

MANAGEMENT LEGACY EFFECTS ON COVER CROP PRODUCTIVITY AND
POTENTIAL TO REDUCE NUTRIENT LEACHING FROM FARMS

by

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A thesis submitted

in partial fulfillment of the requirements

for the degree of

Master of Science

School for Environment and Sustainability

In the University of Michigan

April 2021

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Abstract

Cover cropping is an ecological management practice that provides a variety of ecosystem functions to farms, such as increasing soil organic matter content and improving nutrient retention. Background soil fertility due to unique management legacies is expected to influence the productivity of cover crops and subsequently mediate their effects on ecosystem functions related to nutrient retention. We used a long-term experiment at Michigan State University's Kellogg Biological Station to test the legacy effects of four distinct management systems (ranging from conventional to certified organic) on the production and function of crimson clover and cereal rye cover crops grown alone and in mixture, with a focus on the potential to reduce nitrate leaching. Cover crops were planted following winter wheat harvest in the summer of 2019 and corn was planted in all treatments the following spring after cover crop termination. We applied bromide as a conservative tracer of water and anion flow in the soil profile to assess management legacy and cover crop treatment effects on potential anion leaching.

We found that via changes to soil fertility, management legacies influenced cover crop productivity and species composition in mixture: in the legacies with lower soil fertility, crimson clover was more competitive than rye and dominated the mixture. Legacies with higher soil fertility—which were those with a history of ecological nutrient management—had higher average cover crop biomass (mean = 3357 kg ha⁻¹) and a higher percent recovery of bromide (mean = 33.28%), an estimate of soil anion retention. Further, percent recovery of bromide was positively correlated with cover crop biomass ($r^2=.17$) and the free particulate organic matter fraction ($r^2=.29$), indicating that the effect of management legacy on cover crop function is mediated by cover crop production and background soil fertility. This experiment reveals

complex interactions between soil fertility, cover crop growth, and nutrient leaching potential that depend on soil conditions resulting from specific management regimes.

Acknowledgements

The completion of my thesis could not have been possible without the help of many incredible people. First, I would like to thank Dr. Jennifer Blesh for her invaluable guidance as my thesis advisor and her long-time mentorship, without which I likely would not have come to graduate school at all. I would like to thank Dr. Donald Zak for being on my committee and offering his expertise on this project, and for all he has taught me. For his expertise and advice on all things bromide, I would like to thank Dr. Steve Hamilton.

This project took many early drives, hot field days, and long sample processing hours. I cannot thank Beth VanDusen, Etienne Harrick, Dev Gordin, and the rest of the Blesh lab enough for their help. Thank you to Kevin Kahmark, the Robertson lab, and everyone at KBS for hosting us, helping us collect samples, and all the work you do behind the scenes. I am so grateful for Kent Connell, who graciously and patiently offered endless support throughout my analysis. To Alison Bressler, thank you for all the advice, moral support, and friendship you have given me along the way.

Lastly, I would like to thank the most important people in my life, without whom I would have not been able to complete this thesis. To my family, thank you for always encouraging me. To my friends, I am so lucky to lean on you. And to my sister Katherine, my biggest cheerleader and best friend, thank you for everything.

This project was partnered with the Kellogg Biological Station Long Term Ecological Research site. Funding was provided by the USDA AFRI Foundational Program.

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Introduction

The global intensification of agriculture has resulted in increasing dependence on chemical fertilizers to ensure crop yields and a corresponding decrease in soil fertility (Matson et al., 1997). This dependence, called the “fertilizer treadmill”, promotes chronic application of synthetic fertilizers, particularly for nitrogen (N), which is the nutrient that most often limits crop productivity (Drinkwater & Snapp, 2007). Inorganic N fertilizers are often applied in excess on conventional farms, leading to large N surpluses and associated nitrate (NO_3^-) leaching losses from fields (Blesh & Drinkwater, 2013; Syswerda et al., 2012). Nitrate leaching threatens public health and it causes contamination and eutrophication of major bodies of water, leading to low-oxygen “dead zones” which impair aquatic ecosystem functioning (Diaz & Rosenberg, 2008; Galaviz-Villa et al., 2010).

In response, ecological nutrient management practices focus on increasing crop functional diversity to support ecosystem functions and reduce the use of external fertilizer inputs, which can substantially reduce NO_3^- leaching losses from fields (Robertson et al., 2014; Syswerda et al., 2012; Syswerda & Robertson, 2014). Cover crops, which are non-harvested crops planted in rotation with main crops, are an example of such management, and can provide a variety of ecosystem services to farms (Snapp et al., 2005). Because cover crops are planted in between the harvest and planting of cash crops, they provide plant cover over what would otherwise be bare land. This extended time of living plant cover allows for increased assimilation and immobilization of potentially leachable N through multiple mechanisms (Thorup-Kristensen & Nielsen, 1998; Tonitto et al., 2006).

Cover crops and soil fertility

The adoption of cover crops can recouple carbon and nutrient cycles to reduce the leakiness of agroecosystems (Drinkwater & Snapp, 2007). Organic carbon and N in cover crop residues are decomposed following their incorporation into soil, contributing to soil organic matter (SOM), which is vital to nutrient cycling and availability, as well as nutrient retention. The biochemical composition and concentration of SOM impacts N retention and mineralization; in intensive agricultural systems, SOM is depleted, which increases N leaching potential due to reduced capacity to immobilize and retain N. In these systems, nitrification occurs at a greater rate than mineralization, which increases the soil NO_3^- pool and potential for N losses (Booth et al., 2005). Winter cover crops, however, can increase net carbon inputs to soil, which can increase SOM content (King & Blesh, 2018; Puget & Drinkwater, 2001). They can also support increased microbial activity (e.g. via root exudation and litter inputs to soil) and microbial byproducts have been shown to comprise a large part of the stable SOM pool (Cotrufo et al., 2013). The continuous plant growth offered by cover crops also encourages beneficial interactions between plants and soil microbes that can better couple N mineralization with plant N assimilation, ultimately reducing N surplus and potential loss (Drinkwater & Snapp, 2007; Syswerda & Robertson, 2014; Thorup-Kristensen & Dresbøll, 2010).

Cover crop functional diversity and nitrate leaching

Two common functional groups of cover crops are grasses and legumes, which have different N acquisition strategies. Grasses have high capacity for N retention: they have fibrous and extensive root systems that assimilate soil N and later return it to the soil when they decompose and die. This organic recycling of N through organic matter inputs can reduce excess inorganic N

in soil and thus, N losses. Indeed, nonlegume cover crops significantly reduce NO_3^- leaching (Finney et al., 2016; Kaspar et al., 2012; Tonitto et al., 2006).

Although they have a lower capacity for soil N assimilation and retention compared to grasses, legumes can similarly decrease N surpluses, thereby reducing leaching, especially when they are grown as a N source to reduce the use of synthetic N fertilizers (Blesh & Drinkwater, 2013; Tonitto et al., 2006). Biological N_2 fixation (BNF) by legumes is energy-intensive; the plant must provide energy in the form of carbohydrates to symbiotic bacteria that fix N_2 in return. If soil N availability is high, there is a decreased need for fixed N and legumes can down-regulate BNF to save energy. Thus, legumes can respond to environmental conditions, like soil N availability, and adjust N acquisition correspondingly, which limits excess N in agroecosystems (Blesh, 2019; Schipanski et al., 2010).

Given their complementary functional, mixtures of legumes and grasses can potentially support both N supply and N retention services, including reducing NO_3^- leaching (Finney & Kaye, 2017; Gabriel et al., 2012; Hayden et al., 2014; Kaye et al., 2019). Managing cover crop stands that have greater functional trait diversity may therefore optimize these complementary ecosystem functions. In particular, grass-legume mixtures may reduce potentially leachable N to levels comparable to a sole grass, while supplying an organic N source to soil. Functionally diverse mixtures can also reduce leaching potential more than sole-planted legumes, although there may be tradeoffs between N retention and N supply in mixtures. A previous on-farm study testing 3- and 4-species mixtures found that increasing non-legume presence in mixture decreased N supply services to the subsequent crop but allowed greater N retention and reductions in NO_3^- leaching (White et al., 2017). While previous studies have examined such tradeoffs among multiple ecosystem functions and services in cover crop bicultures (Bergkvist et

al., 2011; Ranells & Wagger, 1997; Tosti et al., 2014), their effects on NO_3^- leaching across variable soil conditions that result from distinct, long-term management systems histories is not known.

Interactions between farm management history and cover crop function

Farm management legacies influence NO_3^- leaching through their effects on multiple SOM pools and associated microbial processes (Drinkwater & Snapp, 2007). The primary dimensions of management that influence overall soil fertility and health are the intensity of tillage (e.g. soil disturbance), crop diversity, and levels of external input use (Zimnicki et al., 2020). For example, simplified crop rotations with large inputs of synthetic fertilizer and intensive tillage practices tend to have low SOM stocks, low rates of mineralization, and decreased microbial activity. These soil conditions, in turn, influence the establishment and productivity of plants, particularly for cover crops which do not typically receive chemical inputs. Nonlegume cover crops, for example, typically thrive in high fertility soils whereas legumes perform better than nonlegumes in nutrient-poor soils because of their ability to fix N_2 . In mixtures of both functional types, interspecific competition—which depends on nutrient and resource availability—and trait complementarity will determine the success of species in mixture (Brooker et al., 2015). Background soil fertility due to unique management legacies is thus expected to influence the composition and productivity of cover crops. This subsequently impacts the functional traits that are expressed, which mediate the effects of cover crops on ecosystem functions including nutrient retention, N supply, and NO_3^- leaching.

While long-term use of cover crops and reduced tillage can reduce NO_3^- leaching compared to conventional management (Syswerda et al., 2012), it is not yet clear how the

adoption of cover crop mixtures may interact with management history to affect nutrient losses from soil. To better understand how distinct management legacies impact the effectiveness of cover crops, we investigated the establishment and productivity of legume and grass cover crops grown alone and in a mixture across a management gradient. The Kellogg Biological Station (KBS) Long-term Ecological Research (LTER) site established four annual cropping systems, hereafter referred to as management legacies, in 1988: 1) conventional, chisel plowed; 2) conventional, no-till, 3) reduced synthetic input, chisel plowed; and 4) biologically-based, certified organic (Robertson et al., 2014). Both the reduced-input and organic legacies have a long-term history of cover crop use while the conventional and no-till systems do not. We utilized this long-standing gradient of management to determine whether soil fertility properties that reflect these distinct management histories affect cover crop biomass and outcomes.

Specifically, to understand the subsequent effect on ecosystem functions, we quantified potential anion loss from the soil in the growing season following the winter cover crop treatments. In addition to measuring soil NO_3^- concentrations in a deep soil core, we used a surface-added bromide tracer applied in the fall after cover crop planting, as bromide percolation can serve as a conservative estimate of water flow in the soil profile, to contribute to our understanding of the cover crops' influence on leaching potential (Hess et al., 2018). Bromide and NO_3^- move similarly in subsoil, so bromide can be a useful proxy for estimating potential NO_3^- losses by leaching (Smith & Davis, 1974). Field studies have shown that bromide and NO_3^- movement in soil is correlated, and although bromide movement likely overestimates NO_3^- leaching because it is not subject to the many biological transformations of the N cycle, bromide is useful in investigating qualitative patterns of anion movement and maximum potential for leaching losses (Clay et al., 2004; Onken et al., 1977). We quantified bromide concentrations in

the fall before frost, and in the summer growing season after the overwintering cover crops, to test how management legacy influences the impact of cover crops grown alone and in mixture on anion movement in soil.

