006) and Steenwerth and Belina (2008). However, dissolved organic N was lower under CCs, which resulted in lower TN losses (Vinther et al., 2006). Dissolved organic C can act as a source of energy for the denitrifying bacteria in the soil, which will reduce nitrate leaching losses to groundwater (Vinther et al., 2006). Soil water content and the C inputs to soil determine the leaching of C (Steenwerth & Belina, 2008; Vinther et al., 2006). Kindler et al. (2011) reported that the amount of DOC leaching in the topsoil layers was positively related to their C/N ratio, whereas in subsoils, DOC retention was inversely related to the ratio of organic C to Fe + Al hydroxides. The DOC can provide C for microbial respiration and can be positively related to soil C dioxide efflux (Steenwerth & Belina, 2008). There is limited research available on the interactive effects of tillage and CCs on water-extractable forms of soil C and N in corn–soybean [*Glycine max* (L.) Merr.] rotation systems. Previous research mostly focused on CCs' impact on nitrate leaching. However, it is also important to understand the impact of CCs under different tillage systems on leaching of DOC in a corn–soybean rotation.

Therefore, the objective of this study was to evaluate the effect of tillage and CCs on soil C and N pools including POXC, WEC, water-extractable N (WEN), total C (TC), and TN, as well as on leaching of DOC and TN. We hypothesized that corn–soybean rotation with CC and NT systems will help build active pools of C and N in soil by adding more biomass inputs in the soil, and that it can lead to increased losses of DOC. However, the TN leaching losses might be reduced with use of nonlegume CCs compared with legume CCs.

2 | MATERIALS AND METHODS

2.1 | Experiment layout and crop management practices

The study site was established in fall 2013 with three crop rotations and two tillage systems at Southern Illinois University Farm in Carbondale, IL (37°71′06′′ N, 89°27′33′′ W). The experimental design was a completely randomized design with three replications. Three crop rotations included in this study were: corn–cereal rye–soybean–hairy vetch (C–R–S–HV), corn–cereal rye–soybean–oat + radish (C–R–S–OR), and corn–soybean rotation without winter CC (C–S). The two tillage systems included were CT and NT. Crop rotations were setup as a 2-yr rotation to complete one cycle of a rotation with a plot size of 18 × 120 m for each treatment. The dominant soil series at the study site was Hosmer silt loam (fine-silty, mixed, active, mesic Oxyaquic Fragiudalfs). The mean 20-yr (1994–2013) annual total precipitation was 1,135 mm with a mean annual temperature of 13 ˚C at the study site (Illinois Climate Network, 2019). This study was not irrigated, and the field was not tile drained.

The field used for this research was under CT before the start of this experiment until fall 2013. Details of field operations, planting dates, harvesting or termination dates, and sample collection dates are provided in Table 1. Cover crops were planted after harvesting cash crops in fall and terminated in spring using Roundup Weathermax (glyphosate) at 1.26 kg acid equivalent ha^{-1}, 2,4-D (2,4-dichlorophenoxyacetic acid) at 0.80 kg ae ha^{-1}, and ammonium sulfate (AMS) 2.5% (v/v). Tillage treatments were performed in the spring after the termination of CCs (Table 1). Cover crop residue was

TABLE 1 Crop sequence for different rotations from 2014 to 2018 and dates for field operations and data collection for the cover crop (CC) and tillage study

Crop rotation	Oct. 2014–Apr. 2015	May 2015–Sept. 2015	Sept. 2015–Apr. 2016	May 2016–Oct. 2016	Oct. 2016–Apr. 2017	Apr. 2017–Sept. 2017	Oct. 2017–Apr. 2018
C-S	No CC	Corn	No CC	Soybean	No CC	Corn	No CC
C-R-S-HV	Hairy vetch	Corn	Cereal rye	Soybean	Hairy vetch	Corn	Cereal rye
C-R-S-OR	Oat + radish	Corn	Cereal rye	Soybean	Oat + radish	Corn	Cereal rye
Field operations	**Dates**						
Tillage	–	8 May 2015	–	3 June 2016	–	25 Apr. 2017	–
N fertilizer application	–	20 May 2015	–	–	–	9 May 2017	–
Planting	9 Oct. 2014	3 June 2015	28 Sept. 2015	10 June 2016	17 Oct. 2016	9 May 2017	6 Oct. 2017
Termination/harvest	24 Apr. 2015	18 Sept. 2015	28 Apr. 2016	10 Oct. 2016	13 Apr. 2017	27 Sept. 2017	28 Apr. 2018
End of season	Spring 2015	Fall 2015	Spring 2016	Fall 2016	Spring 2017	Fall 2017	Spring 2018
Soil Sampling	24 Apr. 2015	28 Sept. 2015	27 Apr. 2016	13 Oct. 2016	20 Apr. 2017	2 Oct. 2017	18 Apr. 2018
Soil solution collection events	–	7	14	6	12	4	9

Note. C–S, corn–soybean; C–R–S–HV, corn–cereal rye–soybean–hairy vetch; C–R–S–OR, corn–cereal rye–soybean–oat + radish. Cover crop was terminated using Roundup Weathermax (glyphosate) at 1.26 kg acid equivalent (a.e.) ha^{-1}, 2,4-D (2,4-dichlorophenoxyacetic acid) at 0.80 kg a.e. ha^{-1}, and ammonium sulfate (AMS) 2.5% (v/v). Soil solution collection events are the total number of water sampling events per season (if the end of the season is spring 2018, then the number of soil solution collection events between planting and termination of cover crops was nine).

incorporated in the soil after burndown application with two passes of disking, followed by a single pass of the roller before planting of the cash crops in the CT treatments. No such tillage operations were performed in the NT treatments. Soybean and corn were planted with a John Deere seed drill at a row spacing of 76.2 cm using seeding rates of 395,000 and 71,000 seeds ha^{-1}, respectively. Soybean received a pre-plant application of 23 kg P ha^{-1} as diammonium phosphate (DAP). Corn received a pre-plant application of 179–34–53 N–P–K kg ha^{-1} as urea, DAP, and muriate of potash (KCl), respectively. All fertilizers were broadcast applied. Cover crops were planted after the harvest of grain crops using a grain drill at a row spacing of 19.1 cm. Cereal rye, hairy vetch, and oat + radish were planted at a seeding rate of 87, 28, and 39 kg ha^{-1}, respectively. A ratio of 16:1 was used for the oat + radish mix.

2.2 | Zero-tension lysimeter installation

Zero-tension lysimeters (ZTLs) were installed in the transition horizon of soil at a depth varying between 23 and 30 cm in all CC and tillage treatments in February 2015. Hosmer soils have a distinct A horizon with a restricting subsurface B horizon. Restricting subsoil creates a perched water table during spring precipitation events; therefore, ZTLs were installed above the restricting layer to prevent potential groundwater or perched water movement in the lysimeters. Zero-tension lysimeters had a surface area of 0.086 m^2 and were installed in an undisturbed soil by excavating pit and digging the sidewall of the soil pit to install soil solution collection pans. Lysimeters were connected to a conical collection container of 22.7 L (0.0227 m^3) capacity. Details of ZTLs installation and design were discussed in Singh, Kaur, Williard, Schoonover, and Kang (2018). After installing ZTLs, excavated soil pits were backfilled and two tubes—the soil solution sample collection tube and the zero-tension tube—were the only tubes that were exposed to the soil surface. Zero-tension lysimeters were left for 17 wk for calibration in the field. Soil solution samples collected after installation until the beginning of June 2015 were not used for any analysis.

