ABSTRACT

Title of Thesis:	NITROGEN CYCLING BY GRASS-BRASSICA MIXTURES IN THE MID-ATLANTIC
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Mixtures of cover crop species may be more effective than monocultures at internal nutrient cycling due to their ability to occupy different niches. Our study investigates nitrogen (N) cycling of radish (*Raphanus sativus* L.) and rye (*Secale cereal* L.) in monocultures and mixtures compared to a no cover crop control. The study was established on fine-textured soils near Laurel, MD where we estimated N leaching losses, quantified mineral soil N (to 60 cm), and cover crop biomass N for two years. Forage radish suppressed estimated N leaching in the fall, while cereal rye suppressed estimated N leaching in the spring. In this study, growing radish in a mixture with rye decreased the risk of N leaching losses and enhanced N cycling due to the difference in timing of N uptake and release. Our research indicates that grass-brassica mixtures are a flexible management tool for mitigating N leaching in the Mid-Atlantic.

NITROGEN CYCLING BY GRASS-BRASSICA MIXTURES IN THE MID-ATLANTIC

by

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Introduction

Nitrogen (N) loss from agricultural fields via leaching and runoff has an impact on the health of the Chesapeake Bay watershed (Kemp et al., 2005; Lassaletta et al., 2014). Nitrate-N losses from agriculture are highest during the fallow season, which generally extends from late fall through the following mid-spring (Dinnes et al., 2002). Cover crops planted in the fall and grown throughout the spring capture surplus N left on fields after the cash crop harvest (Meisinger et al., 1991; Dinnes et al., 2002; Tonitto et al., 2006; Quemada et al., 2013; Chatterjee and Clay, 2017; Tully and Ryals, 2017; Thapa et al., 2018a). In addition to reductions in NO_3^--N leaching, cover crops provide other agroecosystem services such as weed suppression, soil carbon sequestration, reduction in runoff and erosion, alleviation of soil compaction, improved microbial properties, and soil aggregate formation (Blanco-Canqui et al., 2015; Tully and Ryals 2017).

In addition to the benefits attained by management factors (e.g. planting date, termination timing, method of incorporation, etc.), the degree to which cover crops provide agroecosystem services may depend on the cover crop species planted (Blanco-Canqui et al., 2015; Chatterjee and Clay, 2017; Thapa et al., 2018a, b). Specifically, the variation in root diameter may explain a large amount of the variation in root traits among plant species (Ma et al., 2018), and thus agroecosystem services provided by each cover crop species. For example, forage radish (*Raphanus sativus* L.) has tap roots that grow quickly, alleviate soil compaction, and rapidly scavenge residual soil N that may reduce NO_3^--N leaching losses in the fall (Dean and Weil 2009; White and Weil 2011). Forage radish is able to reach soil depths up to 1 m in 43 days, whereas cereal rye (*Secale cereal* L.) takes 69 days to reach the same soil depth (Kristensen and Thorup-Kristensen, 2004). The small diameter portions (fine roots) of the forage radish's tap root that exceed 50 cm soil depth can penetrate up to 2 m into the soil column (Chen and Weil, 2009). In addition to the quick establishment of forage radish, another benefit is that it winter-kills when exposed to air temperatures below -4 °C for several days (Lawley et al., 2012). Winter-kill reduces the labor, use and cost of agrochemicals, and fuel needed for terminating cover crops (Weil and Kremen, 2007). However, there is a potential risk of NO_3^--N leaching via the tap-root channel after forage radish winter-kills and the ground thaws (Dean and Weil, 2009).

Grasses such as cereal rye (*Secale cereal* L.) and triticale (*Triticosecale* Witt.) grow slowly in the fall, but are winter-hardy, and are effective at scavenging residual soil N and suppressing NO_3^--N leaching from winter to spring (Meisinger et al., 1991; Thapa et al., 2018a). Compared to forage radish, cereal rye has a thinner, more fibrous root structure, which allows it to widely explore the upper portions of the soil profile (Kristensen and Thorup-Kristensen, 2004; Chen and Weil, 2009). Additionally, cereal rye's fibrous roots can increase soil organic matter (SOM) and improve the water-holding capacity of the surface soil (Blanco-Canqui et al., 2015; Chatterjee and Clay, 2017). Thus, the mixture of forage radish and cereal rye may optimize agroecosystem services provisioning due to the combined benefits of both species (Blanco-Canqui et al., 2015; Thapa et al., 2018b).

Measuring and modeling N leaching losses

Accurately estimating NO_3^--N leaching losses from cover crop mixtures requires data on (1) soil solution chemistry and (2) drainage past a defined lower boundary in the soil. To date, researchers have adopted several methods to quantify NO_3^--N leaching: (1) installation of porous cup lysimeters (also called tension or ceramic cup lysimeters); (2) drainage lysimeters (also known as monoliths); (3) subsurface drains or pans; or (4) periodic soil collection and extraction of mineral soil N (Lamba et al., 2013; Vanek and Drinkwater, 2013; Meisinger and Ricigliano, 2017; Russo et al., 2017). Among these methods, drainage lysimeters and subsurface drains or pans are labor and cost-intensive (Lamba et al., 2013), and require a large sampling area to obtain a field representative sample (Netto, et al., 1999; Pampolino et al., 2000).