Building on prior research at this site showing differences among legacies in soil properties that reflect management, like increased soil organic carbon and greater soil N availability in the sustainably-managed systems (Grandy & Robertson, 2007; Syswerda et al., 2012; Syswerda & Robertson, 2014), we expected that SOM and other biochemical indicators of soil fertility would be lowest in the conventional legacy due to its history of high-input, intensive management, while more sustainable management in the organic legacy would result in higher background soil fertility. As an indicator biological soil fertility, we measured particulate organic matter (POM), active fractions of SOM that turnover on year to decadal timescales and thus can more sensitively reflect land-use and management-induced changes to the soil (Marriott & Wander, 2006; Nascente et al., 2013).

Our hypothesis was that this management legacy gradient would lead to differences in cover crop establishment and productivity, such as increased legume biomass in the conventional legacies due to lower background soil fertility, and a resulting lower capacity for N retention in that legacy. In contrast, ecologically-based management histories increase soil fertility, which may enhance complementary resource use and multifunctionality in cover crop mixtures through increased evenness. We also expected these differences in cover crop biomass, particularly in mixture composition, to impact potential anion loss. For example, we expected estimates of NO_3^- leaching to be lowest overall in the sole grass treatment because grasses have high capacity for N retention. However, we expected the mixture treatment to achieve levels of nutrient retention comparable to the grass treatment while also providing N supply.

Methods

Site description and treatment design

Measurements were taken from June 2019 - Fall 2020 in the Main Cropping System Experiment (MCSE) of the KBS LTER site (www.lter.kbs.msu.edu) in southwest Michigan (42°240 N, 85°240 W; 288 m elevation). The mean annual air temperature at KBS is 9.2 °C ranging from a monthly mean of -4.1 °C in January to 21.9 °C in July. Rainfall averages 933 mm yr⁻¹. KBS is the only LTER with row crop agriculture and was established in 1988 to reflect a range of field-crop landscapes in the upper Midwest. Each treatment is replicated six times on 1-ha plots and includes four annual cropping systems with corn-soy-wheat rotations and management practices ranging from conventional to organic, three perennial ecosystems (alfalfa, poplar, and coniferous forest), and three successional ecosystems. The MCSE sits on a glacial outwash plain with well drained loam, sandy loam, and sandy clay loam soils in the Kalamzaoo and Oshtemo series which are mixed, mesic Typic Hapludalfs (Crum & Collins, 1995).

We established our cover crop experiment in 12.2 x 12.2 m sections of the northern end of all six replicates of the four annual cropping systems: 1) conventional, chisel plowed; 2) conventional, no-till; 3) reduced synthetic input, chisel plowed; and 4) biologically-based, certified organic. The two conventional legacies receive typical levels of chemical input for farms in the Midwest; the reduced-input legacy receives one-third the amount of herbicide and N fertilizer as the conventional legacies; the biologically based legacy has no chemical inputs, compost, nor manure. In the reduced-input and organic legacies, the winter wheat crop is followed by a red clover cover crop (typically frost-seeded into wheat), while corn harvest is followed by a cereal rye cover crop.

On July 21, 2019, in a randomized complete block design, we planted three cover crop treatments using a no-till drill into each plot following winter wheat harvest: a sole legume, crimson clover (*Trifolium incarnatum* L.) seeded at 16.8 kg ha⁻¹, a sole grass, cereal rye (*Secale cereal* L.) seeded at 100.9 kg ha⁻¹, and a cereal rye (50.4 kg ha⁻¹)-crimson clover (9.0 kg ha⁻¹) mixture, and we compared them to a weedy fallow treatment, which served as our control. Each cover crop treatment plot was 3.1 x 12.2 m. Our design simulates a transition to cover cropping in the conventional legacies and a diversification of cover crop management in the reduced-input and organic legacies. Pesticide was applied to the conventional and reduced-input legacies on May 23rd and 28th, 2020 respectively. Herbicide was applied to the reduced-input legacy on June 25th, 2020. On June 24th, 2020, synthetic N fertilizer (28 UAN, urea-ammonium nitrate with 28% nitrogen) was applied to rye and fallow treatment plots in the two conventional legacies at a rate of 79 kg ha⁻¹.

Aboveground biomass sampling and analysis

We sampled aboveground biomass (AGB) from all treatments in fall 2019 and spring 2020, from one random 0.25 m² quadrat in each replicate plot avoiding edges. Shoot biomass was cut at the soil surface, separated by species, dried at 60 °C for 48 hours, weighed, and coarsely ground (< 2 mm) in a Wiley mill. We analyzed the biomass for total carbon and N content by dry combustion on a Leco TruMac CN Analyzer (Leco Corporation, St. Joseph, MI). Fall cover crop AGB was sampled in all plots on October 24th, 2019. Spring AGB was sampled in plots in the conventional legacy (L1) on May 4, 2020, in the no-till legacy (L2) on May 5, 2020, in the reduced-input legacy (L3) on May 12, 2020, and in the organic legacy (L4) on May 26, 2020. While cover crop biomass in the organic legacy was sampled later than the other legacies, this is accordance with

the cropping system design at KBS, as the organic cropping system typically has a longer growth period for cover crops and later termination date due to later planting of corn. Corn was planted following cover crop biomass sampling in each legacy.

Baseline soil sampling

On June 11 and 12, 2019, we collected a composite, baseline soil sample in the standing wheat crop to determine initial soil conditions. In each plot, we composited 10 soil cores (2 cm diameter by 20 cm depth). We estimated bulk density by taking the fresh weight of the 10 cores and then adjusting for soil moisture. We determined soil moisture gravimetrically by drying duplicate, 20g samples at 105 °C for 48 hours, and then weighing dry soil. We extracted inorganic N ($\text{NO}_3^- + \text{NH}_4^+$) in triplicate with 2 mol L⁻¹ KCl from a subsample passed through a 2-mm sieve. The amount of NO_3^- and NH_4^+ in each sample was analyzed colorimetrically on a discrete analyzer (AQ2; Seal Analytical, Mequon, WI). To determine potential N availability from organic matter decomposition, we sieved subsamples of fresh soil in triplicate to 2 mm for a 7-day anaerobic N mineralization incubation. We then extracted samples in 2 mol L⁻¹ KCl and analyzed them colorimetrically for ammonium. Potentially mineralizable N was calculated as the difference in the initial amount of ammonium in the soil and the ammonium released during the 7-day incubation (Drinkwater et al. 1996).

Subsamples of ~100 g of sieved dried soil were analyzed for pH, organic matter by loss on ignition, Bray-1 P, K^+ , Mg^{2+} , Ca^{2+} , and cation exchange capacity at the A & L Great Lakes Laboratories (Fort Wayne, Indiana, USA). We analyzed soil texture for each sample using the hydrometer method (Gavlak et al., 2005). After air-drying the composite soil samples, we sieved a well-mixed subsample through a 2 mm sieve. Approximately 50 g of soil was mixed and

soaked with 100 mL of sodium hexametaphosphate, blended for 5 min, and transferred to a glass sedimentation cylinder which was filled to the 1L mark with tap water. The slurry was mixed with a metal plunger and then hydrometer readings were taken 40 seconds and 2 hours after the plunger was removed. Percent sand was calculated from the 40 second reading and percent clay from the 2-hour reading. All soil weights were readjusted for soil moisture.

We determined particulate organic matter (POM) by using a combined size and density fractionation method to isolate POM > 53 μm from triplicate 40-g subsamples of unsieved, air-dried soil (Marriott & Wander, 2006).. The subsamples were first gently shaken for 1 h in sodium polytungstate (1.7 g/cm^3), allowed to settle for 16 h, and light fraction POM (also called free POM) floating on top of the solution was removed by aspiration. The remaining sample was shaken with 10% sodium hexametaphosphate to disperse soil aggregates and then rinsed through a 53- μm filter. The material larger than 53 μm was retained, and the intra-aggregate POM (i.e., physically protected POM) was separated from sand by decanting. The carbon and N of both POM fractions were measured on an ECS 4010 CHNSO Analyzer (Costech Analytical Technologies, Valencia, California, USA). Total soil carbon and N (to 20 cm) were measured on dried, sieved soil by dry combustion on a Leco TruMac CN Analyzer.

Bromide application and anion analysis

Bromide was applied as a conservative tracer of water flow through the soil profile, to aid in interpretation of measurements of NO_3^- concentration at depth following cover crop incorporation. On October 17th, 2019, we applied 0.037M KBr in water to four of the six replicates (64 plots total) by using a water transfer pump to spray KBr on a 9 m^2 subplot. The following summer, between July 6-10, 2020, we took a 7.5cm diameter soil core to 120cm using

a Geoprobe from each subplot and divided each core into three depth increments: 0-30cm, 30-60cm, and 60-120cm. We calculated bulk density of every 0-30cm increment by sieving the entire section to 4 mm and separately weighing the sieved soil and the rocks/gravel bigger than 4mm. We calculated bulk density for the other increments (30-60cm and 60-120cm increments) for one randomly selected core per plot (16 total) with the same method we used for the top increment. We determined soil moisture gravimetrically by drying duplicate, 20g samples of 4mm sieved soil from each depth increment at 105 °C for 48 hours, and then weighing dry soil. Subsamples of sieved soil (4 mm) from each increment were extracted with 2 M KCl and analyzed for soil inorganic N colorimetrically on a discrete analyzer (AQ2; Seal Analytical, Mequon, WI). Sieved soil samples were also extracted in deionized water, filtered, and analyzed for bromide concentrations on a Dionex ICS-1000 Ion Chromatography system with a detection limit of 0.02 mg L⁻¹.

Nitrate and bromide content were expressed as a concentration and in kg ha⁻¹ using measured soil bulk density. Bromide retention in the soil was calculated as a percentage, by dividing the concentration of bromide recovered in the deep soil core by the total concentration of bromide applied to that area. We used this calculated percent recovery of bromide to analyze associations between soil fertility, cover crop productivity, and bromide retention in soil.

Statistical analysis

All statistical analysis was performed in R (R Core Team, 2013). To test for differences in soil fertility properties, cover crop biomass, NO₃⁻ concentrations, bromide concentrations, and percent recovery of bromide between management legacies and cover crop treatments, two-way ANOVA analyses were conducted using the *lme4* package for mixed-effect linear models (Bates

et al., 2015). Replicate was treated as a fixed effect and significance was determined at an alpha value of 0.05. We used the *emmeans* function of the *emmeans* package to conduct pairwise comparisons between cover crop treatments and legacy. To test whether baseline soil fertility properties in the different management legacies and cover crop biomass in the experimental treatments influenced potential bromide (or NO_3^-) loss below the root zone, we used linear regressions to model percent recovery of bromide as a function of soil fertility and cover crop biomass. Outliers in the data that were greater than three standard deviations away from the variable mean were removed. Final models were tested for assumptions of independence and normality of residuals. Fall biomass, spring biomass, NO_3^- core concentration and percent recovery of bromide (per depth and total sum) data were transformed using the *transformTukey* function to determine the most precise transformation to meet assumptions.