2.3 | Data collection and analysis

Soil samples were collected twice a year that is after the harvest of grain crop but before planting of CC and after the termination of CC but before tillage and grain crop planting (Table 1). A total of six soil sampling events occurred starting fall 2015 until spring 2018. The soil samples from spring 2015 were not archived to be used later for the C and N pools analysis for the current study. Soil samples were collected within a 5-m^2 area of ZTLs to a depth of 45 cm in increments of 15 cm (0–15, 15–30, and 30–45 cm) using a push probe (JMC soil samplers). In total, seven soil cores were collected for each depth around ZTLs and composited for sample analysis. Collected soil samples were air dried, ground using a soil grinder (Gilson Company), passed through a 2-mm sieve opening, and analyzed for TC and TN using a CNS analyzer (Thermo Fisher Scientific). For WEC and WEN extraction, 10 g of soil sample was shaken with 40 ml of deionized water for 60 min using an oscillating shaker at 120 rpm. The extracted soil solution was filtered using MF-Millipore membrane filter (0.45-µm pore size) and analyzed for WEC and WEN on a Shimadzu TOC-L analyzer (Shimadzu Corporation)

(Burford & Bremner, 1975). The POXC was analyzed using the method described by Weil et al. (2003) with some modifications to account for the elevated quantities of C that might occur in soil samples from CC treatments (Singh et al., 2018). The shaking time was increased from 2 to 15 min, and the volume to weight ratio of 0.02 mol L^{-1} $KMnO_4$ was changed from 8:1 to 16:1. Soil samples for bulk density were also collected to a depth of 45 cm in 15-cm increments and were used for converting C and N concentrations to a Mg ha^{-1} or kg ha^{-1} basis. The cash crop and CC biomass was also collected from each plot to determine biomass yield, C/N ratio, and N uptake in this study (Singh, Thilakarathne, et al., 2020; Supplemental Tables S3 and S4).

Soil solution samples after corn planting in June 2015 until cereal rye termination in April 2018 were collected after every significant rainfall event (>20 mm) from ZTLs. In total, soil solution samples were collected 52 times over the study period from June 2015 to April 2018 (Table 1). Soil solution samples were collected through the exposed sample collection tubes using a self-priming pump. Collected soil solution samples were measured for their total volume. A subsample of soil solution of ∼500 ml was collected in a bottle and transferred to Southern Illinois University water quality laboratory. In the water quality laboratory, subsamples were vacuum filtered using 0.45-μm filter disks. Filtered soil solution samples were analyzed for DOC and TN using a Shimadzu TOC-L analyzer (Shimadzu Instruments). The nonpurgeable organic C (NPOC) was measured and samples were acidified and sparged for analysis of DOC. All soil solution data in concentration (mg L^{-1}) were converted to DOC and TN loads (kg ha^{-1}) based on recorded volume from ZTLs in the field and the area of ZTLs (0.086 m^2) as

$$\text{DOC}\ \left(\text{kg ha}^{-1}\right) = \frac{\text{DOC}\ \left(\text{mg L}^{-1}\right)\ \times \text{volume}\ (\text{L})}{0.086\ \left(\text{m}^2\right)} \times 0.01$$

$$\text{TN}\ \left(\text{kg ha}^{-1}\right) = \frac{\text{TN}\ \left(\text{mg L}^{-1}\right)\ \times \text{volume}\ (\text{L})}{0.086\ \left(\text{m}^2\right)} \times 0.01$$

2.4 | Statistical analysis

The UNIVARIATE procedure in SAS version 9.4 (SAS Institute) was used for testing the normality of data. Based on Shapiro–Wilk and Kolmogorov–Smirnov tests used for determining the normality of data, several variables were log-transformed for analysis and were back-transformed for the presentation of results. The experimental design was a completely randomized design with three replications. All soil and water data were analyzed by season using the GLIMMIX procedure in SAS. For analyzing soil data, the independent variables were crop rotation, tillage, depth, and their interaction and the dependent variables were WEN, TN, WEC, POXC, and TC. Replications for the treatments were treated as a random factor. The three-way interaction between crop rotation, tillage, and depth was not significant. However, the two-way interaction between crop rotation and tillage as well as between crop rotations and depth were significant for at least a season. For comparison of means, all variables were analyzed using the T-grouping and least-squares means were calculated at $\alpha = .1$ (Tables 2 and 3). Additionally, to analyze the trends for building labile or bulk pools of C and N, we split out the crop rotation and tillage based on depth or combined over depths and regressed the data over time (six soil collection events). The regression analysis was performed using the REG procedure in SAS at $\alpha = .1$.

The crops grown in a season changed over a 2-yr crop rotation period, therefore, soil solution data were split by season for soil solution analysis. For example, cereal rye CC planted after corn harvest in fall 2015 had 14 soil solution collection events (Table 1). The 14 soil solution collection events were analyzed together, and the mean values from the analysis given in Table 5 were called cover crop spring 2016. Similarly, there were six soil solution collection events collected between planting and harvesting of soybean in 2016 and the end of the season was called fall 2016. Again, the six soil solution collection events were analyzed together for soybean fall 2016. Soil solution samples were analyzed using a repeated measure model in SAS using the GLIMMIX procedure. The independent variables were crop rotation, tillage, and their interaction and dependent variables were DOC and TN. A repeated measure statement for collection events was added having an exponential spatial or temporal covariance structure type = SP[EXP(c-list)]. The significant differences between the mean values were determined using a post hoc T-grouping analysis at the α level of .1. Cumulative DOC and TN loss from the ZTLs for a season were calculated by the summation of loads for a season. Cumulative DOC and TN loss were analyzed using the GLIMMIX procedure and post hoc T-grouping analysis was used for determining the significant differences.

3 | RESULTS

3.1 | Carbon pools

3.1.1 | Soil

The WEC, POXC, and TC mostly decrease with depth for all rotations and tillage systems (Tables 2 and 3). The WEC was affected by the interaction of crop rotation and depth during the corn growing season in fall 2015 and 2017 (Table 2, Supplemental Table S1). When data was averaged over tillage systems, rotations with CCs (C–R–S–HV and C–R–S–OR)

TABLE 2 Mean water-extractable C (WEC) and permanganate oxidizable C (POXC) in soil as affected by the crop rotation, tillage, and depth. Within a column and within a given factor or combination of factors, means followed by the same letter are not statistically different (α = .10)

Crop rotation[a]	Tillage[b]	Depth	Corn, fall 2015		Cover crop, spring 2016		Soybean, fall 2016		Cover crop, spring 2017		Corn, fall 2017		Cover crop, spring 2018	
			WEC	POXC	WEC	POXC	WEC	POXC	WEC	POXC	WEC	POXC	WEC	POXC
		cm	kg ha^{-1}											
C–S			176b	599	187	635	125	809	111	815	126	858	274	853
C–R–S–HV			197a	648	193	635	123	793	122	799	135	830	274	913
C–R–S–OR			187ab	679	192	642	118	778	106	769	129	829	261	858
	CT		185	683	194	659	122	811	117	846a	123b	868	273	889
	NT		188	601	187	616	122	775	109	743b	136a	810	266	861
		0–15	192a	1,522a	292	1,500a	215a	1,514a	188a	1,530a	196a	1,480a	286a	1,566a
		15–30	135b	327b	144	351b	82b	558b	81b	561b	103b	639b	250b	645b
		30-–45	132b	77c	136	61c	68b	307c	69b	293c	91c	398c	273ab	413c
C–S	CT		171	674	191	658	125	859	124	879	130ab	953a	283	916
C–S	NT		180	524	183	612	125	759	97	751	121ab	762c	265	790
C–R–S–HV	CT		197	648	195	629	121	805	125	854	124ab	841b	286	913
C–R–S–HV	NT		196	648	190	641	124	780	119	744	145a	820b	262	913
C–R–S–OR	CT		188	728	196	690	120	770	102	806	115b	809b	250	838
C–R–S–OR	NT		186	631	187	594	116	785	110	733	142a	848b	271	879
C–S		0–15	259b	1,359	286	1,588	220	1,612	187	1,633	172a	1,543	308	1,654
C–S		15–30	136c	327	148	284	85	528	79	544	102b	631	253	527
C–S		30–45	132c	111	128	33	69	287	66	268	103b	399	261	379
C–R–S–HV		0–15	308a	1,566	303	1,508	219	1,479	201	1,515	213a	1,456	300	1,582
C–R–S–HV		15–30	134c	310	141	334	80	583	88	568	107b	648	244	699
C–R–S–HV		30–45	148c	69	135	62	68	315	77	315	85b	387	279	458
C–R–S–OR		0–15	310a	1,643	287	1,405	206	1,451	175	1,442	202a	1,441	250	1,462
C–R–S–OR		15–30	128c	345	143	436	82	564	76	570	99b	638	252	710
C–R–S–OR		30–45	124c	50	145	86	66	318	67	296	85b	407	280	403

[a]Crop rotation: C–S, corn–soybean; C–R–S–HV, corn–cereal rye–soybean–hairy vetch; C–R–S–OR, corn–cereal rye–soybean–oat + radish.
[b]Tillage: CT, conventional tillage; NT, no-tillage.