Soil NO_3^--N concentrations (from extractions) may provide an indirect estimate of NO_3^--N leaching and require multiple collections, but are sensitive to changes in soil texture and precipitation. Porous cup lysimeters can be easily installed with a soil probe, sampled daily, and minimize soil disturbance compared to other instruments (Russo et al., 2017; Tully and Weil, 2014). Because porous cup lysimeters minimally disturb the soils, they are useful for conducting research on farms or small research plots. However, porous cup lysimeters may only sample macropore flow and overestimate soil solution concentrations rather than capturing diluted solution passing through micropore drainage pathways (van der Laan et al. 2010; Russo et al., 2017). Soil type and crop species both significantly change soil N dynamics through soil columns (Ajdary et al., 2007; Perego et al., 2012). In this study, we examined the effect of cover crop species on soil solution NO_3^--N concentrations using porous cup lysimeters.

Soil solution NO_3^--N concentrations and simulated drainage rates can be used to estimate NO_3^--N leaching in a soil hydraulic model (Ajdary et al., 2007; Perego et al., 2012; van der Laan et al., 2014; Russo et al., 2017). The VS2D (Lappala et al. 1987) and HYDRUS 1-D models (Šimůnek, et al., 2016) are public domain, hydraulic modeling programs for the analysis of water flow and solute transport through variably-saturated, porous media. Both of these programs numerically solve the Richards equation (Richards, 1931) or the Green Ampt equation (Green and Ampt, 1911). Soil hydraulic models provide an understanding of the complex interactions occurring between precipitation and nutrient application, crop root uptake, yield, erosion, runoff, and leaching (Ajdary et al. 2007). However, these models require calibration for specific soil textures, crop species, meteorological, and atmospheric conditions. We used a hydraulic model, HYDRUS 1-D, which was specifically calibrated for our medium textured soils to estimate drainage past 60 cm soil depth. HYDRUS 1-D simulated drainage and soil solution NO_3^--N from porous cup lysimeters were used to estimate the NO_3^--N leaching losses from the different cover crop species combinations.

The objective of our study was to determine if grass-brassica mixtures are more effective at reducing NO_3^--N leaching in both the fall and the spring as compared to their monoculture counterparts. We tested our hypothesis by estimating NO_3^--N leaching losses over the course of two growing seasons in a radish+rye mixture, monocultures, and no cover crop control.

Methods

Study sites

This study was conducted near Laurel, Maryland at the University of Maryland Central Maryland Research and Education Center (UMD CMREC; 39°01'40" N 76°50'14" W) from 2016-2018. Soils are Russett-Christiana complex where Russetts are classified as loam or sandy loam in the surface soils and Christiana surface soils are classified as silt loams (Table 1.1 and 1.2). In 1988, the land was converted from pasture into research plots under no-till management. The bulk density of the soils ranges from 1.3-1.8 g cm^{-3} depending on the soil texture and depth.

The site receives an average of 1046 mm annual rainfall, has a mean minimum temperature of 6.8° C, and a mean maximum temperature of 19° C (USDA-ARS, 2018). Our sites received 377 mm of rainfall from the first soil solution collection date to the last collection date in year 1 (3 Jan 2016-27 May 2017) and 577 mm in year 2 (31 Oct 2017-3 Jun 2018).

Experimental design

Experimental plots were established in a randomized complete block design with four cover crop treatments (1) forage radish (*Raphanus sativus*), (2) cereal rye (*Secale cereal*), (3) forage radish-cereal rye mixture, and (4) no cover crop control (weeds only, *Lamium amplexicaule* L. and *Cerastium arvense* L.). Cover crops received 140 kg-N ha^{-1} before planting to mimic the high residual soil N left after a poor cash crop harvest. Cover crops were planted at 17.8 cm (7 in) spacing. Plots were (3.1 x 10.7 m), with a 0.31 m buffer between each plot using a no-till grain drill. Cereal rye was seeded at 126 kg ha^{-1} and forage radish at 19.8 kg ha^{-1} in both years. The radish+rye mixture was seeded at 2.8/63 kg ha^{-1} (radish/rye) in year 1 and at 4.5/84 kg ha^{-1} (radish/rye) in year 2. Forage radish was winter-killed in mid-December - late January and cereal rye was terminated using glyphosate (N-(phosphonomethyl)-glycine) in early June of both study years (Table 1.3).

Porous cup lysimeters (22 mm diameter; Soil Solution Access Tubes, Irrometer, Riverside, California, USA) were installed at 60 cm below the ground surface following planting (Table 1.3). Four lysimeters were installed in each plot (two in cover crop rows and two in between the rows) to account for spatial variability in leaching within the plots (total of 32 and 64 lysimeters installed in year 1 and 2, respectively; Table 1.3). Lysimeters were installed with a 23.8 mm diameter soil probe and a slurry using the deepest soil was poured into the hole before inserting the lysimeter to ensure good soil contact. Finally, lysimeters were sealed at the soil surface with a bentonite/sand mixture to avoid preferential flow of water down the sides of the tube. Pilot studies confirmed that soil solution collection was only possible following rain events that were greater than or equal to 6 mm, thus soil solution was only collected following rain events of this level. Each day before sampling, lysimeters were purged of any water and an internal pressure of -60 to -70 kPa was applied. Soil solutions were collected, filtered (Whatman No. 42; 2.5 μm), and stored in a freezer at -20°C until further analysis. The soil solutions were analyzed on a LACHAT QuikChem 8500 (HACH Co., Loveland, CO) for ammonium (NH_4^+-N) and NO_3^--N using the sodium salicylate (US EPA, 1983) and cadmium reduction (Fisherman and Friedman, 1989) methods, respectively. Samples analyzed for NH_4^+-N were either at or below the method detection limit, while samples analyzed for NO_3^--N were diluted if they exceeded the highest calibration standard within the detectable range of the colorimeter.