Results

Baseline soil characteristics

Baseline soil analysis conducted prior to planting the cover crop treatments revealed a gradient of soil fertility across management legacies. Based on several biological and chemical soil properties including SOM, nutrient content, and indicators of microbial activity, the reduced-input and organic legacies had higher fertility than the conventional and no-till legacies (Table 1). Specifically, the organic legacy had significantly higher total organic matter (OM) content, light- and physically-protected particulate organic matter (fPOM and oPOM, respectively), soil NO_3^- concentrations, total inorganic N concentration, and potentially mineralizable N (PMN) compared to the conventional and no-till legacies, due to its long history of biologically-based

management. The reduced-input legacy had significantly higher OM, total carbon and N, and PMN than the two conventional legacies.

Table 1: Mean baseline soil properties by management legacy (values followed by different letters are significantly different at $\alpha < 0.05$; greatest values are bolded).

	<i>Conventional</i>	<i>No-till</i>	<i>Reduced-input</i>	<i>Organic</i>	<i>p-value</i>
OM (%)	1.25 ^a	1.33 ^a	1.74^b	1.74^b	< 0.0001
fPOM (g kg ⁻¹)	2.28 ^a	1.83 ^a	1.83 ^a	2.92^b	< 0.0001
oPOM (g kg ⁻¹)	1.65 ^{ab}	1.47 ^a	2.51 ^{bc}	2.75^c	< 0.0001
C (mg kg ⁻¹)	.747 ^a	.813 ^a	1.02^b	.932 ^{ab}	< 0.0001
N (mg kg ⁻¹)	.066 ^a	.074 ^{ab}	.092^c	.089 ^{bc}	< 0.0001
[NO ₃] (mg kg ⁻¹)	.33 ^a	.17 ^a	.27 ^a	.70^b	< 0.0001
NO ₃ + NH ₄ (mg kg ⁻¹)	.44 ^a	.34 ^a	.40 ^a	.95^b	< 0.0001
PMN (kg ha ⁻¹)	7.95 ^a	5.69 ^a	17.02^b	13.39^b	< 0.0001
P (mg kg ⁻¹)	25.38^b	19.00 ^{ab}	22.62^b	9.31 ^a	0.0004
K (mg kg ⁻¹)	91.81 ^b	103.63 ^{bc}	112.81^c	60.19 ^a	< 0.0001
sand (%)	46.01	49.30	43.63	41.29	0.5811
silt (%)	36.08	35.30	39.96	39.30	0.7573
clay (%)	17.92	15.40	16.40	19.40	0.2833

Cover crop productivity

At the fall sampling, aboveground biomass in the three cover crop treatments ranged from 84 kg ha⁻¹ to 3440 kg ha⁻¹ across all treatments and legacies. The reduced-input management legacy had the highest mean cover crop aboveground biomass overall (mean \pm standard error = 1729 kg ha⁻¹ \pm 184.9; Figure 1A). Both treatment and legacy had a significant effect on biomass and there was a significant treatment and legacy interaction (Table 2). In the conventional and no-till legacies, the sole clover (1585 \pm 214.2 kg ha⁻¹ and 1316 \pm 267.0 kg ha⁻¹, respectively) and ryeclover mixture (1399 \pm 221.1 kg ha⁻¹ and 1143 \pm 255.5 kg ha⁻¹, respectively) treatments had higher mean biomass than the sole rye (533 \pm 81.9 kg ha⁻¹ and 249 \pm 62.9 kg ha⁻¹, respectively)

or weed biomass in the fallow ($354 \pm 81.4 \text{ kg ha}^{-1}$ and $353.33 \pm 123.9 \text{ kg ha}^{-1}$, respectively). In the reduced-input legacy, clover biomass ($2458 \pm 273.8 \text{ kg ha}^{-1}$) was higher on average than rye ($853 \pm 170.8 \text{ kg ha}^{-1}$) and fallow ($1408 \pm 380.4 \text{ kg ha}^{-1}$) treatments. Although there was low biomass in all treatments in the organic legacy in the fall, there was less clover biomass than in the three conventional management legacies, as expected due to its high fertility.

In the spring, biomass ranged from 291 kg ha^{-1} to 5013 kg ha^{-1} . Again, treatment, legacy, and interactions between treatment and legacy were significant (Table 2). Spring biomass trends in the conventional legacies were similar to those in the fall, with higher mean biomass in the sole clover (conventional $2246 \pm 457.7 \text{ kg ha}^{-1}$ and no-till $2953 \pm 129.2 \text{ kg ha}^{-1}$) and mixture (conventional $2056 \pm 176.3 \text{ kg ha}^{-1}$ and no-till $2860 + 281.0 \text{ kg ha}^{-1}$) treatments, following our expectations about soil fertility (Figure 1B). Legume biomass in the sole clover and rye-clover mixture was comparable in the conventional legacies. There were no treatment differences in the reduced-input legacy, but in the organic legacy, the mixture (mean = $4268 \pm 235.2 \text{ kg ha}^{-1}$) had significantly more biomass than the rye (mean = $2935 \pm 188.2 \text{ kg ha}^{-1}$) or weed biomass in the

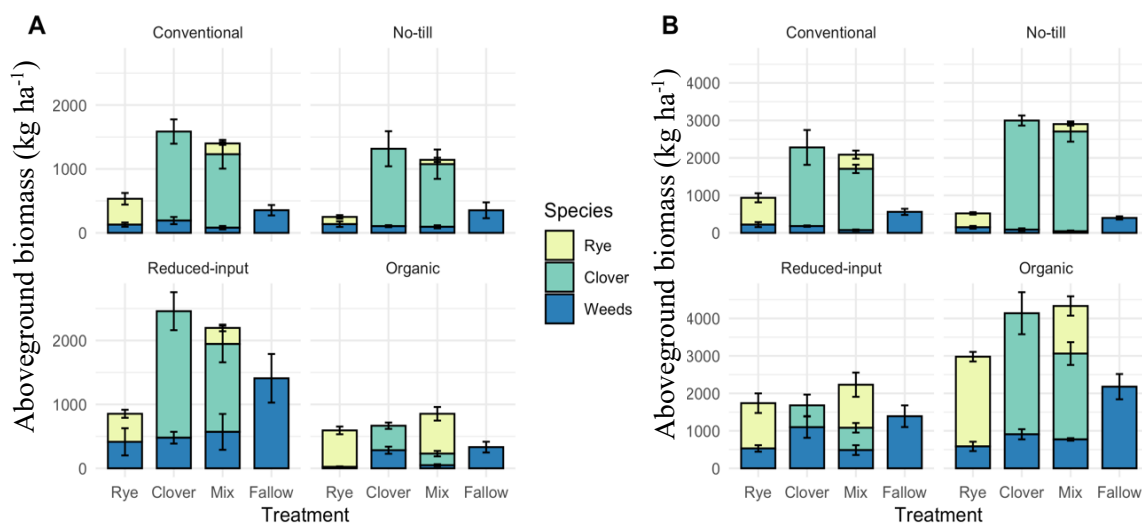


Figure 1: Mean cover crop biomass (with standard error) sampled in the fall (A) and spring (B) by treatment and legacy.

fallow (mean = 2147 ± 332.0 kg ha⁻¹). Overall, the organic legacy had significantly higher mean aboveground biomass means than the other legacies due to the later sampling date.

Our results demonstrate a relationship between higher soil fertility and greater cover crop productivity: the reduced-input and organic legacies had higher fertility soils (Table 1), and cover crop treatments were most productive in the reduced-input legacy in the fall and the organic legacy in the spring. Further, soil fertility also influenced cover crop composition: across legacies, higher fertility generally led to lower legume productivity in both sole-planted and mixture treatments.

Table 2: Results of two-way ANOVA analyses by treatment and legacy for linear mixed-effects models; significant effects are bolded.

	p-value		
	<i>Treatment</i>	<i>Legacy</i>	<i>Treatment*Legacy</i>
Fall AGB	<0.0001	<0.0001	0.0024
Spring AGB	<0.0001	<0.0001	<0.0001
[NO ₃ ⁻]	0.2281	0.7287	0.6206
[Br ⁻]	0.1496	<0.0001	0.2731
Percent recovery (total core)	0.0564	<0.0001	0.3215
Percent recovery (0-30cm)	0.1362	<0.0001	0.9196
Percent recovery (30-60cm)	0.0326	<0.0001	0.7646
Percent recovery (60-120cm)	0.0893	0.6440	0.0055

Nitrate and bromide in soil after cover crop incorporation

At the time of sampling in July 2020, there were no significant differences in mean NO_3^- concentrations between cover crop treatment or management legacy in the deep cores for any depth increment (Table 2). Mean NO_3^- concentrations ranged from 0.62 to 27.75 mg kg^{-1} in the cores (Figure 2A, values in kg ha^{-1} by depth are shown in Supplemental Figure 1A). The pattern for cover crop treatments in the organic legacy, while not statistically significant, tended to follow our expectations, with the highest soil NO_3^- concentrations in the sole clover treatment,