TABLE 3 Total C in soil as affected by the crop rotation, tillage, and depth. Within a column and within a given factor or combination of factors, means followed by the same letter are not statistically different (α = .10)

Crop rotation[a]	Tillage[b]	Depth	Total C Corn, fall 2015	Cover crop, spring 2016	Soybean, fall 2016	Cover crop, spring 2017	Corn, fall 2017	Cover crop, spring 2018
		cm	Mg ha^{-1}					
C–S			12.9	12.2	12.3	12.3	12.2	11.6
C–R–S–HV			12.1	11.6	11.6	11.6	11.8	12.0
C–R–S–OR			12.9	11.6	11.6	11.6	11.6	11.5
	CT		13.0	12.2	12.3	12.3a	12.4	12.0
	NT		12.2	11.4	11.4	11.4b	11.3	11.4
		0–15	22.6a	21.3a	21.7a	21.7a	21.2a	20.9a
		15–30	9.2b	8.8b	8.6b	8.6b	8.9b	8.7b
		30–45	6.1c	5.2c	5.3c	5.3c	5.6c	5.5c
C–S	CT		13.7	12.7	13.1	13.1	14.1	12.3
C–S	NT		12.0	11.7	11.6	11.6	10.3	10.8
C–R–S–HV	CT		12.1	11.8	12.0	12.0	11.8	12.2
C–R–S–HV	NT		12.1	11.3	11.2	11.2	11.9	11.9
C–R–S–OR	CT		13.2	12.1	11.8	11.8	11.4	11.5
C–R–S–OR	NT		12.5	11.1	11.5	11.5	11.8	11.5
C–S		0–15	22.8	22.5	23	23	22.3	21.4
C–S		15–30	9.3	8.8	8.5	8.5	8.6	8.1
C–S		30–45	6.5	5.3	5.5	5.5	5.7	5.3
C–R–S–HV		0–15	21.7	21.1	21.2	21.2	21.1	21.5
C–R–S–HV		15–30	8.8	8.4	8.6	8.6	8.7	8.8
C–R–S–HV		30–45	5.8	5.2	5.1	5.1	5.7	5.9
C–R–S–OR		0–15	23.2	20.5	20.9	20.9	20.2	19.9
C–R–S–OR		15–30	9.5	9.4	8.7	8.7	9.3	9.3
C–R–S–OR		30–45	5.9	5.0	5.4	5.4	5.3	5.4

[a]Crop rotation: C–S, corn–soybean; C–R–S–HV, corn–cereal rye–soybean–hairy vetch; C–R–S–OR, corn–cereal rye–soybean–oat + radish.
[b]Tillage: CT, conventional tillage; NT, no-tillage.

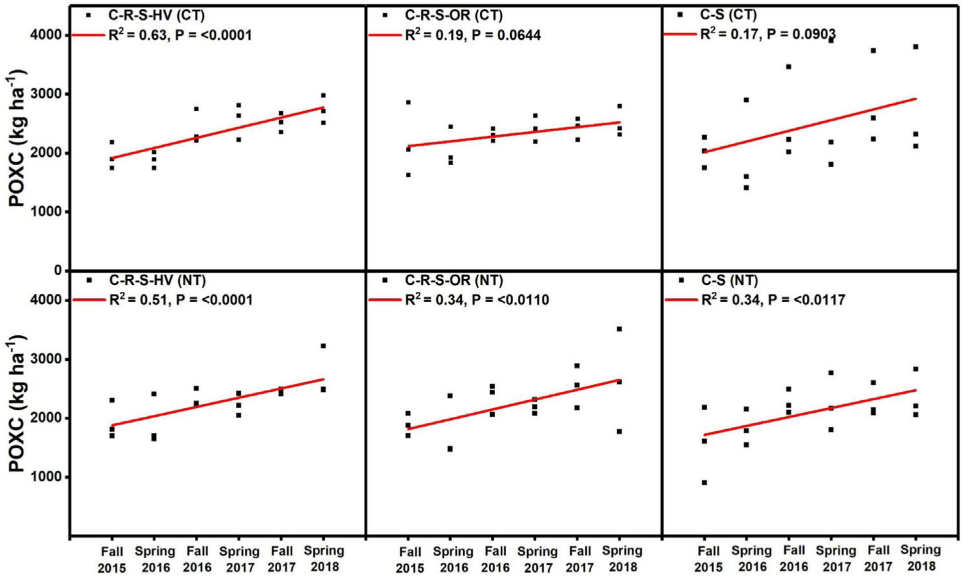

FIGURE 1 Linear regression between permanganate oxidizable C (POXC, summed by depth) and time of collection of soil samples. The three squares indicate three replicates for each crop rotation and tillage treatments at an individual soil collection time. Rotations were corn–soybean (C–S), corn–cereal rye–soybean–hairy vetch (C–R–S–HV), and corn–cereal rye–soybean–oat + radish (C–R–S–OR). Tillage treatments were conventional tillage (CT) and no-tillage (NT)

resulted in 49 to 51 kg ha^{-1} greater WEC than the C–S rotation at a depth of 0–15 cm in fall 2015 during the corn growing season. Within each rotation, the WEC was greater in surface soil depth (0–15 cm) than at deeper soil layers (15–30 and 30–45 cm) during corn growing season in fall 2015 and 2017. No differences were obtained due to rotations and tillage for WEC and POXC in 2016. During the CC growing season in spring 2017, the CT system increased the POXC and TC by 103 kg ha^{-1} and 0.9 Mg ha^{-1} compared with the NT system, when data were averaged over rotations and depth (Tables 2 and 3).

During the corn growing season in fall 2017, the interaction of crop rotation and tillage significantly affected the WEC and POXC content in soil (Table 2, Supplemental Table S1). The NT increased WEC by 27 kg ha^{-1} than CT for C–R–S–OR rotation, whereas it reduced the POXC by 191 kg ha^{-1} than CT for C–S rotation without any CCs. The C–S rotation having no CCs had 112 and 144 kg ha^{-1} higher POXC than the C–R–S–HV and C–R–S–OR rotations, respectively, under the CT system in fall 2017 during the corn growing season (Table 2). However, the trend was opposite for the POXC under the NT system in fall 2017, where rotations with CCs had greater POXC than the C–S rotation under NT system.

The WEC decreased over time at a depth of 0–15 cm from fall 2015 to spring 2018 under the CT system in C–R–S–OR rotation having cereal rye and oat + radish CCs, as indicated by the linear regression between WEC and collection timing of soil samples (Table 4). However, WEC increased with time at a depth of 30–45 cm under the NT system in C–S rotation without any CCs, as well as in C–R–S–OR rotation having cereal rye and oat + radish CCs. The POXC in soil depths 15–30 and 30–45 cm increased significantly over time from fall 2015 to spring 2018 for all rotations under both tillage systems (Table 4, Figure 1). Total C in soil showed no changes over time in this study from fall 2015 to spring 2018, except a decrease with time at depth 30–45 cm under CT system in C–R–S–OR rotation having rye and oat + radish CCs.