Soil volumetric water content and temperature were collected continuously (every 10 minutes) throughout the cover crop growing season using time-domain reflectometers (TDRs; True TDR-310S, Acclima, Meridian, Idaho, USA). During the first study year (2016-2017), TDRs were installed at three depths: 0-10, 20-30, and 50-60 cm on 14 Feb 2017 and removed on 24 May 2017 in two of the four blocks (total of 18 TDRs). We found little moisture variability at 50-60 cm, so in the second study year (2017-2018), TDRs were only installed at 0-10 and 10-20 cm on 28 Oct 2017 and removed on 5 Jun 2018 (total of 24 TDRs) in all four of the blocks. During both study years, the TDRs were installed in only the radish, rye, and the no cover crop control plots. The TDRs were excluded from the radish+rye plots since HYDRUS 1-D lacks the functionality to model mixtures (only monocultures). We will discuss how we compensate for this shortcoming below.

Estimating N leaching losses

Real-time soil volumetric water content data were used to calibrate the estimated soil volumetric water contents simulated in HYDRUS 1-D version 4.17.0140 (Šimůnek, et al., 2016). A linear interpolation on soil solution NO_3^--N concentration was used to obtain daily concentrations for each block and cover crop treatment. The interpolated soil solution NO_3^--N concentrations were multiplied by the estimated daily drainage at the block-level and scaled to kg ha^{-1} to estimate NO_3^--N leaching losses from each plot. There was no option for a two or more species mixture in HYDRUS1-D. Therefore, we used the estimated drainage under rye for the radish+rye mixture since rye was the only crop that persisted for the entire duration of the cover crop growing season within the mixture.

We used a public domain, finite element water movement and solute transport model, HYDRUS 1-D, for analysis of one-dimensional water flow (Šimůnek, et al., 2016). The soil profile geometry for our model consisted of four soil materials at 0-15, 15-30, 30-45, and 45-60 cm. To simulate one-dimensional, vertical fluid flow, the upper boundary conditions were defined as atmospheric with surface layer in HYDRUS 1-D (measured precipitation). The lower boundary was characterized as free drainage to simulate gravity-driven flow, and the root water uptake parameter was used to simulate transpiration (Table 1.4). Runoff was estimated using the National Resource Conservation Service curve method for each cover crop treatment using the hydrologic soil group, D (poor drainage; USDA, NRCS, 1986). The curve number (CN) for rye was 89 and 94 for the no cover crop control for the entire duration of both cover crop growing season. For radish, the CN was 89 before the winter kill event and 94 after for each study year (Table 1.3). Runoff percentages were calculated based on daily precipitation and applied to hourly rainfall for rain events that exceed the hourly infiltration rate of the soils, which is calculated as:

$$(1000/CN - 10) * 0.2 \tag{1}$$

Surface soil boundary and meteorological conditions were determined using 15-minute continuous precipitation, solar radiation, temperature, relative humidity, and wind speed data. The data came from a nearby (1.2 km) meteorological tower (USDA-ARS, 2019), and were adjusted to hourly values. We used the van Genuchten equation (van Genuchten, 1980) to calculate the water retention curve for our single porosity, hydraulic model. In year 1, the hydraulic model began on 3 Jan 2017 (first soil solution collection date), and the modeled volumetric water contents were calibrated using 10 weeks (20 Mar-30 May 2017) of measured, continuous (every 10 minutes) volumetric water contents (VWC) from 0-10 and 20-30 cm from two of the four blocks (Figure 1.1). In year 2, the hydraulic model began on 31 Oct 2017 (first soil solution collection date), and the modeled volumetric water contents were calibrated to fit 18 weeks (31 Oct-21 Dec and 21 Mar-5 Jun) of measured, continuous VWC at 0-10 and

10-20 cm from all four blocks (Figure 1.2). The measured, continuous VWC were averaged over one hour for both years. The periods when soil temperatures were below freezing (0° C) were eliminated due to inaccurate soil moisture readings.

Theta saturation (θ_s), theta residual (θ_r), alpha (α), and *n* were adjusted to fit the modeled to the measured volumetric water contents at the block-level (Table 1.5 and 1.6). Theta saturation is the soil water content near saturation of the pores with water, θ_r is the water content below which liquid water movement is minimal, and n and α (alpha) determine the slope of the water retention curve at various soil water potentials. Another important parameter for calibration is the relative permeability (K_{sat}), which we determined using data from the Web Soil Survey for each of the four soil materials by soil depth at our sites (Soil Survey Staff, NRCS, USDA). Crop height, rooting depth, and solar radiation were used to estimate evapotranspiration (ET) of rye and radish in HYDRUS 1-D. Rye and radish crop heights were estimated based on monthly field observations. The growth rate for radish roots and shoots increased linearly until the winter-kill event, whereas the rye increased linearly during the fall and plateaued in the winter due to dormancy. Growth then recommenced after the ground thawed in the early spring reaching a maximum height of 114 cm. The maximum rooting depth in the model was set to 60 cm for both radish and rye due to soil compaction of 1.7 g cm^{-3} at this depth.

An estimated water balance was created from data simulated in HYDRUS 1-D to ensure the accuracy of the models (Table 1.7). There was 0.2-3.9% error in the measured rainfall and the rainfall output from the model. The evaporation, transpiration, and drainage were graphed over time for each study year to ensure there were no over- or under-estimation of these data (Figure 1.3; Figure 1.4; Figure 1.5).