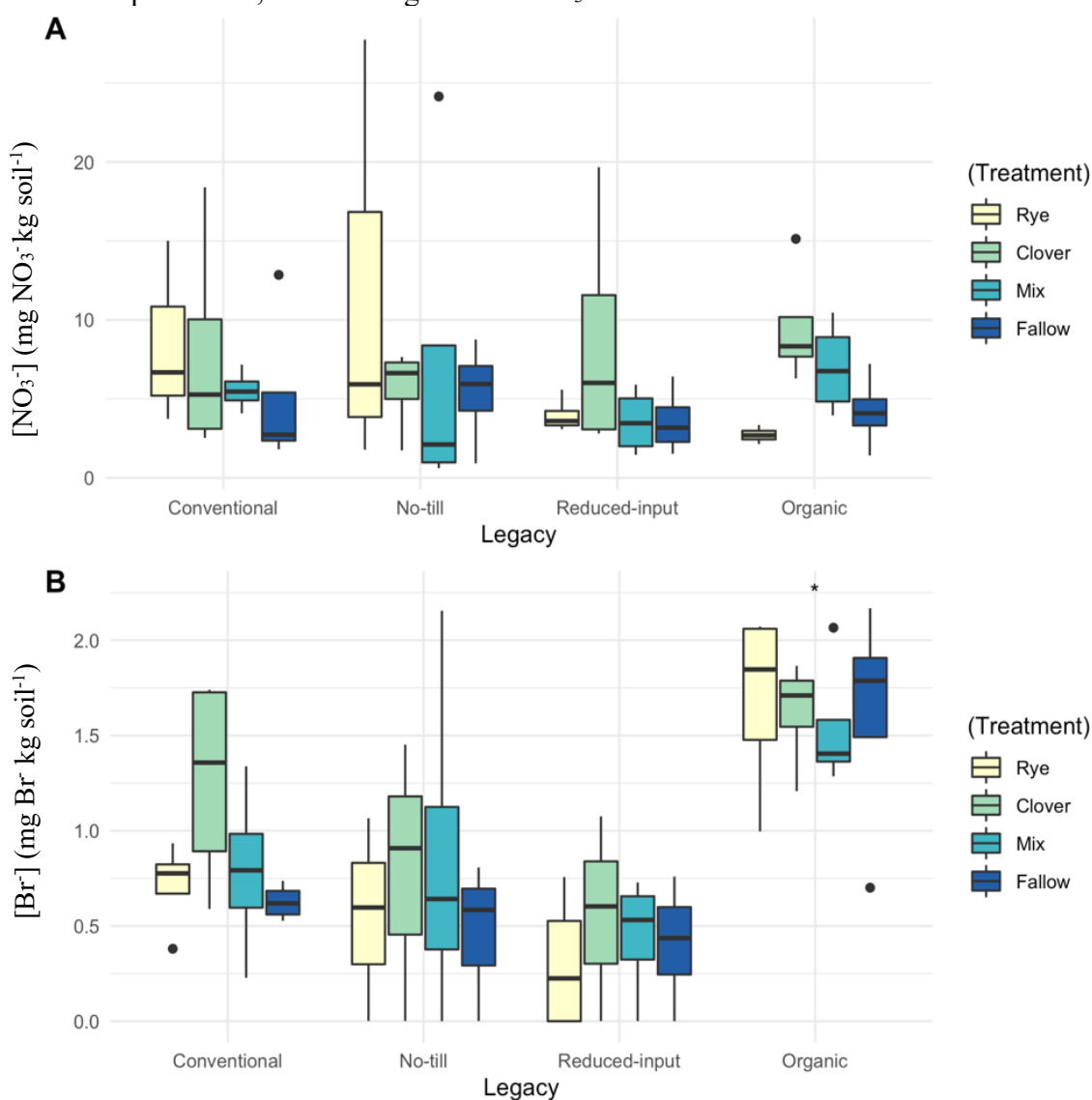


Figure 2: Mean nitrate (A) and bromide (B) concentrations in deep soil cores for entire depth sampled July 6-10, 2020. Asterisks (*) indicate significant difference at $\alpha < 0.05$.

followed by the rye-clover mixture, and the sole rye (Figure 2A). Although mean NO_3^- concentrations did not differ by legacy, the mean concentration of bromide in the organic system was two to three times higher than in the other legacies (Figure 2B; Table 2). Mean bromide concentrations in all cores ranged from 0.17 to 2.17 mg kg^{-1} . This legacy effect was present at all depths, with higher mean amounts of bromide in the organic legacy in each depth increment (Supplemental Figure 1B).

The higher bromide concentration in the core corresponded with a significantly higher percent recovery of added bromide in the soil; the organic legacy had a higher mean percent recovery of bromide throughout the deep core than did the other legacies (mean = 33.28%; Table 2). Percent recovery across legacies ranged from 4.16% to 53.09%. By depth increment, the top segment of the core (0-30cm) had a significantly higher average percent recovery of bromide in

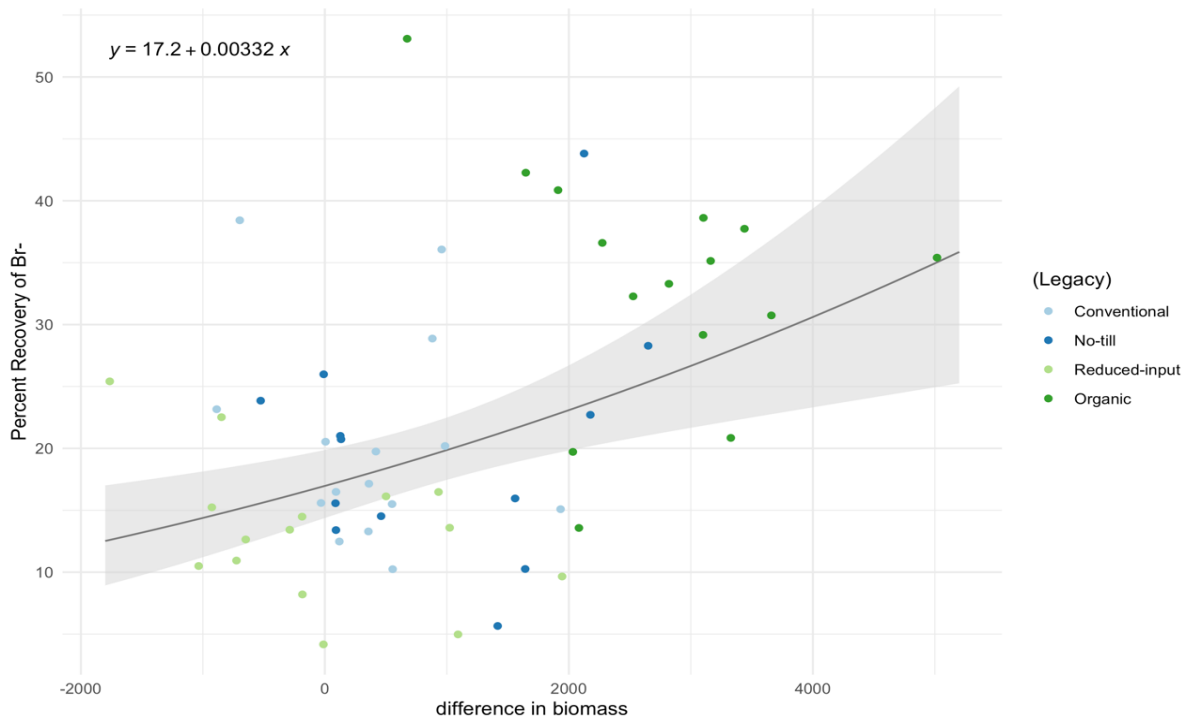


Figure 2: An increase in cover crop biomass from fall to spring was associated with increased percent recovery of bromide, $R^2 = .17$.

the organic legacy compared to the other legacies (Supplemental Figure 2, Table 2). In the middle increment (30-60cm), there was both a significant treatment and legacy effect: the conventional and the organic legacies had higher mean percent recovery than the reduced-input legacy and the clover treatments had higher percent recovery compared to the fallow. In the deepest segment (60-120cm), there was a significant interaction between treatment and legacy, with the highest overall recovery in the fallow treatment in the organic legacy.

Across treatments and legacies, percent recovery of bromide in the deep soil cores was positively associated with an increase in cover crop biomass from fall to spring (Figure 3; p -value = 0.0007; $R^2 = .17$). Percent recovery of added bromide was also strongly positively correlated with light fraction POM (fPOM), indicating that cover crops improved water retention in soil through increased soil fertility (Figure 4; p -value < .0001; $R^2 = .29$).

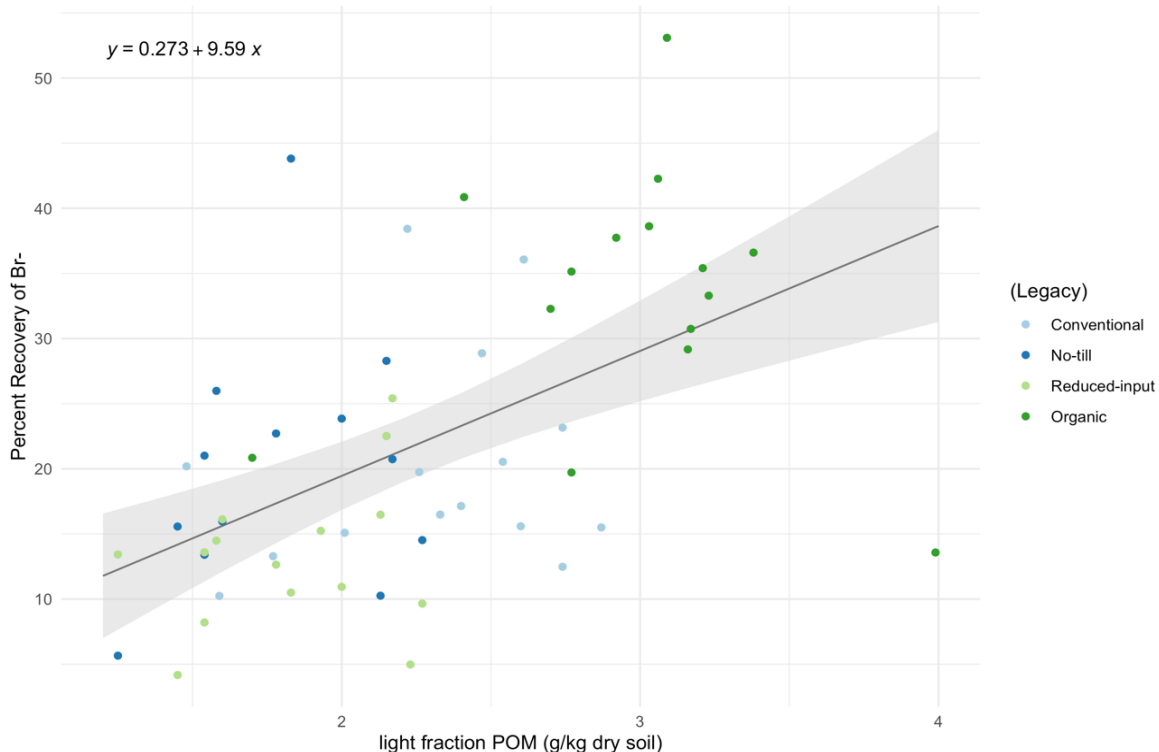


Figure 3: Light fraction particulate organic matter concentration was positively associated with percent recovery of bromide.

Discussion

To determine the effects of agroecosystem management history on cover crop performance, we measured background soil fertility as well as cover crop biomass and composition in four distinct management legacies in a long-term experiment. We applied bromide to estimate water flow through the soil profile to determine how management legacy and overwintering cover crop treatments interact to influence anion movement in soil. We found that a history of ecological management increased multiple soil fertility characteristics (e.g. SOM content, microbial activity, and NO_3^- availability) compared to conventional management legacies. This, in turn, influenced the establishment and growth of cover crops: there was greater overall spring biomass and mixture evenness in the organic legacy compared to the conventional legacies. Both cover crop biomass and indicators of biological soil fertility, particularly free particulate organic matter, were correlated with greater bromide recovery in the soil. Our results indicate that the productivity of cover crops and their potential benefits depend on the past management of the systems into which they are adopted.

Management legacy effects on cover crop establishment and productivity

The four long-term management histories at KBS have led to significant differences in soil health, as evidenced by several soil fertility metrics, such as larger POM fractions and higher rates of microbial activity (e.g., PMN), and higher total OM content in the organic legacy compared to the conventional systems (Table 1). Increases in biological and chemical measures of fertility, in turn, positively impacted cover crop composition and growth.