3.1.2 | Water

The mean DOC leaching loss as measured by the ZTL was affected by the interaction of tillage and crop rotation in all seasons, except in fall 2017 and spring 2018 (Table 5). No-tillage increased mean DOC leaching compared to CT by 4 and 3.8 kg ha^{-1} for C–R–S–HV and C–R–S–OR rotations having hairy vetch and oat + radish CCs, respectively, in fall 2015 during the corn growing season. Within the NT system, hairy vetch CC in C–R–S–HV and oat + radish CC in C–R–S–OR increased mean DOC leaching by 6.2 and 4.4 kg ha^{-1} than C–S rotation without any CCs, respectively, in fall 2015. In spring 2016 during the CC growing season within the NT system, the cereal rye CC in C–R–S–OR rotation resulted in 4.6 kg ha^{-1} greater mean DOC leaching than the C–S rotation without any CCs. Mean DOC leaching was 3.6 and 6.6 kg ha^{-1} greater under the CT system than under NT in C–S and C–R–S–HV rotations, respectively, in fall 2016. Under the NT system in fall 2016 during the soybean growing season, the C–R–S–HV had 6.6 and 3.9 kg ha^{-1} lower mean DOC leaching than the C–R–S–OR and C–S, respectively. In spring 2017

TABLE 4 Depth-wise linear regression between C (water-extractable C [WEC], permanganate oxidizable C [POXC], and total C [TC]) pools and collection time of soil samples

			WEC				POXC				TC			
			Variables				Variables				Variables			
Crop rotation[a]	Tillage[b]	Depth	Intercept	Slope	Pr > *F*	R^2	Intercept	Slope	Pr > *F*	R^2	Intercept	Slope	Pr > *F*	R^2
		cm	———kg ha^{-1}———				———kg ha^{-1}———				———Mg ha^{-1}———			
C–R–S–OR	CT	0–15	328.41	−24.92	.0029	.43	1,623.97	−30.52	.4500	.04	24.15	−0.70	.1315	.14
		15–30	73.55	14.99	.1002	.16	380.66	49.31	.0016	.47	9.54	−0.04	.7641	.01
		30–45	68.11	14.47	.1638	.12	33.09	61.87	<.0001	.65	5.67	−0.20	.0592	.21
	NT	0–15	264.99	−8.48	.3117	.06	1,484.50	−15.39	.6604	.01	21.34	−0.23	.5245	.03
		15–30	89.78	12.67	.1159	.15	219.62	89.95	.0003	.57	9.07	−0.04	.8224	.00
		30–45	67.59	19.84	.0700	.19	−54.38	92.74	<.0001	.75	5.36	0.06	.7471	.01
C–R–S–HV	CT	0–15	286.80	−7.78	.3146	.06	1,510.07	0.14	.9922	.00	22.11	0.01	.9736	.00
		15–30	85.34	13.74	.1187	.15	274.05	85.30	.0018	.47	9.10	0.05	.8671	.00
		30–45	80.70	14.43	.2070	.10	−41.46	86.72	<.0001	.83	4.94	0.07	.4744	.03
	NT	0–15	293.99	−11.16	.1925	.10	1,533.45	−2.52	.9090	.00	20.91	0.04	.8888	.00
		15–30	87.66	12.41	.0903	.17	198.77	78.79	.0002	.58	8.01	−0.03	.8277	.00
		30–45	80.10	15.02	.1354	.13	−6.85	80.19	<.0001	.78	5.38	0.08	.7466	.01
C–S	CT	0–15	236.11	2.71	.7814	.01	1,512.61	51.48	.4536	.04	24.54	0.13	.9174	.00
		15–30	91.71	13.08	.1255	.14	299.21	60.65	.0919	.17	9.83	−0.20	.6327	.01
		30–45	71.29	16.47	.1077	.15	21.19	69.69	.0005	.54	6.00	−0.11	.4257	.04
	NT	0–15	267.35	−10.07	.2378	.09	1,344.60	26.24	.4546	.04	21.93	−0.55	.1255	.14
		15–30	87.68	12.05	.1838	.11	236.55	56.83	.0039	.42	8.72	−0.20	.4754	.03
		30–45	68.42	15.84	.0888	.17	−13.27	68.68	<.0001	.64	6.25	−0.18	.3243	.06

[a]Crop rotation: C–S, corn–soybean; C–R–S–HV, corn–cereal rye–soybean–hairy vetch; C–R–S–OR, corn–cereal rye–soybean–oat + radish.
[b]Tillage: CT, conventional tillage; NT, no-tillage.

TABLE 5 Mean and cumulative dissolved organic C collected using zero-tension lysimeters installed below A horizon. Within a column and within a given factor or combination of factors, means followed by the same letter are not statistically different (α = .1)

		Dissolved reactive C											
		Corn, fall 2015		Cover crop, spring 2016		Soybean, fall 2016		Cover crop, spring 2017		Corn, fall 2017		Cover crop, spring 2018	
Crop rotation[a]	**Tillage**[b]	**Mean**	**Cumulative**	**Mean**	**Cumulative**	**Mean**	**Cumulative**	**Mean**	**Cumulative**	**Mean**	**Cumulative**	**Mean**	**Cumulative**
		kg ha^{-1}											
C–S		3.0b	19.5b	3.4b	52.7	11.8	36.5	4.5a	79.2	0.8	1.8	4.3b	51.1
C–R–S–HV		6.9a	44.9a	2.4ab	42.4	9.4	30.8	0.6b	49.2	1.9	2.7	9.4a	93.2
C–R–S–OR		5.2a	28.9ab	5.3a	77.4	14	48.5	4.1a	79.4	0.3	0.6	7.4ab	74.8
	CT	3.9b	25.1	4.5a	67a	14a	48.7a	5.6a	104.1a	1.6	2.7	8.3	84.7
	NT	6.2a	37.1	2.8b	47.9b	9.4b	28.5b	0.5b	34.4b	0.3	0.7	5.8	61.4
C–S	CT	3.4c	21.7	5.6ab	26.1	13.6a	46.8	8.6a	133.4a	1.5	3.1	5.8	64.8ab
C–S	NT	2.7c	17.4	1.1b	79.2	10b	26.3	0.3c	25b	0.2	0.5	2.8	37.4b
C–R–S–HV	CT	4.9bc	30.8	3.1ab	34.2	12.7ab	44.3	0.8c	64.8ab	3.3	4.5	12.3	120.8a
C–R–S–HV	NT	8.9a	58.9	1.7ab	50.5	6.1c	17.4	0.5c	33.6ab	0.5	0.9	6.5	65.5ab
C–R–S–OR	CT	3.3c	22.7	4.9ab	83.5	15.7a	54.9	7.4a	114.2ab	0.2	0.5	6.7	68.4ab
C–R–S–OR	NT	7.1ab	35.1	5.7a	71.4	12.2ab	42	0.8c	44.6ab	0.3	0.7	8.2	81.2ab
Source of variation	df	*p* value											
Rotation	2	.0031	.0637	.0101	.4847	.5336	.5524	.0325	.3375	.6034	.2776	.0541	.1375
Tillage	1	.0073	.1492	.0002	.0962	<.0001	.0226	.0013	.0224	.1508	.6486	.9123	.1661
Rotation × tillage	2	.0189	.2737	.0001	.3968	.0571	.576	<.0001	.0749	.1942	.4308	.1126	.0481

[a]Crop rotation: C–S, corn–soybean; C–R–S–HV, corn–cereal rye–soybean–hairy vetch; C–R–S–OR, corn–cereal rye–soybean–oat + radish.
[b]Tillage: CT, conventional tillage; NT, no-tillage.