Statistical approach

To examine the effect of cover crop species on soil solution NO_3^--N concentrations, we used a linear mixed effect model (*lmer* package for R; Bates et al., 2013) with cover crop treatment and collection date as the main effects and block as a random effect. Seasons were defined using the equinox and solstice for fall (23 Sep-20 Dec), winter (21 Dec-20 Mar), and spring (21 Mar-21 June). To examine the effect of cover crop species on soil solution NO_3^--N concentrations by season, we used another linear mixed effect model with cover crop treatment and season as the main effects and block as a random effect. Differences were considered significant at $P < 0.05$. If necessary, concentration data were transformed using a Box-Cox transformation to satisfy the assumptions of the statistical models (Box and Cox, 1964). Tukey *post hoc* tests were used to examine pairwise comparisons among cover crop treatments for individual rain events and seasons (*multcomp*; Hothorn et al., 2008). All data were analyzed in the R environment for PC (R Core Team 2018).

Linear models were used to test the effect of cover crop treatment on estimated NO_3^--N leaching losses over the course of each study year and by season. In year 1, the lysimeters were only installed in two blocks, which caused extremely high variability and a lack of statistical power for any discernible statistical differences between cover crop treatments. In year 2, the lysimeters were installed in every block (total of four), which increased the statistical power.

<u>*Results*</u>

Cover crops altered evapotranspiration and drainage

In year 1, we determined that radish winter-killed between 9-11 Dec 2016, when air temperatures dipped below -4° C. In year 2, we determined that radish winter-killed between 5-7 Jan 2018 when soil temperatures dipped below -4° C. Our models of transpiration and evaporation matched expected patterns based on cover crop growth patterns and ranged from 0-40 mm across the study period. For example, the rye treatment had high transpiration in the winter through the spring since there was a crop present for the entire duration of each modeled growing season (Figure 1.3). On the other hand, transpiration was highest in the fall in radish treatment until the winter-kill event (Figure 1.3). Modeled evaporation showed the exact opposite patterns of transpiration for both radish and rye (e.g. low evaporation in the fall and high evaporation in the spring under radish; Figure 1.4). The model estimated that no transpiration occurred in the no cover crop control plots, but rather all water was lost in the form of evaporation (Figure 1.3 and 1.4). However, we suspect that some transpiration occurred from the small weed population in those treatments.

Cover crops also altered estimated runoff, which played a more important role in moderating water loss than evapotranspiration. The cumulative evapotranspiration ranged from 4.7-14.5% of the total precipitation, while cumulative runoff ranged from 35.2-53.5% of the total precipitation over both study years. The highest cumulative runoff was found in the control for both study years since there was no cover crop to suppress the amount of runoff (Table 1.7).

Finally, we found that higher precipitation rate in year 2 than year 1 (by 200 mm) led to higher cumulative drainage in year 2 than year 1, regardless of species (Table 1.8 and Figure 1.6). Our model illustrated that less runoff and more drainage occurs with a longer duration of cover crop presence (Figure 1.6). For example, rye and radish+rye had the highest drainage and least amount of runoff, followed by radish, and lastly the no cover crop control with the least drainage and highest runoff (Table 1.8).

Soil solution NO_3^--N concentrations through time

In year 1 (2016-2017), soil solution NO_3^--N concentrations varied dramatically throughout the year in all cover crop treatments (effect of season*cover crop species: $P<0.001$) ranging from 0 to 90 mg NO_3^--N L^{-1}. Overall, concentrations under rye and radish+rye were significantly lower than under radish and no cover crop control ($P< 0.001$; Table 1.9). In year 1, soil solution was only collected in the winter and spring due to minimal precipitation and drought conditions in the fall (Table 1.9). In the winter 2017, soil solution NO_3^--N concentrations were significantly lower under rye and radish+rye than radish and the no cover crop control ($P<0.001$; Table 1.9). There were significantly lower soil solution NO_3^--N concentrations under the no cover crop control than radish in the winter ($P<0.001$; Table 1.9). In the spring of 2017, soil solution NO_3^--N concentrations under radish+rye and rye were significantly lower than the no cover crop control ($P<0.001$; Table 1.9). There were no significant differences in soil solution NO_3^--N concentrations between the radish and the other cover crop treatments in the spring ($P<0.002$; Table 1.9).

In year 2 (2017-2018), soil solution NO_3^--N concentrations were significantly lower under radish, rye, and radish+rye than the no cover crop control ($P<0.001$; Table 1.9). In fall 2017, soil solution NO_3^--N concentrations were significantly lower under radish than under rye and the no cover crop control ($P<0.05$), but not radish+rye (Table 1.9). Soil solution NO_3^--N concentrations were similar between rye and the no cover crop control in the fall (Table 1.9). In the winter and spring, soil solution NO_3^--N concentrations were lower under radish, rye, and radish+rye than the no cover crop control ($P<0.001$; Table 1.9).

Estimated N leaching losses

In year 1, estimated NO_3^--N leaching losses followed the pattern: radish > no cover crop control > radish+rye > rye (Table 1.10), but due to a lack of statistical power, there was no significant differences detected among the treatments. Estimated NO_3^--N leaching losses under rye and radish+rye were 83 and 75% lower (respectively) than under the no cover crop control, while estimated NO_3^--N leaching losses were 4% lower under the no cover crop control than radish. The estimated NO_3^--N leaching losses from the radish and no cover crop control treatments primarily occurred in the winter and spring, while most of the NO_3^--N leaching loss under radish+rye and rye occurred in the winter (Figure 1.7).