Cover crop biomass largely followed our predictions regarding soil fertility and cover crop establishment. For example, fall clover biomass in the conventional legacies, which have

low N availability from microbial turnover of SOM, was three to four times higher than in the organic legacy, following ecological understanding that legumes are more competitive in low N soils (Schipanski & Drinkwater, 2012; Vitousek et al., 1987). In the spring, clover continued to dominate the rye-clover mixture biomass in the conventional legacies, while the mixture treatment in the organic legacy had a more even species composition. Rye was more competitive in the organic legacy mixture treatments compared to the other legacies, demonstrating that higher background soil fertility fosters nonlegume growth and increases species evenness in mixture, perhaps enhancing the interaction of agroecosystem N cycling functions provided by legumes and nonlegumes.

Variation in cover crop mixture composition has been previously shown to reflect soil N availability, with low soil inorganic N concentrations favoring legume species in mixture (Baraibar et al., 2020). Further, soil N availability also influences the functions provided by grasses and legumes in mixture. Blesh (2019) found lower rates of BNF by hairy vetch with higher levels of soil N availability from decomposition of POM fractions, and White et al. (2017) found that lower concentrations of soil NO_3^- at cover crop planting allowed for increased legume growth and subsequent N supply without compromising N retention by nonlegumes in cover crop mixtures. Soil fertility resulting from management history thus influences cover crop establishment and composition; this effect is particularly important in cover crop mixtures, where species composition and evenness determines the balance of functions provided.

Cover crop biomass was positively associated with percent recovery of bromide. This is likely due to increased water use by vegetation which decreases flow out of the soil profile, as well as improved nutrient cycling and water retention in soil provided by cover crops. In the 30-60cm core segment, the clover treatment had significantly higher percent recovery of bromide

than the other cover crop treatments across all four legacies, likely because of significantly higher clover biomass compared to rye in most of the legacies at both fall and spring sampling times. In the deepest core segment, there was a significant interaction between treatment and legacy. In the organic legacy, recovery was unexpectedly highest in the fallow plots; however, as the only system without herbicide inputs, it is possible that high weed biomass contributed to the outcome. Otherwise, there were no significant treatment effects of cover crops grown alone or in mixture on anion concentrations or movement in the soil profile.

We expected that increases in cover crop biomass and mixture evenness would decrease NO_3^- content in the soil, as previous research has indicated that grass-legume mixtures can reduce soil NO_3^- content as well as or better than sole grass cover crops (Bergkvist et al., 2011; Sainju et al., 2007; Tosti et al., 2014). There was unusually low rye biomass in the conventional legacies (Figure 1), perhaps due to the summer planting date. Without substantial grass biomass in the mixture, N retention capacity and the potential benefits of multifunctionality in mixture treatments were reduced, as evidenced by the lack of significant differences between cover crop treatments. The mixture and sole-clover treatments were both dominated by clover, which limited our ability to understand the effects of distinct cover crop treatments and mixture composition on soil N dynamics.

Another explanation for the lack of significant differences in soil NO_3^- by treatment or legacy is our deep core sampling date. Due to restrictions on research following COVID-19 we were unable to collect a deep soil core earlier in the growing season. When we were able to sample in late summer, corn was actively growing, and we had missed a key window of N mineralization immediately following cover crop incorporation into soil. Thus, the NO_3^- concentrations measured do not directly reflect the effect of the winter cover treatments, but

instead capture two months of corn growth, N fertilizer inputs in the conventional treatments, and related N cycling dynamics. Given the timing of soil sampling in the plots, it is likely that N fertilizer inputs in the conventional systems ahead of corn planting would have masked any potential differences in soil NO_3^- resulting from residue decomposition in the cover crop and fallow treatments.

Management legacy and anion movement in the soil profile

Our results suggest that long-term use of ecological nutrient management practices, including legume cover crops in rotation, reduces anion movement and potential loss through the soil profile. The organic management legacy had the greatest proportion of bromide recovered from our initial application, suggesting that there was a lower potential for anion movement out of the soil compared to the other legacies, due to the long-term effects of cover crops on soil fertility and hydrology.

Winter cover crops have been shown to improve water infiltration and storage by altering soil physical properties, such as aggregate stability and plant available water, as well as biochemical properties, like SOM, but these effects often take several years to establish (Basche et al., 2016; Basche & DeLonge, 2017; Villamil et al., 2006). Rorick and Kladvko (2017) found that a cereal rye cover crop slightly improved soil aggregate stability and, subsequently, water infiltration but that bulk density and water retention were unchanged after a four years with a winter cover crop in rotation, suggesting that it may take many years of cover crop use before effects on soil water dynamics are measurable. Similarly, Beehler et al. (2017) determined that while a winter rye cover crop increased soil carbon after three years, this time frame was not long enough to establish significant changes to soil chemical properties that would have

improved water retention. While cover crop adoption can introduce numerous benefits to agroecosystems, some benefits, especially those related to soil organic matter and water dynamics, may not be detectable in early years.

Because the benefits of cover crops are more apparent years after adoption, we would expect to see differential effects on nutrient leaching below the root zone in legume and grass cover crops planted alone and in mixture if the experiment was extended (Drinkwater, 2002; Sharma et al., 2018). For example, in Acuña & Villamil's (2014) study, a one-season adoption of a brassica-grass mixture increased NO_3^- retention in a conventional system—likely due to extended plant growth and subsequent N assimilation—but was not enough time to significantly impact soil biogeochemical properties. The strongest association we found between baseline soil properties and anion movement was with free POM, which is a fraction of organic matter that responds to management changes on shorter time scales than the total SOM pool (Nascente et al., 2013). Overall, the long-term influence of management systems on soil properties—rather than short-term changes to management practices, like the introduction of cover crops in the two conventional legacies and cover crop diversification in the reduced-input and organic legacy—account for the relationships we found. The organic system's long history of cover crop use, coupled with its long-term sustainable nutrient management, may best explain its high percent recovery of bromide.

Our calculation of percent recovery assumes that any bromide not recovered in the soil profile would have been lost via leaching, however, Hamilton et al. (2020) found that grasses take up a small portion of applied bromide. The cover crops may have taken up bromide, which would influence our estimates of anion movement; however, we collected the deep cores six to nine weeks after cover crop termination and incorporation back into the soil, so the

decomposition of cover crop residues would have likely returned any bromide assimilated by cover crops to the soil. Further, from an ecosystem perspective, bromide retained in plant residues still reduces its loss via leaching. This potential for plant uptake may help explain the relationship between cover crop biomass and percent recovery of bromide that we found. Future studies should test for possible plant uptake of bromide to ensure all movement of the tracer is considered. It is also possible that greater evapotranspiration in treatments with higher cover crop biomass drove this effect.

Although we did not find significant differences in mean soil NO_3^- concentrations among legacies or cover crop treatments at any depth, differences in average bromide concentrations and percent recovery may correlate with trends in anion movement that could be extended to potential NO_3^- leaching below the root zone (Ottman et al., 2000; Schuh et al., 1997; Smith & Davis, 1974). There were several rain events (Supplemental Figure 3) before our sampling date, so soil NO_3^- may have been lost before we sampled. Based on calculated percent recovery, mean estimates of bromide lost by leaching range from 15.68 kg ha⁻¹ in the organic legacy to 18.76 kg ha⁻¹ in the conventional legacy. These are estimates of maximum potential NO_3^- leaching; because it does not undergo the same biologically-mediated transformations as NO_3^- , bromide only represents one pathway of N movement (i.e., it does not reflect N that may be lost as a gas or assimilated into plants, microbial biomass, and SOM pools), but can qualitatively represent potential NO_3^- loss (Onken et al. 1977). Kessavalou et al. (1996) determined bromide to be a convenient estimate of short-term NO_3^- leaching in corn production systems, as it followed movement patterns of a ¹⁵N tracer through the soil, although bromide leaching measurements were higher than NO_3^- because of N volatilization losses and the immobilization of N in SOM. Clay et al (2004) similarly found that bromide can be an adequate tracer of NO_3^- , but it

overestimates leaching potential if used as a quantitative proxy because it moves faster than NO_3^- through the soil profile. Even though we did not detect differences in soil NO_3^- below the root zone, our results show significant relationships between anion movement through the soil, cover crop productivity, and soil fertility that may coincide with similar relationships involving NO_3^- leaching. For example, Syswerda et al (2014) found that not only did the organic legacy at KBS leach less NO_3^- than the other annual row crop legacies, but that soil carbon, an indicator of soil fertility, was negatively correlated with leaching, suggesting that the patterns we found accurately reflect NO_3^- leaching potential.

Implications for agroecological management

During transitions to more diverse crop rotations, results of this study suggest that farmers should consider the legacy effects of past management when choosing cover crop types. There are also other factors at play in addition to ecological interactions—economic and labor costs of planting cover crops, for example—and farmers must account for multiple dimensions at once to achieve the intended benefits from cover cropping (Bergtold et al., 2019). For example, we found that legume biomass was comparable in monoculture and mixture in the conventional legacies; thus, planting a sole legume in lower-fertility soils may be more economically advantageous than a mixture while providing the same ecosystem function in the early years of adoption.

Studies that investigated the fate of different N sources, like fertilizer versus legume N, have found that conventional farm management with regular fertilizer use and/or low crop diversity results in large N surpluses and N-saturated fields (Blesh & Drinkwater, 2013; Gardner & Drinkwater, 2009) that are much more susceptible to N loss via leaching (Drinkwater & Snapp, 2007; Ross et al., 2008). In a low fertility system with high N surpluses, a mixture may

therefore be desirable for scavenging surplus N and improving agroecosystem nutrient retention during the early stages of adoption. Indeed, mixtures improve ecosystem functions better than sole-grown legumes in a variety of contexts (Nyfeler et al., 2011; Reiss & Drinkwater, 2020). Still, due to the complex interactions between management history and cover crop functions, benefits of mixtures may not be seen until later years, after cover crops begin to improve fertility. Future studies should consider how multiple seasons of cover cropping across a management gradient affect anion movement to more clearly understand the interaction between background soil fertility, biomass production, and potential anion loss for specific cover treatments.

While these complex dynamics may not be revealed until several years after cover crop adoption, short-term use of cover crops can improve soil health (Blesh, 2019). A recent meta-analysis of cover crop impacts on soil health found that several indicators increased after short-term (< 1 year) cover crop adoption, including microbial activity, water infiltration and runoff, and N mineralization (Stewart et al., 2018). Gentry et al. (2013) found that increases in soil N availability were fully realized after just one year of legume cover crop introduction to a conventional system. Although it may take several years for ecosystem functions provided by cover crops to be fully demonstrated, this suggests that transitioning to cover cropping, particularly in low-fertility conventional systems with degraded soil health, may prove worthwhile even in early adoption years.

Field- and farm-level management practices clearly influence cover crop productivity and subsequent impacts on ecosystem services related to N cycling. Cover crop effects on soil health and fertility, N retention and loss, and water dynamics, may be further optimized by precise management of cover crops themselves. Modeling of rye cover growth determined that specific planting dates and growth periods can maximize reductions in N loss (Feyereisen et al., 2006).