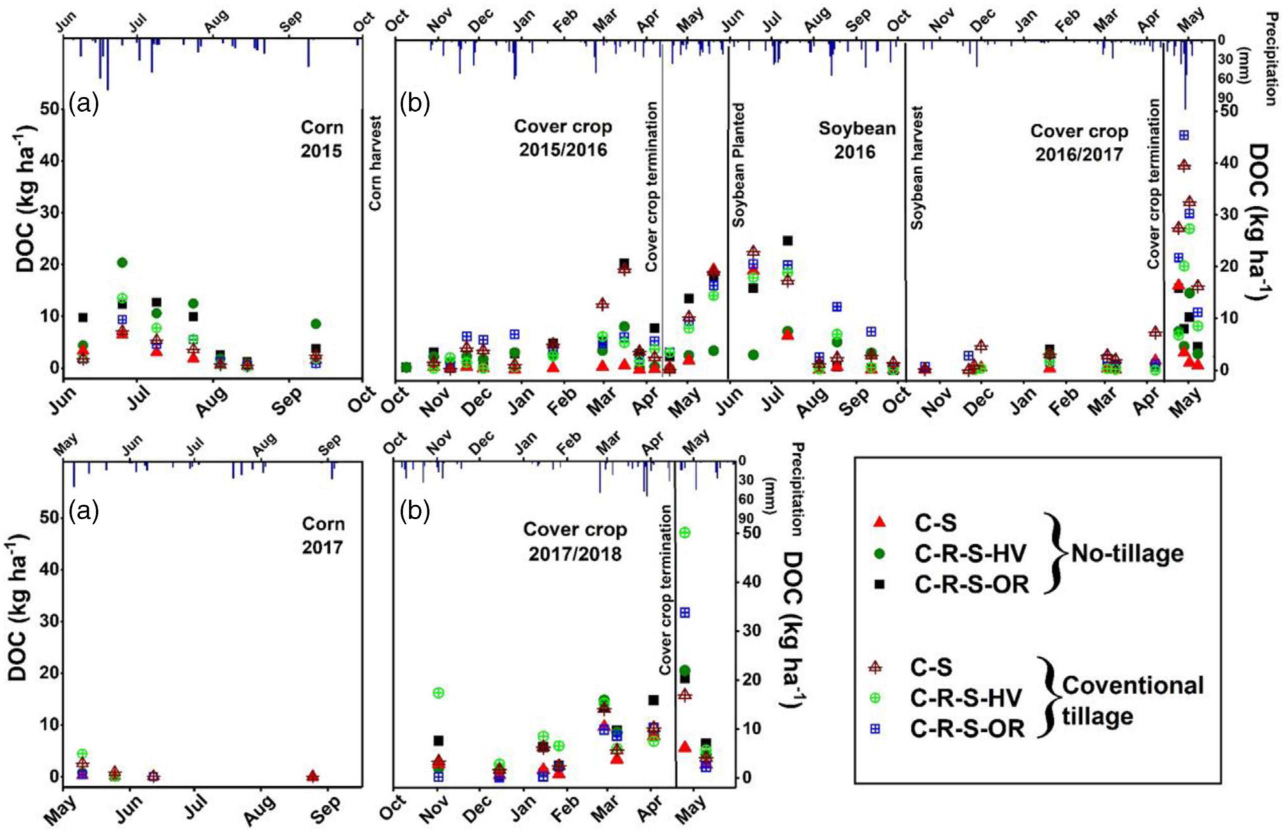

FIGURE 2 Mean dissolved organic C (DOC) leaching across sampling dates for cover crop and no-cover crop rotations under no-tillage and conventional tillage system. Blue bars represent the average daily precipitation received at the research site. Note the scale on *y* axis changes on the figure from left to right and on the bottom. Rotations were corn–soybean (C–S), corn–cereal rye–soybean–hairy vetch (C–R–S–HV), and corn–cereal rye–soybean–oat + radish (C–R–S–OR)

during the CC growing season, CT resulted in greater mean DOC leaching by 8.3 and 6.6 kg ha^{-1} than the NT under the C–S rotation having no CCs and C–R–S–OR rotation having oat + radish CC, respectively. Within the CT system in spring 2017, hairy vetch in C–R–S–HV rotation reduced mean DOC leaching compared with the C–R–S–OR rotation having oat + radish CCs and C–S rotation having no CCs by 6.6 and 7.8 kg ha^{-1}, respectively. The mean DOC leaching in spring 2018 was 5.1 kg ha^{-1} greater in C–R–S–HV rotation having cereal rye CC compared with C–S rotation having no CC. Mean DOC leaching across sampling dates for CC and no-CC rotations under NT and CT is presented in Figure 2.

Crop rotation resulted in differences in cumulative DOC leaching during the cash crop growing season only in fall 2015, whereas no differences were obtained among rotations during the soybean and corn growing season in 2016 and 2017 (Table 5). In fall 2015, the C–R–S–HV rotation having hairy vetch CC planted in previous spring resulted in 25.4 kg ha^{-1} greater cumulative DOC leaching loads than C–S rotation that had no CC planted (Table 5). Cumulative DOC leaching during the CC growing season was affected by the interaction of crop rotation and tillage in spring 2017 and 2018, but not in spring 2016. Conventional tillage increased cumulative DOC leaching compared with NT by 19.1 and 20.2 kg ha^{-1} in spring and fall of 2016, respectively. Conventional tillage increased cumulative DOC leaching compared with NT by 108.4 kg ha^{-1} only in rotation without any CCs (C–S) during spring 2017.

3.2 | Nitrogen pools

3.2.1 | Soil

Similar to C pools, the WEN and TN decreased with soil depth in all crop rotations and tillage systems (Table 6). The WEN was significantly affected by the interaction of crop rotation and depth during the corn season in fall 2015 and 2017 (Table 6, Supplemental Table S2). In fall 2015 during the corn season, the CT system increased WEN by 2 kg ha^{-1} compared with the NT system, but it reduced TN content by 0.07 Mg ha^{-1} compared with NT. Differences in WEN between crop rotations were only seen at the depth of 0–15 cm in fall 2015, where C–R–S–HV rotation with hairy vetch CC had 7 kg ha^{-1} greater WEN than the rotation without CC (C–S). During the CC growing season in spring 2016, the

TABLE 6 Mean water-extractable N (WEN) and total N (TN) in soil determined by crop rotation, tillage, and depth. Within a column and within a given factor or combination of factors, means followed by the same letter are not statistically different (α = .10)

Crop rotation[a]	Tillage[b]	Depth	Corn, fall 2015		Cover crop, spring 2016		Soybean, fall 2016		Cover crop, spring 2017		Corn, fall 2017		Cover crop, spring 2018	
			WEN	TN	WEN	TN	WEN	TN	WEN	TN	WEN	TN	WEN	TN
		cm	kg ha^{-1}	Mg ha^{-1}	kg ha^{-1}	Mg ha^{-1}	kg ha^{-1}	Mg ha^{-1}	kg ha^{-1}	Mg ha^{-1}	kg ha^{-1}	Mg ha^{-1}	kg ha^{-1}	Mg ha^{-1}
C–S			17b	1.63	14a	1.55	19	1.71	17ab	1.52	18b	1.62	20a	1.45
C–R–S–HV			19a	1.52	13b	1.56	19	1.58	17a	1.55	26a	1.66	16b	1.51
C–R–S–OR			18ab	1.51	12b	1.50	17	1.58	14b	1.50	19b	1.54	15b	1.49
	CT		19a	1.52b	13	1.53	19	1.67	17a	1.61a	24	1.66	19a	1.53
	NT		17b	1.59a	13	1.54	18	1.58	14b	1.43b	21	1.56	16b	1.44
		0–15	35a	2.42a	24a	2.42a	36a	2.5a	27a	2.41a	35a	2.45a	29a	2.34a
		15–30	11b	1.28b	8b	1.25b	11b	1.33b	10b	1.2b	16b	1.33b	11b	1.21b
		30–45	8c	0.96c	7b	0.94c	7c	1.05c	11b	0.96c	12c	1.05c	11b	0.9c
C–S	CT		18	1.60	15	1.55	21	1.74	21a	1.62	20	1.73	23	1.53
C–S	NT		15	1.66	13	1.56	17	1.67	12b	1.42	17	1.51	18	1.37
C–R–S–HV	CT		19	1.49	13	1.55	18	1.70	18ab	1.67	25	1.70	16	1.54
C–R–S–HV	NT		19	1.53	13	1.57	20	1.47	17ab	1.42	26	1.62	15	1.49
C–R–S–OR	CT		19	1.46	12	1.50	17	1.57	14b	1.54	18	1.53	17	1.52
C–R–S–OR	NT		16	1.59	12	1.49	16	1.59	14b	1.46	19	1.54	14	1.46
C–S		0–15	31b	2.48	25	2.46	39	2.64	26	2.45	28b	2.50	34	2.36
C–S		15–30	11c	1.05	9	1.24	11	1.36	10	1.19	15cd	1.31	12	1.12
C–S		30–45	8c	1.36	8	0.96	7	1.12	14	0.92	12cd	1.06	15	0.87
C–R–S–HV		0–15	38a	2.36	24	2.48	38	2.47	30	2.41	44a	2.52	27	2.40
C–R–S–HV		15–30	11c	0.93	7	1.22	11	1.30	12	1.18	19c	1.36	10	1.21
C–R–S–HV		30–45	8c	1.24	6	0.98	7	0.98	10	1.04	13cd	1.10	10	0.93
C–R–S–OR		0–15	36ab	2.41	22	2.30	33	2.38	23	2.37	32b	2.33	26	2.26
C–R–S–OR		15–30	10c	0.91	7	1.31	10	1.32	9	1.23	14cd	1.31	11	1.29
C–R–S–OR		30–45	7c	1.25	7	0.87	7	1.03	9	0.90	11d	0.98	9	0.91

[a]Crop rotation: C–S, corn–soybean; C–R–S–HV, corn–cereal rye–soybean–hairy vetch; C–R–S–OR, corn–cereal rye–soybean–oat + radish.