In year 2, estimated NO_3^--N leaching losses were significantly lower under radish, rye, and radish+rye than the no cover crop control ($P<0.001$; Table 1.10; Figure 1.7). Estimated NO_3^--N leaching losses were lower under radish, rye, and radish+rye (by 84%, 82%, and 87%, respectively) than the no cover crop control. The majority of the estimated NO_3^--N leaching losses under rye and radish+rye occurred in the fall, while they were consistently low from the fall through the spring under radish (Table 1.10; Figure 1.7).

<u>Discussion</u>

Cover crop altered soil solution NO_3^--N concentrations through time

In the fall of 2016, exceptional drought conditions prevented soil solution collections (NIDIS). In fall 2017, soil solution NO_3^--N concentrations were lower under radish than the no cover crop control and rye, while radish was similar to radish+rye due quick establishment of radish in both the mixture and monoculture. Mean soil solution NO_3^--N concentrations were much higher under radish in year 1 (20.0 mg L^{-1}) than year 2 (3.5 mg L^{-1}) due to differences in planting date between the two years. In year 1, the radish had 52 days to establish, whereas in year 2, the radish had 107 days to establish, leading to lower biomass accumulation in year 1 (0.6 Mg ha^{-1}) than year 2 (3.0 Mg ha^{-1}; Gaimaro et al *in prep*). A similar pattern was found in a global meta-analysis, which showed that NO_3^--N leaching losses are significantly lower than a no cover control if cover crops can accumulate roughly 2 Mg ha^{-1} of aboveground biomass (Thapa et al. 2018a).

Unlike radish, planting later in the fall did not impact the rye's ability to reduce the soil solution NO_3^--N concentrations (in either mixture or monoculture) since most of rye's growth occurs from late winter to spring. Soil solution NO_3^--N concentrations in the radish+rye mixture followed similar patterns to the rye monoculture. However, in the fall of 2017 (year 2), soil solution NO_3^--N concentrations under rye were similar to the no cover crop control, while they were lower under radish+rye than the no cover crop control; this is because the radish was accumulating N in its tissues while the rye was relatively inactive (Table 1.9). Further, the radish+rye mixtures had low soil solution NO_3^--N concentrations (0.0-9.4 mg NO_3^--N L^{-1}; Tables 1.8) in both years despite the different radish seeding rates (year 1: 2.8 kg ha^{-1} radish seed; year 2: 4.5 kg ha^{-1} radish seed) and planting densities (year 1: 1 row rye to 1 row radish; year 2: 2 rows rye to 1 row radish). The radish+rye mixture was able to suppress soil solution NO_3^--N concentrations even with the variation in planting date and seeding rates. Thus, proving the radish+rye mixture to be a flexible tool for managing soil solution NO_3^--N concentrations in mid-Atlantic cropping systems.

Cover crops altered evapotranspiration and drainage

Cover crops not only exercise control over the amount of N susceptible to leaching, but also the amount of water moving through the soil column (Brill and Neal, 1950; Dabney, 1998; Kaspar et al., 2001). For instance, cover crops can lower drainage rates by increasing transpiration or raise drainage rates by decreasing evaporation and runoff. We modeled water loss to transpiration during periods of biomass growth and water loss to evaporation during periods when the soil was bare (Figure 1.3 and 1.4). Overall, cover crop moderation of runoff played a more important role than evaporation or transpiration on cumulative drainage from the cover crop treatments (Table 1.7). This is because these soils belong to a very poor drainage class (Hydrologic Soil Group, D) with a high average bulk density of 1.6 g cm^{-3}. In the presence of cover crops, there was a decrease in runoff with a corresponding increase in drainage.

Cover crops reduce estimated leaching compared to no cover crop control

We found that cover crops reduced soil solution NO_3^--N concentrations and estimated NO_3^--N leaching losses over the course of a cover crop growing season compared to a no cover crop control. Most of the studies investigating NO_3^--N leaching losses of cover crops include monocultures (e.g. brassicas, grasses, and legumes) or grass-legume mixtures. There are no studies to date that investigate the efficacy of grass-brassica mixtures in reducing NO_3^--N leaching losses compared to their monoculture counterparts or a no cover crop control. Grass-legume mixtures grown on similar soils had higher NO_3^--N leaching losses due to biological N fixation in the legumes (~56 kg-N ha^{-1}; Campigila et al., 2010; Faega et al., 2010; Totsi, et al., 2014) compared to the grass-brassica mixture in this study (7.3 kg-N ha^{-1}).

Constantin et al. (2010) examined NO_3^--N leaching losses under rye, radish, and no cover crop control for 14 cover crop growing seasons and found that NO_3^--N leaching losses were smaller under radish (11-15 kg-N ha^{-1}) than rye