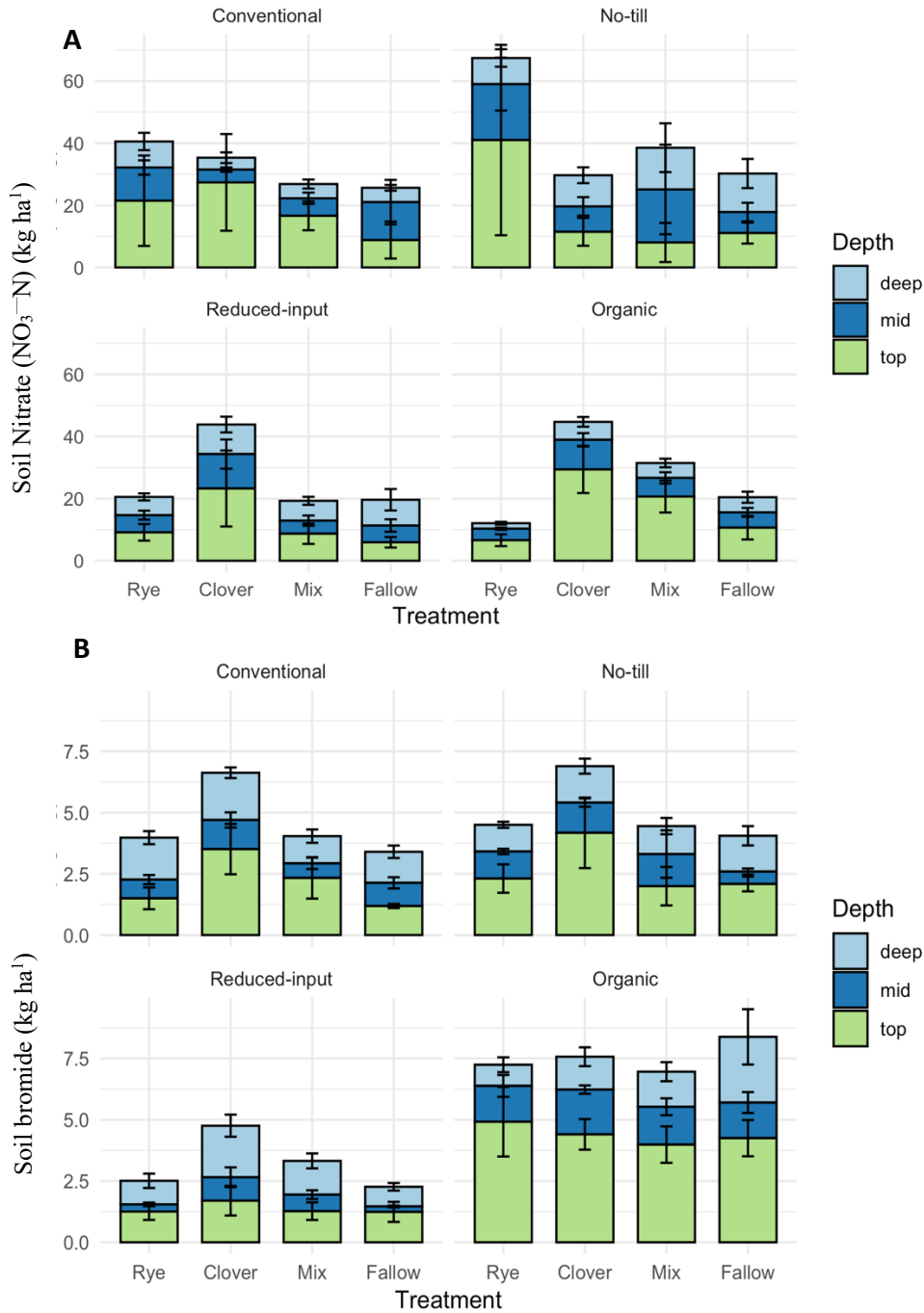
Precise cover crop termination timing and techniques can better synchronize N release from decomposition with N demand of the subsequent crop, further reducing potential for N loss (Dabney et al., 2001; Wortman et al., 2012). Careful attention to management practices may enhance cover crop performance, especially when transitioning to their use (i.e. before long-term benefits are realized).

Conclusions

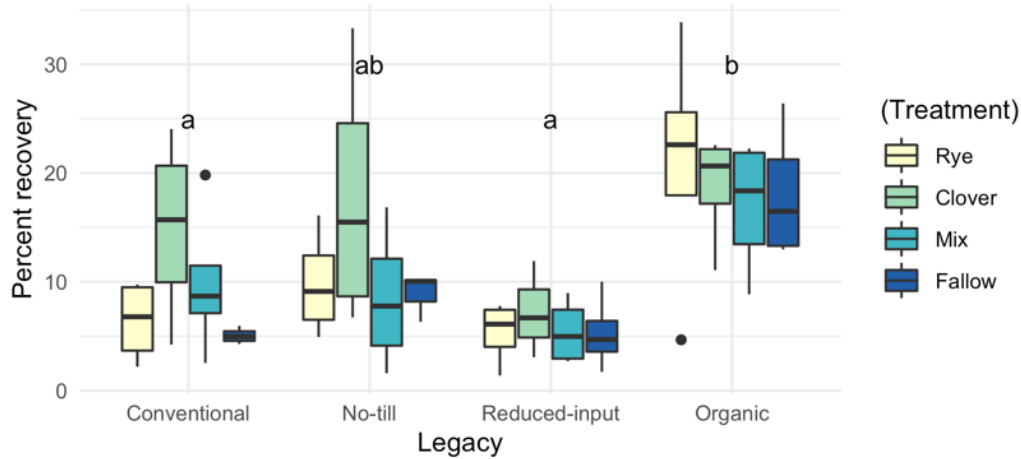
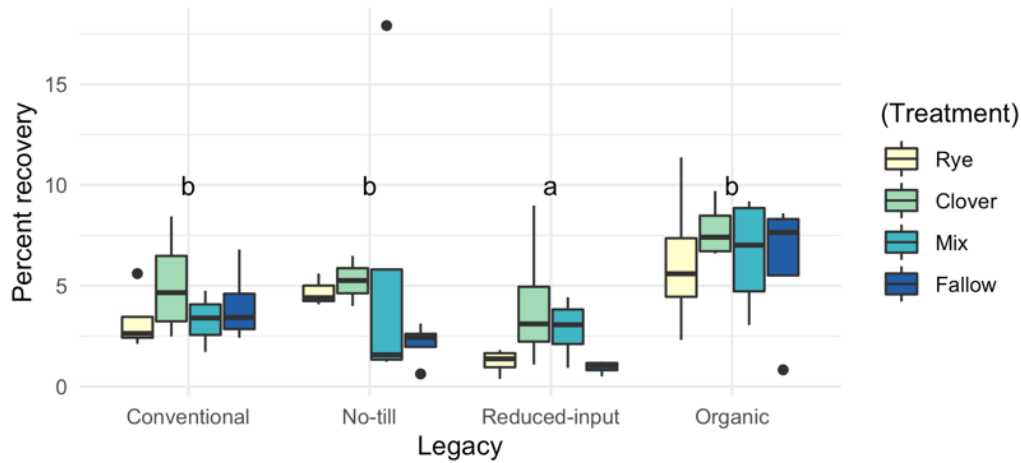
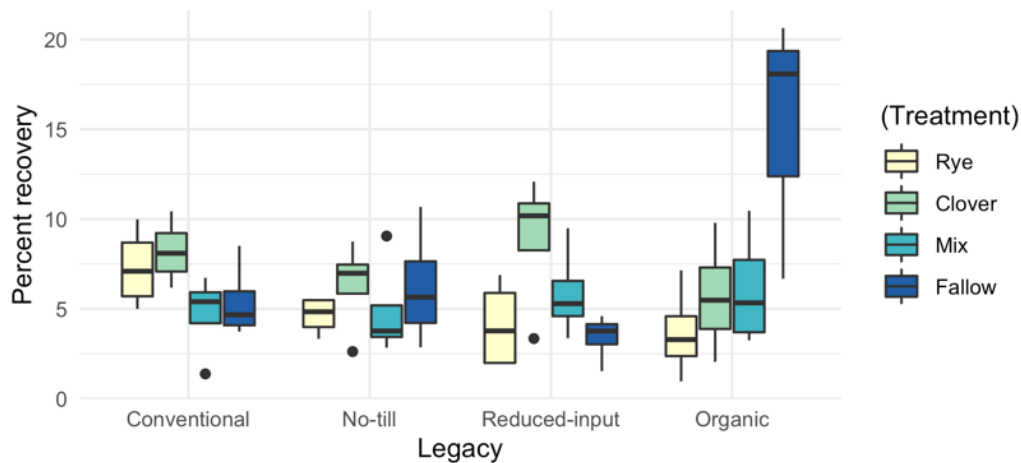
The objective of this study was to determine how cover crop growth and function were influenced by management history. Our results show that long-term use of ecological nutrient management practices established high levels of soil fertility, such as increased OM content, microbial activity, and nutrient availability, that positively affected cover crop production and reduced the potential for nutrient losses via leaching. While distinct cover crop treatments did not have a strong effect on anion movement in any of the legacies, we found significant differences in cover crop establishment among legacies due to their background soil fertility that likely correspond with the systems' capacities to reduce leaching. These findings indicate two things: first, that soil fertility impacts cover crop productivity in ways that may enhance species interactions and ecosystem functions after longer periods of adoption. Second, the reverse relationship—in which cover crop productivity influences soil chemical and physical properties over time—can reduce anion loss from soil. This complex interaction between soil fertility, cover crop growth, and reduced anion potential is not only dependent on initial soil conditions but has the potential to evolve with sustained cover crop use and subsequent soil fertility changes. Future research should further investigate how these interactions change over time and in different agricultural settings. Overall, this study provides valuable insight into the

fundamental relationships that mediate the potential for cover crops to reduce nutrient leaching across management gradients.