[b]Tillage: CT, conventional tillage; NT, no-tillage.

TABLE 7 Depth-wise linear regression between soil N (water-extractable N [WEN] and total N [TN]) pools and collection time of soil samples

Crop rotation[a]	Tillage[b]	Depth	WEN Variables Intercept	WEN Variables Slope	WEN Pr > *F*	WEN R^2	TN Variables Intercept	TN Variables Slope	TN Pr > *F*	TN R^2
		cm	——kg ha^{-1}——				——Mg ha^{-1}——			
C–R–S–OR	CT	0–15	34.33	–0.95	.3504	.05	2.45	–0.02	.6367	.01
		15–30	8.19	0.56	.0704	.19	1.18	0.03	.4175	.04
		30–45	5.51	0.06	.0129	.33	0.82	0.02	.2691	.08
	NT	0–15	29.73	–0.96	.2544	.08	2.37	–0.02	.5967	.02
		15–30	8.19	0.67	.1224	.14	1.37	–0.02	.4692	.03
		30–45	6.00	0.79	.0038	.42	1.01	–0.01	.7963	.00
C–R–S–HV	CT	0–15	33.00	0.20	.8750	.00	2.44	0.03	.4365	.04
		15–30	8.99	0.74	.2990	.07	1.26	0.02	.6281	.02
		30–45	6.13	0.83	.0227	.28	0.91	0.01	.5647	.02
	NT	0–15	33.97	–0.19	.8952	.00	2.37	–0.01	.6854	.01
		15–30	8.28	1.06	.0814	.17	1.20	–0.01	.6241	.02
		30–45	6.18	0.86	.0241	.28	1.00	0.01	.7665	.01
C–S	CT	0–15	30.34	0.99	.4983	.03	2.54	0.03	.7999	.00
		15–30	11.02	0.44	.3777	.05	1.30	–0.01	.9071	.00
		30–45	6.17	1.81	.0392	.24	1.02	–0.01	.5228	.03
	NT	0–15	29.08	–0.53	.5629	.02	2.56	–0.06	.0505	.22
		15–30	7.49	0.85	.0155	.31	1.46	–0.06	.0205	.29
		30–45	4.47	1.18	.0007	.52	1.13	–0.03	.2213	.09

[a]Crop rotation: C–S, corn–soybean; C–R–S–HV, corn–cereal rye–soybean–hairy vetch; C–R–S–OR, corn–cereal rye–soybean–oat + radish.
[b]Tillage: CT, conventional tillage; NT, no-tillage.

inclusion of cereal CC in C–R–S–HV and C–R–S–OR reduced WEN in soil by 1–2 kg ha^{-1} as compared with C–S rotation having no CC.

During the soybean growing season in fall 2016, no differences were obtained due to rotation and tillage systems for WEN and TN. In the spring of 2017 during the CC season, the CT resulted in 9 kg ha^{-1} greater WEN than NT system only in C–S rotation having no CC (Table 6). Under the CT system in spring 2017, the oat + radish CC in C–R–S–OR rotation reduced soil WEN content by 7 kg ha^{-1} compared with C–S rotation with no CC planted. However, the TN was 0.18 Mg ha^{-1} greater under CT than under NT in spring 2017, when data were averaged over rotations and depth. In fall 2017, the C–R–S–HV rotation that had hairy vetch CC in spring season resulted in 16 and 12 kg ha^{-1} greater WEN than the C–S and C–R–S–OR rotations, respectively, at a depth of 0–15 cm. During the CC season in spring 2018, CT had 3 kg ha^{-1} greater WEN in soil than NT, and cereal rye CC in the C–R–S–HV and C–R–S–OR rotations reduced WEN content in soil by 4–5 kg ha^{-1} compared with C–S rotation having no CC.

The WEN increased significantly ($P < .1$) over time from fall 2015 to spring 2018 in all crop rotations under both tillage systems at a depth of 30–45 cm, as well as in C–R–S–HV having cereal rye and hairy vetch CCs and C–S having no CCs under NT at a depth of 15–30 cm (Table 7). However, TN decreased over time from the fall of 2015 to spring 2018 under the NT system in the C–S rotation having no CC at depths of 0–15 and 15–30 cm.

3.2.2 | Water

The mean TN leaching during the corn growing season was affected by main effects of crop rotations and tillage in 2015 and by their interaction in 2017, whereas differences in mean TN leaching were only due to tillage systems during the soybean season in fall 2016. Inclusion of CCs in the C–R–S–HV and C–R–S–OR rotations resulted in higher mean TN leaching compared with C–S rotation without any CC during the corn growing season in fall 2015 (Table 8). Conventional tillage increased mean TN leaching than NT by 4.3 and 1.6 kg ha^{-1} in fall 2015 and fall 2016, respectively. In spring 2016, mean TN leaching was significantly affected by the interaction of tillage and crop rotation. Conventional tillage system increased mean TN losses compared with NT only in the C–S rotation, which had no CC planted, during spring 2016. Under the CT system, cereal rye CC in the C–R–S–HV and

TABLE 8 Mean and cumulative total N collected using zero-tension lysimeters installed below the A horizon. Within a column and within a given factor or combination of factors, means followed by the same letter are not statistically different (α = .1)

		Total N											
		Corn, fall 2015		Cover crop, spring 2016		Soybean, fall 2016		Cover crop, spring 2017		Corn, fall 2017		Cover crop, spring 2018	
Crop rotation[a]	**Tillage**[b]	**Mean**	**Cumulative**	**Mean**	**Cumulative**	**Mean**	**Cumulative**	**Mean**	**Cumulative**	**Mean**	**Cumulative**	**Mean**	**Cumulative**
		kg ha^{-1}											
C–S		20.9b	91.5b	0.5a	8.1a	2.6	12.1	0.6b	9.6b	3.2b	7.4b	3.0a	27.3a
C–R–S–HV		38.1a	210.4a	0.1b	3.2b	2	9.3	1.2b	14.4b	13.4a	18.4a	2.3b	20.8b
C–R–S–OR		33.8a	150.7ab	0.3b	5.1b	2.6	11.5	3.1a	29.0a	2.1b	3.5b	1.5b	13.9b
	CT	33.1a	171.5a	0.4a	6.5a	3.2a	14.6a	1.5b	17.5	10.3	15.7	2.7	25.8
	NT	28.8b	130.2b	0.2b	4.3b	1.6b	7.3b	1.8a	17.8	2.2	3.8	1.7	15.5
C–S	CT	26.8	128.7	0.9a	12.1a	3.6	16.9	1bc	15.8	5.4b	12.5b	3.9a	36
C–S	NT	14.9	54.3	0.2b	4.1b	1.6	7.2	0.1c	3.4	1b	2.4b	2.0ab	18.6
C–R–S–HV	CT	34.4	186.9	0.1b	3.3b	2.7	12.8	1.5abc	17	24a	32.0a	3.8ab	34.1
C–R–S–HV	NT	41.9	233.8	0.1b	3.1b	1.2	5.8	1bc	11.9	2.8b	4.7b	0.8b	7.4
C–R–S–OR	CT	38	198.9	0.2b	4.2b	3.2	14	2ab	19.8	1.6b	2.6b	0.5b	7.4
C–R–S–OR	NT	29.6	102.5	0.3b	5.8b	2	9	4.2a	38.1	2.6b	4.4b	2.4ab	20.4
Source of variation	df	*p* values											
Rotation	2	<.0001	.0095	<.0001	.0481	.934	.7464	.0001	.0287	.0436	.0153	.0125	.0966
Tillage	1	.0248	.0311	.0021	.0318	.0004	.0365	.0327	.1391	.6271	.7805	.3173	.422
Rotation × tillage	2	.1093	.5834	<.0001	.0155	.2013	.7767	.0176	.4158	.0225	.0313	.0173	.1285

[a]Crop rotation: C–S, corn–soybean; C–R–S–HV, corn–cereal rye–soybean–hairy vetch; C–R–S–OR, corn–cereal rye–soybean–oat + radish.