(39 kg-N ha^{-1}), however, they planted radish well before winter-kill in early August - late September. In year 1, we planted our cover crops in mid-October and found the opposite trend with smaller NO_3^--N leaching losses under rye (5.4 kg-N ha^{-1}) than radish (32.7 kg-N ha^{-1}). By planting radish earlier in year 2 (mid-September), estimated NO_3^--N leaching losses were much lower (8.4 kg-N ha^{-1}) compared to year 1 under radish (32.7 kg-N ha^{-1}; Tables 1.10). Of note, our fall N treatment was surface-applied rather than deeper N following a corn harvest, which may have led to an advantage for rye over radish (Kristensen and Thorup-Kristensen, 2004; Dean and Weil, 2009). We estimated NO_3^--N leaching losses of 5.4-9.6 kg-N ha^{-1} under rye, which was similar to other studies reporting around 13.6 kg-N ha^{-1} (Shepherd 1999; Macdonald et al. 2005; Daigh et al. 2015). On similar soils, NO_3^--N leaching losses ranged from 2-124 kg-N ha^{-1} under rye (Brandi-Dohrn et al., 1997; Faega et al., 2010; Meisinger and Ricigliano, 2017), suggesting that N leaching losses measured in our study are on the low end. However, there were differences in planting and termination dates (Beckwith et al., 1998; Kaspar et al., 2012; Heinrich et al., 2014), precipitation (Constantin et al., 2010; Faega et al., 2010), residual soil N levels, and farm management (Brandi-Dohrn et al., 1997), which could explain why our N leaching losses were on the lower end as compared to the other studies. For instance, rye can suppress NO_3^--N leaching losses more effectively during drier years (Meisinger and Ricigliano, 2017; Thapa et al. 2018a). This could explain why we observed an increase in estimated NO_3^--N leaching from 5.4 kg NO_3^--N ha^{-1} in year 1 to 9.6 kg NO_3^--N ha^{-1} in year 2 under the rye cover crop treatment since there was more cumulative drainage and

precipitation in year 2. Notably, this increase in cumulative drainage and precipitation from year 1 to year 2 did not impact the ability of the radish+rye mixture to reduce the estimated NO_3^--N leaching, which remained around 6.7-7.9 kg-N ha^{-1} per year. We show that a grass-brassica mixture may be less sensitive to variation in precipitation and drainage compared to a rye monoculture, and less sensitive to a later planting date than a radish monoculture, thus allowing them to more consistently reduce NO_3^--N leaching across a wide range of managements and climates.

This study may have been limited by the monoculture presets of HYDRUS 1-D for simulating the drainage under cover crop mixtures, porous cup lysimeters sampling macropore-dominated flow, and the estimated rooting depths. The drainage may have been overestimated in the fall due to the radish in the radish+rye mixture and underestimated in the winter and spring following the radish winter-kill. Additionally, the porous cup lysimeters may only sample macropore-dominated flow and lead to an overestimation of the soil solution NO_3^--N concentrations and estimated NO_3^--N leaching. The primary ways for determining rooting depth of plants can be laborious and destructive to small, no-till research plots (e.g. excavating) or expensive (e.g. minirhizotrons), so we did not measure the exact rooting depth of each cover crop species. This could have led to an under or overestimation of rooting depth, transpiration, and drainage. Future studies should determine methods for modeling crop mixtures, the roles of roots, and the efficacy of different radish+rye seeding rates and planting densities for suppressing NO_3^-N leaching.

Conclusion

Cover crop mixtures and monocultures reduced estimated NO_3^--N leaching losses compared to the no cover crop control. The ability of radish to suppress estimated NO_3^--N leaching largely depended on when it was planted and when it died. Monoculture radish planted early in the fall suppressed estimated NO_3^--N leaching losses during the fall and winter months. Rye was more effective than radish at reducing estimated leaching in the spring. Rye's ability to reduce estimated NO_3^--N leaching was largely a function of precipitation and drainage. The radish+rye mixture more consistently reduced estimated NO_3^--N leaching than either species planted in monoculture and shows flexible application across a range of farm managements and climates across the mid-Atlantic region.

Table 1.1. Soil textural properties by block and depth for plots in year 1 of the cover crop mixture experiment.

Block	Treatment	Depth (cm)	% Sand	% Silt	% Clay	Texture
Block 1	No cover crop control	0-15	33	52	15	Silt loam
		15-30	25	51	24	Silt loam
		30-45	16	38	46	Clay
		45-60	34	26	41	Clay
	Radish	0-15	33	52	15	Silt loam
		15-30	25	51	24	Silt loam
		30-45	16	38	46	Clay
		45-60	34	26	41	Clay
	Rye and Radish+Rye	0-15	33	52	15	Silt loam
		15-30	25	51	24	Silt loam
		30-45	16	38	46	Clay
		45-60	34	26	41	Clay
Block 4	No cover crop control	0-15	58	35	7	Sandy loam
		15-30	57	29	14	Sandy loam
		30-45	46	35	19	Loam
		45-60	34	40	26	Loam
	Radish	0-15	58	35	7	Sandy loam
		15-30	57	29	14	Sandy loam
		30-45	46	35	19	Loam
		45-60	34	40	26	Loam
	Rye and Radish+Rye	0-15	58	35	7	Sandy loam
		15-30	57	29	14	Sandy loam
		30-45	46	35	19	Loam
		45-60	34	40	26	Loam

Table 1.2. Soil textural properties by plot and depth for plots in year 2 of the cover crop mixture experiment.