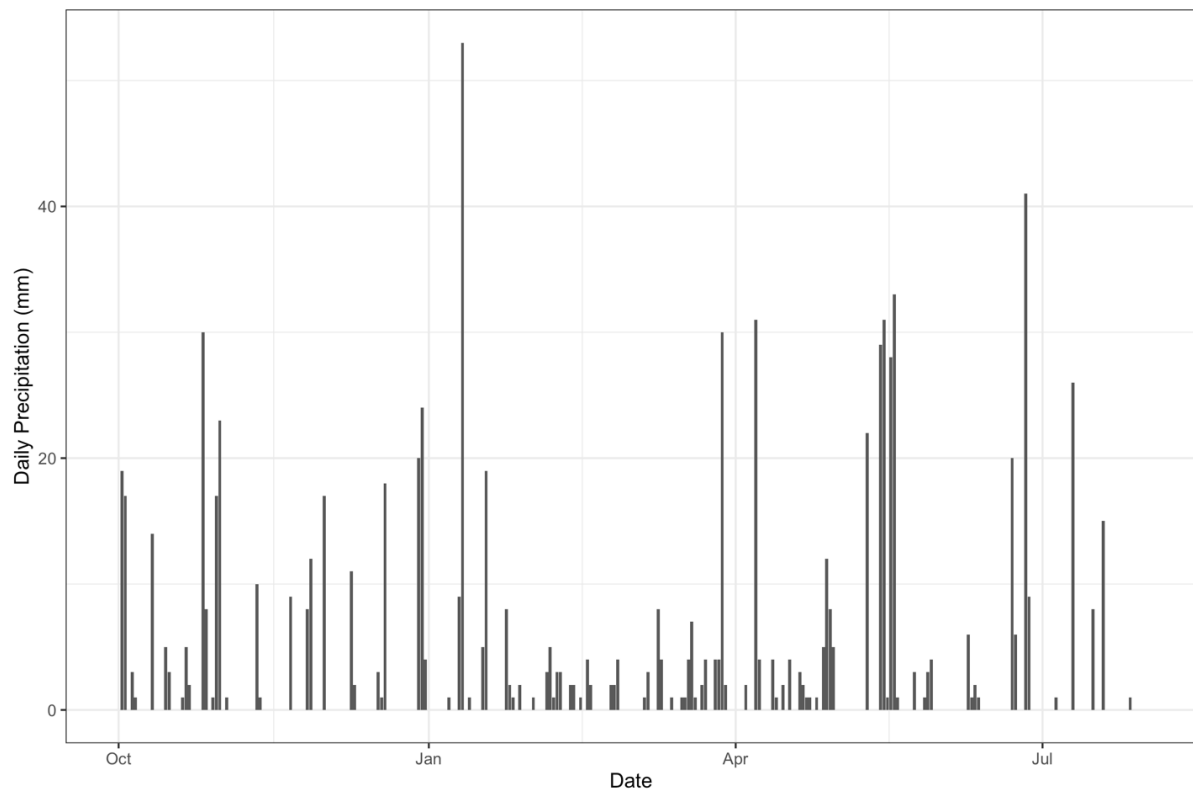
Appendix



Supplemental Figure 1: Mean amount of nitrate (A) and bromide (B) in kilograms of $\text{NO}_3^- \text{N}$ or Br per hectare (with standard error) by depth, sampled July 6-10, 2020. There were no significant differences in the mean nitrate concentrations in any depth increment (A), but the organic legacy had significantly higher mean concentrations of bromide compared to the other legacies in each depth increment (B).

A 0-30cm segment**B** 30-60cm segment**C** 60-120cm segment

Supplemental Figure 2: Mean percent recovery of applied bromide by depth (different letters indicate significant difference between legacies at an $\alpha < 0.05$). In the 60-120cm segment, there was a significant treatment by legacy interaction in which fallow plots in the organic legacy had a higher mean percent recovery of bromide.



Supplemental Figure 3: Daily precipitation at KBS from the time of bromide application in fall 2019 to deep core sampling in summer 2020.

Bibliography

- Acuña, J. C. M., & Villamil, M. B. (2014). Short-term effects of cover crops and compaction on soil properties and soybean production in Illinois. *Agronomy Journal*, *106*(3), 860–870. <https://doi.org/10.2134/agronj13.0370>
- Baraibar, B., Murrell, E. G., Bradley, B. A., Barbercheck, M. E., Mortensen, D. A., Kaye, J. P., & White, C. M. (2020). Cover crop mixture expression is influenced by nitrogen availability and growing degree days. *PLoS ONE*, *15*(7 July), 1–15. <https://doi.org/10.1371/journal.pone.0235868>
- Basche, A. D., Kaspar, T. C., Archontoulis, S. V., Jaynes, D. B., Sauer, T. J., Parkin, T. B., & Miguez, F. E. (2016). Soil water improvements with the long-term use of a winter rye cover crop. *Agricultural Water Management*, *172*, 40–50. <https://doi.org/10.1016/j.agwat.2016.04.006>
- Basche, A., & DeLonge, M. (2017). The Impact of Continuous Living Cover on Soil Hydrologic Properties: A Meta-Analysis. *Soil Science Society of America Journal*, *81*(5), 1179–1190. <https://doi.org/10.2136/sssaj2017.03.0077>
- Bates, D., Mächler, M., Bolker, B. M., & Walker, S. C. (2015). *Fitting linear mixed-effects models using lme4*.
- Beehler, J., Fry, J., Negassa, W., & Kravchenko, A. (2017). Impact of cover crop on soil carbon accrual in topographically diverse terrain. *Journal of Soil and Water Conservation*, *72*(3), 272–279. <https://doi.org/10.2489/jswc.72.3.272>
- Bergkvist, G., Stenberg, M., Wetterlind, J., Båth, B., & Elfstrand, S. (2011). Clover cover crops under-sown in winter wheat increase yield of subsequent spring barley-Effect of N dose and companion grass. *Field Crops Research*, *120*(2), 292–298. <https://doi.org/10.1016/j.fcr.2010.11.001>
- Bergtold, J. S., Ramsey, S., Maddy, L., & Williams, J. R. (2019). A review of economic considerations for cover crops as a conservation practice. *Renewable Agriculture and Food Systems*, *34*(1), 62–76. <https://doi.org/10.1017/S1742170517000278>
- Blesh, J., & Drinkwater, L. E. (2013). The impact of nitrogen source and crop rotation on nitrogen mass balances in the Mississippi River Basin. *Ecological Applications*, *23*(5), 1017–1035. <https://doi.org/10.1890/12-0132.1>
- Blesh, Jennifer. (2019). Feedbacks between nitrogen fixation and soil organic matter increase ecosystem functions in diversified agroecosystems. *Ecological Applications*, *29*(8), 1–12. <https://doi.org/10.1002/eap.1986>
- Blesh, Jennifer, Hoey, L., Jones, A. D., Friedmann, H., & Perfecto, I. (2019). Development pathways toward “zero hunger.” *World Development*, *118*, 1–14. <https://doi.org/10.1016/j.worlddev.2019.02.004>
- Booth, M. S., Stark, J. M., & Rastetter, E. (2005). Controls on nitrogen cycling in terrestrial ecosystems: A synthetic analysis of literature data. *Ecological Monographs*, *75*(2), 139–157. <https://doi.org/10.1890/04-0988>
- Brooker, R. W., Bennett, A. E., Cong, W. F., Daniell, T. J., George, T. S., Hallett, P. D., Hawes, C., Iannetta, P. P. M., Jones, H. G., Karley, A. J., Li, L., Mckenzie, B. M., Pakeman, R. J., Paterson, E., Schöb, C., Shen, J., Squire, G., Watson, C. A., Zhang, C., ... White, P. J. (2015). Improving intercropping: A synthesis of research in agronomy, plant physiology and ecology. *New Phytologist*, *206*(1), 107–117. <https://doi.org/10.1111/nph.13132>
- Clay, D. E., Zheng, Z., Liu, Z., Clay, S. A., & Trooien, T. P. (2004). Bromide and Nitrate

- Movement through Undisturbed Soil Columns. *Journal of Environmental Quality*, 33(1), 338–342. <https://doi.org/10.2134/jeq2004.3380>
- Cotrufo, M. F., Wallenstein, M. D., Boot, C. M., Deneff, K., & Paul, E. (2013). The Microbial Efficiency-Matrix Stabilization (MEMS) framework integrates plant litter decomposition with soil organic matter stabilization: Do labile plant inputs form stable soil organic matter? *Global Change Biology*, 19(4), 988–995. <https://doi.org/10.1111/gcb.12113>
- Crum, J. R., & Collins, H. P. (1995). KBS Soils. KBS LTER Special Publication. *Zenodo*, 1–2.
- Dabney, S. M., Delgado, J. A., & Reeves, D. W. (2001). Using winter cover crops to improve soil and water quality. *Communications in Soil Science and Plant Analysis*, 32(7–8), 1221–1250. <https://doi.org/10.1081/CSS-100104110>
- Diaz, R. J., & Rosenberg, R. (2008). Spreading dead zones and consequences for marine ecosystems. *Science*, 321(5891), 926–929. <https://doi.org/10.1126/science.1156401>
- Drinkwater, L. E. (2002). Cropping systems research: Reconsidering agricultural experimental approaches. *HortTechnology*, 12(3), 355–361. <https://doi.org/10.21273/horttech.12.3.355>
- Drinkwater, L., & Snapp, S. (2007). Nutrients in Agroecosystems: Rethinking the Management Paradigm. *Advances in Agronomy*, 92(May 2018), 163–186. [https://doi.org/10.1016/S0065-2113\(04\)92003-2](https://doi.org/10.1016/S0065-2113(04)92003-2)
- Feyereisen, G. W., Wilson, B. N., Sands, G. R., Strock, J. S., & Porter, P. M. (2006). Potential for a rye cover crop to reduce nitrate loss in southwestern Minnesota. *Agronomy Journal*, 98(6), 1416–1426. <https://doi.org/10.2134/agronj2005.0134>
- Finney, D. M., & Kaye, J. P. (2017). Functional diversity in cover crop polycultures increases multifunctionality of an agricultural system. *Journal of Applied Ecology*, 54(2), 509–517. <https://doi.org/10.1111/1365-2664.12765>
- Finney, D. M., White, C. M., & Kaye, J. P. (2016). Biomass production and carbon/nitrogen ratio influence ecosystem services from cover crop mixtures. *Agronomy Journal*, 108(1), 39–52. <https://doi.org/10.2134/agronj15.0182>
- Gabriel, J. L., Muñoz-Carpena, R., & Quemada, M. (2012). The role of cover crops in irrigated systems: Water balance, nitrate leaching and soil mineral nitrogen accumulation. *Agriculture, Ecosystems and Environment*, 155(3), 50–61. <https://doi.org/10.1016/j.agee.2012.03.021>
- Galaviz-Villa, I., Landeros-Sánchez, C., Castañeda-Chávez, M. del R., P. Martínez-Dávila, J., Pérez-Vázquez, A., Nikolskii-Gavrilov, I., & Lango-Reynoso, F. (2010). Agricultural Contamination of Subterranean Water with Nitrates and Nitrites: An Environmental and Public Health Problem. *Journal of Agricultural Science*, 2(2). <https://doi.org/10.5539/jas.v2n2p17>
- Gardner, J. B., & Drinkwater, L. E. (2009). The fate of nitrogen in grain cropping systems: A meta-analysis of 15N field experiments. *Ecological Applications*, 19(8), 2167–2184. <https://doi.org/10.1890/08-1122.1>
- Gavlak, R., Horneck, R., & Miller, R. O. (2005). Soil, Plant and Water Reference Methods for the Western Region. *Western Regional Extension Publication (WREP) 125, 1*, 129–134.
- Gentry, L. E., Snapp, S. S., Price, R. F., & Gentry, L. F. (2013). Apparent red clover Nitrogen credit to corn: Evaluating cover crop introduction. *Agronomy Journal*, 105(6),