[b]Tillage: CT, conventional tillage; NT, no-tillage.

C–R–S–OR rotations reduced mean TN leaching losses compared with the C–S rotation having no CC by 0.7–0.8 and 7.9–8.8 kg ha^{-1}, respectively, in spring 2016. In spring 2016, no differences were observed for TN due to tillage for two rotations having CCs (Table 8).

The mean TN leaching during the CC season was influenced by the interaction of crop rotation and tillage systems in spring 2016, 2017, and 2018 (Table 8). In spring 2017, the oat + radish CC in C–R–S–OR rotation resulted in 3.2 and 4.1 kg ha^{-1} greater mean TN leaching loss than C–R–S–HV rotation having hairy vetch CC and C–S rotation having no CC, respectively, within the NT system (Table 8). Mean TN was 21.2 kg ha^{-1} greater under CT than under NT in fall 2017, only in the C–R–S–HV rotation. Under the CT system in fall 2017, C–R–S–HV rotation that had hairy vetch CC plated during the previous spring had 18.6 and 22.4 kg ha^{-1} greater mean TN losses than C–S and C–R–S–OR rotations, respectively. In spring 2018 under the CT system, the mean TN leaching loss was 3.4 kg ha^{-1} greater from C–S than from C–R–S–OR rotation that had cereal rye CC.

Planting of hairy vetch CC in the preceding spring in the C–R–S–HV rotation increased cumulative TN leaching loads compared with the rotation without any CC (C–S) by 118.9 kg ha^{-1} during fall 2015 (Table 8). Cereal rye CC in the C–R–S–HV and C–R–S–OR rotations reduced cumulative TN leaching by 7.9–8.8 kg ha^{-1} compared with C–S rotation that had no CC planted during the spring 2016 under the CT system only. Averaged over rotations, the CT system increased cumulative TN leaching compared with the NT by 7.3 kg ha^{-1} in fall 2016. When data were averaged over tillage systems in spring 2017, oat + radish CC in the C–R–S–OR rotation led to 14.6 and 19.4 kg ha^{-1} greater cumulative TN leaching compared with C–R–S–HV rotation having hairy vetch CC and C–S rotation having no CC, respectively. Conventional tillage resulted in 27.3 kg ha^{-1} greater cumulative TN leaching than the NT system in fall 2017 only in the C–R–S–HV rotation that had hairy vetch CC planted in the preceding spring. Within the CT system in fall 2017, the C–R–S–HV rotation had 19.5 and 29.4 kg ha^{-1} greater cumulative TN than C–S and C–R–S–OR rotations, respectively. Cereal rye CC in the C–R–S–HV and C–R–S–OR rotations reduced the cumulative TN by 6.5 and 13.4 kg ha^{-1}, respectively, as compared with the rotation without any CC (C–S) in spring 2018.

4 | DISCUSSION

4.1 | Soil C and N pools

Carbon pools decrease with soil depth from the surface to 45 cm in our study. The upper soil layers receive more leaf litter and root biomass than the deeper soil layers. Greater biomass return to upper soil layers results in more organic matter additions to soil and, consequently, higher C and N pools in these topsoil layers. Similar to our study, Grebliunas et al. (2016) reported greater water-extractable organic C in the upper soil depth (0–5 cm) than in the lower soil profile (5–20 cm). Cover crops such as tillage radish and cereal rye usually have lower C.N ratio than the corn (>60:1) and soybean crops (30:1), which promotes faster decomposition and higher C and N content in upper soil layers (Olson et al., 2010; Peregrina et al., 2010; Steenwerth & Belina, 2008).

In our study, CT had lower WEC than NT for rotation C–R–S–OR in fall 2017, when data were averaged over crop rotations and depth. Similarly, Linn and Doran (1984) reported lower WEC under CT compared with NT in the topsoil depth from 0–7.5 cm but found no differences between tillage systems in deeper soil depths. Greater C and N concentrations in soil under NT in surface results from the reduced decomposition of residues due to less contact of residues with the soil particles and microbes (Blanco-Canqui et al., 2013; Varvel & Wilhelm, 2010). However, greater POXC content was present under CT than NT in the spring 2017 and in C–S rotation during fall 2017. Gregorich, Liang, Drury, Mackenzie, and McGill (2000) found that the incorporation of crop residues by tillage increases water-extractable organic C via stimulating the microbial decomposition of residues. Higher POXC with NT compared to CT at 0-to-2.5-cm soil depth was also reported by Jagadamma, Essington, Xu, and Yin (2019).

Differences in soil C pools were observed due to rotations in fall 2015 and 2017. Inclusion of hairy vetch and oat + radish CCs increased WEC in fall 2015 and POXC in fall 2017 under the NT system compared with no CC control containing winter weeds. Similarly, Mukumbareza, Muchaonyerwa, and Chiduza (2016) reported that the inclusion of hairy vetch CC resulted in higher water-soluble C by 0.24 and 0.23 mg g^{-1} soil at depths of 0–5 and 5–20 cm, respectively, when compared with weedy fallow. Greater C and N concentrations in CC treatments such as rye, hairy vetch, crimson clover (*Trifolium incarnatum* L.), when compared with no CC control, were also reported by Sainju et al. (2002). However, the POXC was lower in CC rotations compared with no CC rotation under the CT system. For the C–S rotation, the NT had lower POXC than CT in fall 2017. Plant species in crop rotation affect the amount and type of C inputs, as well as the released material via root exudates and consequently, affect the labile and bulk fractions of C in soil (Chantigny, 2003). An increase in C and N pools in soil with CCs compared with no CC control can be attributed to greater C and N inputs from CC biomass than from the winter weeds. Few studies have shown that including legumes in a crop rotation increases the amount of WEC in soil (Campbell et al., 1999; Mazzarino et al., 1993). However, a study conducted by Sainju et al. (2002) found that rye resulted in greater organic C and N concentrations than hairy vetch due to its greater biomass yield, C concentration, and C/N ratio. Sainju et al. (2002) found that soil organic C and N concentrations can be conserved by using NT along with CC, with nonlegumes being better than legumes for increasing the SOC and N content in soil (Sainju et al., 2002). Legume CCs can undergo faster decomposition after termination due to their lower C/N ratio and release N quickly during the cash crop growing season, resulting in higher N leaching. No differences were obtained for C pools between CC rotations containing hairy vetch or oats + radish in our study.