Block	Treatment	Depth (cm)	% Sand	% Silt	% Clay	Texture
Block 1	No cover crop control	0-15	42	41	17	Loam
		15-30	31	42	27	Clay loam
		30-45	27	41	32	Clay loam
		45-60	34	35	31	Clay loam
	Radish	0-15	42	40	18	Loam
		15-30	39	39	22	Loam
		30-45	33	41	26	Loam
		45-60	35	37	29	Clay loam
	Rye and Radish+Rye	0-15	50	35	16	Loam
		15-30	32	42	27	Clay loam
		30-45	20	41	39	Silty clay loam
		45-60	22	33	45	Clay
Block 2	No cover crop control	0-15	51	34	15	Loam
		15-30	40	37	24	Loam
		30-45	37	32	31	Clay loam
		45-60	32	30	38	Clay loam
	Radish	0-15	52	32	16	Loam
		15-30	39	35	25	Loam
		30-45	26	34	40	Loam
		45-60	21	31	48	Clay
	Rye and Radish+Rye	0-15	52	32	16	Loam
		15-30	39	35	25	Loam
		30-45	26	34	40	Loam
		45-60	21	31	48	Clay
Block 3	No cover crop control	0-15	63	24	13	Sandy loam
		15-30	46	32	22	Loam
		30-45	32	37	32	Clay loam
		45-60	29	35	36	Clay loam
	Radish	0-15	60	27	14	Sandy loam
		15-30	38	36	26	Loam
		30-45	27	38	35	Clay loam
		45-60	25	40	35	Clay loam
	Rye and Radish+Rye	0-15	64	24	12	Sandy loam
		15-30	56	26	18	Sandy loam
		30-45	51	30	19	Loam
		45-60	50	29	22	Loam
Block 4	No cover crop control	0-15	43	40	17	Loam
		15-30	29	48	23	Loam
		30-45	25	42	32	Clay loam
		45-60	22	42	35	Clay loam
	Radish	0-15	43	41	17	Loam
		15-30	33	43	24	Loam
		30-45	21	45	34	Clay loam
		45-60	26	39	35	Clay loam
	Rye and Radish+Rye	0-15	48	38	14	Loam
		15-30	48	34	18	Loam
		30-45	48	31	21	Loam
		45-60	48	30	22	Loam

Table 1.3. Field activities by study year including timing of planting, installation of equipment, and cover crop termination.

Year 1 (2016–17)		Year 2 (2017–18)	
16 Oct 2016	Fertilize, then plant cover crops	*22 Sep 2017*	Fertilize, then plant cover crops
24 Oct 2016	Install lysimeters	*28 Sep 2017*	Install lysimeters
Mid Dec 2016	Radish winter-kill event	*28 Sep 2017*	Install Time Domain Reflectometers
14 Feb 2017	Install Time Domain Reflectometers	*Mid Jan 2017*	Radish winter-kill event
5 Jun 2017	Terminate cover crops	*10 Jun 2018*	Terminate cover crops

Table 1.4. Parameters selected for development of the HYDRUS model. Parameters were used for every block in the experimental design with the exception of the no cover crop control plots when indicated. The soil hydraulic parameters were subjected to calibration by block.

Pre- Processing	Criteria
Main Processes	Water flow and root water uptake (except no cover crop control)
Geometry Information	4 soil materials with a depth of 60 cm
Time Information	Hourly
Soil Hydraulic Properties	van Genuchten equation (van Genuchten, 1980)
Water Flow Boundary Conditions	Upper: Atmospheric BC with Surface Layer
	Lower: Free drainage
Meterological Parameters	Net Radiation with crop (except no cover crop control)

Table 1.5. Soil hydraulic parameters for modeled volumetric water content calibration by species for year 1. Relative permeability (K_{sat}) was determined by depth through Web Soil Survey (Soil Survey Staff, NRCS, USDA). Theta saturation (θ_s), theta residual (θ_r), alpha (α), and *n* were adjusted to fit the modeled to the measured volumetric water contents at the block level. Theta saturation is the soil water content near saturation of the pores with water, theta residual is the water content below which liquid water movement in minimal, n and α (alpha) determine the slope of the water retention curve at various soil water potentials. Theta saturation and theta residual are reported as volumetric water contents (cm^3 cm^{-3}).

Block	**Treatment**	**Depth**	**K_{sat}**	**Θ_r**	**Θ_s**	**α**	***n***
		cm	*cm hr^{-1}*				
Block 1	No cover crop control	0-15	3.00	0.07	0.41	0.075	1.890
		15-30	0.80	0.07	0.31	0.008	1.090
		30-45	0.80	0.07	0.31	0.008	1.090
		45-60	0.80	0.07	0.31	0.008	1.090
	Radish	0-15	3.00	0.07	0.50	0.065	1.890
		15-30	0.80	0.07	0.34	0.012	1.190
		30-45	0.80	0.07	0.34	0.008	1.090
		45-60	0.80	0.07	0.34	0.008	1.090
	Rye and Radish+Rye	0-15	3.00	0.07	0.35	0.020	1.410
		15-30	0.80	0.07	0.31	0.008	1.090
		30-45	0.80	0.07	0.31	0.008	1.090
		45-60	0.80	0.07	0.31	0.008	1.090
Block 4	No cover crop control	0-15	3.00	0.07	0.35	0.020	1.560
		15-30	0.80	0.07	0.29	0.008	1.090
		30-45	0.80	0.07	0.29	0.008	1.090
		45-60	0.80	0.07	0.29	0.008	1.090
	Radish	0-15	3.00	0.07	0.31	0.015	1.650
		15-30	0.80	0.07	0.26	0.011	1.100
		30-45	0.80	0.07	0.26	0.011	1.100
		45-60	0.80	0.07	0.26	0.011	1.100
	Rye and Radish+Rye	0-15	3.00	0.07	0.36	0.015	1.650
		15-30	0.80	0.07	0.26	0.011	1.150
		30-45	0.80	0.07	0.26	0.011	1.150
		45-60	0.80	0.07	0.26	0.011	1.150

Table 1.6. Soil hydraulic parameters for modeled volumetric water content calibration by species for year 2. Relative permeability (K_{sat}) was determined by depth through Web Soil Survey (Soil Survey Staff, NRCS, USDA). Theta saturation (θ_s), theta residual (θ_r), alpha (α), and *n* were adjusted to fit the modeled to the measured volumetric water contents at the block level. Theta saturation is the soil water content near saturation of the pores with water, theta residual is the water content below which liquid water movement in minimal, n and α (alpha) determine the slope of the water retention curve at various soil water potentials. Theta saturation and theta residual are reported as volumetric water contents ($cm^3\ cm^{-3}$).