- 1658–1664. <https://doi.org/10.2134/agronj2013.0089>
- Grandy, A. S., & Robertson, G. P. (2007). Land-use intensity effects on soil organic carbon accumulation rates and mechanisms. *Ecosystems*, *10*(1), 58–73. <https://doi.org/10.1007/s10021-006-9010-y>
- Hamilton, S. K. (2020). *Bromide Addition to Test for Vertical Percolation and Plant Uptake*. November, 1–4.
- Hayden, Z. D., Ngouajio, M., & Brainard, D. C. (2014). Rye-vetch mixture proportion tradeoffs: Cover crop productivity, nitrogen accumulation, and weed suppression. *Agronomy Journal*, *106*(3), 904–914. <https://doi.org/10.2134/agronj2013.0467>
- Hess, L. J. T., Hinckley, E. L. S., Robertson, G. P., Hamilton, S. K., & Matson, P. A. (2018). Rainfall intensification enhances deep percolation and soil water content in tilled and no-till cropping systems of the US midwest. *Vadose Zone Journal*, *17*(1). <https://doi.org/10.2136/vzj2018.07.0128>
- Kaspar, T. C., Jaynes, D. B., Parkin, T. B., Moorman, T. B., & Singer, J. W. (2012). Effectiveness of oat and rye cover crops in reducing nitrate losses in drainage water. *Agricultural Water Management*, *110*(3), 25–33. <https://doi.org/10.1016/j.agwat.2012.03.010>
- Kaye, J., Finney, D., White, C., Bradley, B., Schipanski, M., Alonso-Ayuso, M., Hunter, M., Burgess, M., & Mejia, C. (2019). Managing nitrogen through cover crop species selection in the U.S. Mid-Atlantic. *PLoS ONE*, *14*(4), 1–23. <https://doi.org/10.1371/journal.pone.0215448>
- Kessavalou, A., Doran, J. W., Powers, W. L., Kettler, T. A., & Qian, J. H. (1996). Bromide and Nitrogen-15 Tracers of Nitrate Leaching under Irrigated Corn in Central Nebraska. *Journal of Environmental Quality*, *25*(5), 1008–1014. <https://doi.org/10.2134/jeq1996.00472425002500050012x>
- King, A. E., & Blesh, J. (2018). Crop rotations for increased soil carbon: Perenniality as a guiding principle: Perenniality. *Ecological Applications*, *28*(1), 249–261. <https://doi.org/10.1002/eap.1648>
- Marriott, E. E., & Wander, M. (2006). *Qualitative and quantitative differences in particulate organic matter fractions in organic and conventional farming systems*. *38*, 1527–1536. <https://doi.org/10.1016/j.soilbio.2005.11.009>
- Matson, P. A., Parton, W. ., Power, A. G., & Swift, M. J. (1997). Agricultural Intensification and Ecological Properties. *Science*, *277*(July), 191–218. <https://doi.org/10.2307/j.ctvdjrr1w.13>
- Nascente, A. S., Li, Y. C., & Crusciol, C. A. C. (2013). Cover crops and no-till effects on physical fractions of soil organic matter. *Soil and Tillage Research*, *130*, 52–57. <https://doi.org/10.1016/j.still.2013.02.008>
- Nyfelner, D., Huguenin-Elie, O., Suter, M., Frossard, E., & Lüscher, A. (2011). Grass-legume mixtures can yield more nitrogen than legume pure stands due to mutual stimulation of nitrogen uptake from symbiotic and non-symbiotic sources. *Agriculture, Ecosystems and Environment*, *140*(1–2), 155–163. <https://doi.org/10.1016/j.agee.2010.11.022>
- Onken, A. B., Wendt, C. W., Hargrove, R. S., & Wilke, O. C. (1977). Relative Movement of Bromide and Nitrate in Soils under Three Irrigation Systems. *Soil Science Society of America Journal*, *41*(1), 50–52. <https://doi.org/10.2136/sssaj1977.03615995004100010018x>
- Ottman, M. J., Tickes, B. R., & Husman, S. H. (2000). Nitrogen-15 and Bromide Tracers of

- Nitrogen Fertilizer Movement in Irrigated Wheat Production. *Journal of Environmental Quality*, 29(3), 1500–1508.
- Puget, P., & Drinkwater, L. E. (2001). Short-Term Dynamics of Root- and Shoot-Derived Carbon from a Leguminous Green Manure. *Soil Science Society of America Journal*, 65(3), 771–779. <https://doi.org/10.2136/sssaj2001.653771x>
- Ranells, N. N., & Waggoner, M. G. (1997). Winter annual grass-legume bicultures for efficient nitrogen management in no-till corn. *Agriculture, Ecosystems and Environment*, 65(1), 23–32. [https://doi.org/10.1016/S0167-8809\(97\)00054-6](https://doi.org/10.1016/S0167-8809(97)00054-6)
- Reiss, E. R., & Drinkwater, L. E. (2020). Ecosystem service delivery by cover crop mixtures and monocultures is context dependent. *Agronomy Journal*, 112(5), 4249–4263. <https://doi.org/10.1002/agj2.20287>
- Robertson, G. P., Gross, K. L., Hamilton, S. K., Landis, D. A., Schmidt, T. M., Snapp, S. S., & Swinton, S. M. (2014). Farming for ecosystem services: An ecological approach to production agriculture. *BioScience*, 64(5), 404–415. <https://doi.org/10.1093/biosci/biu037>
- Rorick, J. D., & Kladvik, E. J. (2017). Cereal rye cover crop effects on soil carbon and physical properties in southeastern Indiana. *Journal of Soil and Water Conservation*, 72(3), 260–265. <https://doi.org/10.2489/jswc.72.3.260>
- Ross, S. M., Izaurralde, R. C., Janzen, H. H., Robertson, J. A., & McGill, W. B. (2008). The nitrogen balance of three long-term agroecosystems on a boreal soil in western Canada. *Agriculture, Ecosystems and Environment*, 127(3–4), 241–250. <https://doi.org/10.1016/j.agee.2008.04.007>
- Sainju, U. M., Singh, B. P., Whitehead, W. F., & Wang, S. (2007). Accumulation and crop uptake of soil mineral nitrogen as influenced by tillage, cover crops, and nitrogen fertilization. *Agronomy Journal*, 99(3), 682–691. <https://doi.org/10.2134/agronj2006.0177>
- Schipanski, M. E., Drinkwater, L. E., & Russelle, M. P. (2010). Understanding the variability in soybean nitrogen fixation across agroecosystems. *Plant and Soil*, 329(1), 379–397. <https://doi.org/10.1007/s11104-009-0165-0>
- Schipanski, Meagan E., & Drinkwater, L. E. (2012). Nitrogen fixation in annual and perennial legume-grass mixtures across a fertility gradient. *Plant and Soil*, 357(1), 147–159. <https://doi.org/10.1007/s11104-012-1137-3>
- Schuh, W. M., Klinkebiel, D. L., Gardner, J. C., & Meyer, R. F. (1997). Tracer and Nitrate Movement to Groundwater in the Northern Great Plains. *Journal of Environmental Quality*, 26(5), 1335–1347. <https://doi.org/10.2134/jeq1997.00472425002600050020x>
- Sharma, P., Singh, A., Kahlon, C. S., Brar, A. S., Grover, K. K., Dia, M., & Steiner, R. L. (2018). The Role of Cover Crops towards Sustainable Soil Health and Agriculture—A Review Paper. *American Journal of Plant Sciences*, 09(09), 1935–1951. <https://doi.org/10.4236/ajps.2018.99140>
- Smith, S. J., & Davis, R. J. (1974). Relative Movement of Bromide and Nitrate Through Soils. *Journal of Chemical Information and Modeling*, 53(9), 1689–1699.
- Snapp, S. S., Swinton, S. M., Labarta, R., Mutch, D., Black, J. R., Leep, R., & Nyiraneza, J. (2005). Evaluating Cover Crops for Benefits, Costs and Performance within Cropping System Niches. *Agronomy Journal*, 97(i), 322–332. <https://doi.org/10.2134/agronj2005.0322>
- Stewart, R. D., Jinshi, J., Gyawali, A. J., Thomason, W. E., Badgley, B. D., Reiter, M. S., &

- Strickland, M. S. (2018). What We Talk About When We Talk About Soil Health. *Agricultural & Environmental Letters*, 327–340. <https://doi.org/10.2307/j.ctt1zxz145.18>
- Syswerda, S. P., Basso, B., Hamilton, S. K., Tausig, J. B., & Robertson, G. P. (2012). Long-term nitrate loss along an agricultural intensity gradient in the Upper Midwest USA. *Agriculture, Ecosystems and Environment*. <https://doi.org/10.1016/j.agee.2011.12.007>
- Syswerda, S. P., & Robertson, G. P. (2014). Ecosystem services along a management gradient in Michigan (USA) cropping systems. *Agriculture, Ecosystems and Environment*, 189, 28–35. <https://doi.org/10.1016/j.agee.2014.03.006>
- Thorup-Kristensen, K., & Dresbøll, D. B. (2010). Incorporation time of nitrogen catch crops influences the N effect for the succeeding crop. *Soil Use and Management*, 26(1), 27–35. <https://doi.org/10.1111/j.1475-2743.2009.00255.x>
- Thorup-Kristensen, Kristian, & Nielsen, N. E. (1998). Modelling and measuring the effect of nitrogen catch crops on the nitrogen supply for succeeding crops. *Plant and Soil*, 203(1), 79–89. <https://doi.org/10.1023/A:1004398131396>
- Tonitto, C., David, M. B., & Drinkwater, L. E. (2006). Replacing bare fallows with cover crops in fertilizer-intensive cropping systems: A meta-analysis of crop yield and N dynamics. *Agriculture, Ecosystems and Environment*, 112(1), 58–72. <https://doi.org/10.1016/j.agee.2005.07.003>
- Tosti, G., Benincasa, P., Farneselli, M., Tei, F., & Guiducci, M. (2014). Barley-hairy vetch mixture as cover crop for green manuring and the mitigation of N leaching risk. *European Journal of Agronomy*, 54, 34–39. <https://doi.org/10.1016/j.eja.2013.11.012>
- Villamil, M. B., Bollero, G. A., Darmody, R. G., Simmons, F. W., & Bullock, D. G. (2006). No-Till Corn/Soybean Systems Including Winter Cover Crops. *Soil Science Society of America Journal*, 70(6), 1936–1944. <https://doi.org/10.2136/sssaj2005.0350>
- Vitousek, P. M., Walker, L. R., Whiteaker, L. D., Mueller-Dombois, D., & Matson, P. A. (1987). Biological invasion by *Myrica faya* alters ecosystem development in Hawaii. *Science*, 238(4828), 802–804. <https://doi.org/10.1126/science.238.4828.802>
- White, C. M., DuPont, S. T., Hautau, M., Hartman, D., Finney, D. M., Bradley, B., LaChance, J. C., & Kaye, J. P. (2017). Managing the trade off between nitrogen supply and retention with cover crop mixtures. *Agriculture, Ecosystems and Environment*, 237, 121–133. <https://doi.org/10.1016/j.agee.2016.12.016>
- Wortman, S. E., Francis, C. A., Bernards, M. L., Drijber, R. A., & Lindquist, J. L. (2012). Optimizing cover crop benefits with diverse mixtures and an alternative termination method. *Agronomy Journal*, 104(5), 1425–1435. <https://doi.org/10.2134/agronj2012.0185>
- Zimnicki, T., Boring, T., Evenson, G., Kalcic, M., Karlen, D. L., Wilson, R. S., Zhang, Y., & Blesh, J. (2020). On Quantifying Water Quality Benefits of Healthy Soils. *BioScience*, 70(4), 343–352. <https://doi.org/10.1093/biosci/biaa011>