The TN content in soil is controlled by the climate and vegetation type, and it affects the decomposition of organic matter and humification rates (Guimarães et al., 2013). Lower TN content in soil under NT in spring 2017 might have resulted from higher N accumulation in aboveground biomass in NT (51 kg ha^{-1}) than the CT (39 kg ha^{-1}) as found in our previous work (Singh, Thilakarathne, et al., 2020; Supplemental Table S3). Lower soil WEN in corn–soybean rotation with CCs (C–R–S–HV and C–R–S–OR) than in C–S without any CC during spring 2016 and 2018 might be due to higher C/N ratio and greater N uptake by cereal rye in the C–R–S–HV and C–R–S-OR compared with weeds in C–S (Singh, Thilakarathne, et al., 2020; Supplemental Table S4). In our previous study, N accumulation in aboveground biomass (cereal rye + weeds) in C–R–S–HV and C–R–S–OR was 19 and 16 kg ha^{-1} higher than the C–S in 2016 (Singh, Thilakarathne, et al., 2020; Supplemental Table S4). In fall 2017, C–R–S–HV had greater WEN than the C–S and C–R–S–OR at depth of 0–15 cm (Supplemental Table S3). Hairy vetch (10:1) grown in C–R–S–HV during spring 2017 had a lower C/N ratio than the oat + radish (20:1) in C–R–S–OR and weeds (22:1) in C–S and consequently resulted in faster decomposition of hairy vetch after termination and N release during the corn growing season (Singh, Thilakarathne, et al., 2020; Supplemental Table S3).

The WEN and TN decreased with depth in our study. Similarly, a decline in TN with an increase in soil depth has been reported by multiple studies (Guimarães et al., 2013; Singh et al., 2018). A decrease in N pools with depth might be due to a reduction in root biomass and density in subsoil layers (Singh et al., 2018). It is possible that greater presence of labile N in surface soil might lead to N losses via runoff and erosion, ultimately contributing to poor water quality in the downslope water streams.

4.2 | DOC and TN leaching

Inclusion of cereal rye CC in the C–R–S–HV and C–R–S–OR rotations resulted in lower cumulative TN leaching losses compared with C–S under the CT system in spring 2016 and 2018. Previous studies have reported that CCs such as cereal rye reduce nitrate leaching after harvest of cash crops by scavenging residual N from the soil, and consequently reduce TN leaching in groundwater (Brandi-Dohrn et al., 1997; Dozier et al., 2017; Meisinger & Ricigliano, 2017; Meisinger et al., 1991; Staver & Brinsfield, 1998). Cereal rye grows faster and has greater root density, which results in reducing nitrate leaching to groundwater and surface water (McCracken et al., 1994). However, not all CCs are equally efficient in N scavenging, since the N scavenging depends on many factors including precipitation received (Ball-Coelho & Roy, 1997; Meisinger & Ricigliano, 2017), CC species (legume vs. nonlegume), CC biomass production, planting and termination timing of CCs (Mirsky, Ackroyd, et al., 2017; Mirsky, Spargo, et al., 2017), and amount of residual N in the soil after harvest of cash crop (Kuo & Jellum, 2002).

Greater cumulative TN leaching losses in the C–R–S–OR compared with the other two rotations in spring 2017 might have resulted from lower biomass production and N accumulation in oats (Singh, Thilakarathne, et al., 2020; Supplemental Table S3). The N accumulation in CC + weeds aboveground biomass during spring 2017 in C–R–S–OR was 34.99 kg ha^{-1} as compared with 73.33 and 26.66 kg ha^{-1} in C–R–S–HV and C–S rotations, respectively, in our study (Singh,Thilakarathne, et al., 2020; Supplemental Table S3). Oat are winter killed, which might have caused faster decomposition of biomass, thus releasing N in the soil. However, the use of hairy vetch as a CC in C–R–S–HV rotation did not result in lower cumulative TN loads in soil water as compared with C–S rotation without any CC in spring 2017. Hairy vetch, being a legume, fulfills its N requirement by biological N fixation when compared with nonlegumes CCs, which uptake N from the soil. Previous studies have reported that legumes are not as efficient as nonlegumes in reducing nitrate leaching (McCracken et al., 1994; Tonitto, David, & Drinkwater, 2006).

The variation in mean DOC leaching losses due to CC and tillage were observed during cash crop and cover seasons from 2015 to 2018 except in fall 2017. The cumulative DOC leaching was greater in the CT than in NT in spring 2016, fall 2016, and in spring 2017, only in rotation C–S. Crop residues contribute to DOC loss, as they are potential sources of C and nutrients (Bertol, Engel, Mafra, Bertol, & Ritter, 2007; Lal, Logan, Eckert, Dick, & Shipitalo, 2017). Conventional tillage mixed up crop residues in the tilled soil layers, whereas the residues are left on the soil surface in NT. Mixing of soil residues in the soil makes them more susceptible to decomposition due to more contact with soil particles. The decomposition of residues will produce more labile C fractions in soil and, consequently, result in more DOC loss under the CT system. Cumulative DOC leaching was greater under C–R–S–HV than the other two rotations in spring 2018, whereas DOC leaching during the cash crop season was greater under C–R–S–HV than under C–S in 2015. Greater DOC leaching under CCs compared with no CC and tilled plots was also reported by Steenwerth and Belina (2008). Hairy vetch and oat

+ radish have different growing patterns, resulting in differences in aboveground and belowground biomass production. In our previous study, the aboveground biomass produced by hairy vetch CC + weeds in the C–R–S–HV rotation (900 kg ha^{-1}) was 670 kg ha^{-1} greater than the biomass produced in the C–R–S–OR rotation (230 kg ha^{-1}) by oat + radish CC and weeds in 2015 (Singh, Thilakarathne, et al., 2020; Supplemental Table S3). Greater aboveground and belowground biomass production probably resulted in greater labile C content and, consequently, greater DOC losses under the hairy vetch system in 2015. Seasonal differences in DOC losses might be attributed to changes in soil water content, which is dependent on the amount of precipitation received (Steenwerth & Belina, 2008). In summary, our study shows that the DOC leaching by CCs can depend upon their biomass production and CC species used in rotation. Cover crops and NT might increase labile C pools in soil over time, but the labile pools of C and N vary among these systems depending upon the growing season conditions whether it is CC season or cash crop season.

5 | CONCLUSIONS

Our study evaluated the impact of CCs under different tillage systems on water-extractable forms of soil C and N in the corn–soybean rotation system, as well as DOC and TN leaching. Seasonal differences were observed due to CCs and tillage for WEC and WEN. Cover crops and tillage did not change TC over the 4 yr of this study from 2015 to 2018, except in rotation C–R–S–OR rotation under CT at subsurface depth. Tillage affects the decomposition of organic matter in soil and consequently, changes the concentrations of labile C fractions in soil, depending on the crop rotation. Hairy vetch and cereal rye CCs reduced N leaching during the CC season—however, cereal rye increased cumulative DOC leaching in the 2018 CC season. The mean and cumulative DOC concentrations over different crop seasons do not always follow the same trend. Cereal rye (spring 2016 and 2018) and hairy vetch or oat + radish CCs (2015, 2017) came twice in 4 yr of our study. A more long-term study might be needed to get a better understanding of the DOC and TN leaching trends under CC systems in a corn–soybean rotation.

ACKNOWLEDGMENTS

This publication is a contribution of the National Center for Alluvial Aquifer Research and the Mississippi Agricultural and Forestry Experiment Station. This material is based upon work supported by the Illinois Nutrient Research and Education Council (www.illinoisnrec.org) and the USDA-ARS (Non-Assistance Cooperative Agreement no. 58-6001-7-001).

AUTHOR CONTRIBUTIONS

Gurbir Singh: Conceptualization; Formal analysis; Funding acquisition; Investigation; Methodology; Project administration; Supervision; Writing-original draft; Writing-review & editing. Gurpreet Kaur: Formal analysis; Writing-original draft; Writing-review & editing. Karl W. J. Williard: Funding acquisition; Project administration; Resources; Supervision. Jon E. Schoonover: Funding acquisition; Resources; Supervision.

CONFLICT OF INTEREST

Authors declare no conflict of interest.

ORCID

Gurbir Singh https://orcid.org/0000-0002-4013-3460
Gurpreet Kaur https://orcid.org/0000-0003-0766-3336

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SUPPORTING INFORMATION

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How to cite this article: Singh G, Kaur G, Williard KW, Schoonover JE. Cover crops and tillage effects on carbon–nitrogen pools: A lysimeter study. *Vadose Zone J*. 2021;e20110.
https://doi.org/10.1002/vzj2.20110