Block	Treatment	Depth	K_{sat}	Θ_r	Θ_s	α	*n*
		cm	*cm hr⁻¹*				
Block 1	No cover crop control	0-15	3.00	0.08	0.43	0.036	1.560
		15-30	0.80	0.07	0.29	0.036	1.560
		30-45	0.80	0.07	0.29	0.036	1.560
		45-60	0.80	0.07	0.29	0.036	1.560
	Radish	0-15	3.00	0.07	0.50	0.036	1.560
		15-30	0.80	0.07	0.40	0.036	1.560
		30-45	0.80	0.07	0.40	0.036	1.560
		45-60	0.80	0.07	0.40	0.019	1.310
	Rye and Radish+Rye	0-15	3.00	0.07	0.43	0.020	1.890
		15-30	0.80	0.08	0.30	0.019	1.310
		30-45	0.80	0.09	0.43	0.010	1.230
		45-60	0.80	0.07	0.38	0.008	1.090
Block 2	No cover crop control	0-15	3.00	0.08	0.43	0.036	1.560
		15-30	0.80	0.08	0.43	0.036	1.560
		30-45	0.80	0.10	0.41	0.019	1.310
		45-60	0.80	0.10	0.41	0.019	1.310
	Radish	0-15	3.00	0.07	0.55	0.030	1.890
		15-30	0.80	0.07	0.38	0.030	1.890
		30-45	0.80	0.07	0.38	0.030	1.890
		45-60	0.80	0.07	0.38	0.019	1.310
	Rye and Radish+Rye	0-15	3.00	0.07	0.43	0.036	1.890
		15-30	0.80	0.07	0.38	0.030	1.890
		30-45	0.80	0.07	0.38	0.030	1.890
		45-60	0.80	0.07	0.38	0.019	1.310
Block 3	No cover crop control	0-15	3.00	0.08	0.45	0.036	1.860
		15-30	0.80	0.08	0.43	0.036	1.560
		30-45	0.80	0.10	0.41	0.019	1.310
		45-60	0.80	0.10	0.41	0.019	1.310
	Radish	0-15	3.00	0.07	0.41	0.075	1.890
		15-30	0.80	0.08	0.43	0.036	1.560
		30-45	0.80	0.10	0.41	0.019	1.310
		45-60	0.80	0.10	0.41	0.019	1.310
	Rye and Radish+Rye	0-15	3.00	0.08	0.43	0.036	1.560
		15-30	0.80	0.08	0.43	0.036	1.560
		30-45	0.80	0.10	0.41	0.019	1.310
		45-60	0.80	0.10	0.41	0.019	1.310
Block 4	No cover crop control	0-15	3.00	0.08	0.43	0.036	1.560
		15-30	0.80	0.08	0.43	0.036	1.560
		30-45	0.80	0.10	0.41	0.019	1.310
		45-60	0.80	0.10	0.41	0.019	1.310
	Radish	0-15	3.00	0.08	0.43	0.036	1.560
		15-30	0.80	0.08	0.43	0.036	1.560
		30-45	0.80	0.10	0.41	0.019	1.310
		45-60	0.80	0.10	0.41	0.019	1.310
	Rye and Radish+Rye	0-15	3.00	0.08	0.43	0.042	1.890
		15-30	0.80	0.08	0.41	0.036	1.560
		30-45	0.80	0.08	0.43	0.036	1.560
		45-60	0.80	0.08	0.43	0.036	1.560

Table 1.7. Estimated water balance for each of the model cover crop treatments. Rye was used for the radish+rye since HYDRUS does not enable the modeling of crop mixtures. Runoff was determined by the NRCS curve method (USDA, NRCS, 1986), while drainage and evapotranspiration (ET) were simulated by HYDRUS 1-D. The column total indicates the summation of runoff, drainage, and ET. Rainfall is the total precipitation between the indicated dates for each year as measured by a tipping-bucket rain gauge located 1.2 km from the plots to determine the total amount of rainfall accounted for in the simulated data.

Year 1 (3 Jan 2017 – 27 May 2017)					
Treatment	**Estimated Runoff**	**Estimated Drainage**	**Estimated ET**	**Estimated Total**	**Measured Rainfall**
			cm		
Control	18.15	16.56	2.95	37.66	37.74
Radish	18.15	16.44	2.95	37.54	37.74
Rye	13.28	19.07	5.48	37.84	37.74
Year 2 (31 Oct 2017 – 3 Jun 2018)					
Treatment	**Estimated Runoff**	**Estimated Drainage**	**Estimated ET**	**Estimated Total**	**Measured Rainfall**
			cm		
Control	30.91	22.41	2.70	56.03	57.73
Radish	29.70	24.59	2.98	57.28	57.73
Rye	23.66	28.24	3.55	55.45	57.73

Table 1.8. Estimated drainage past 60 cm soil depth for each season and total drainage by cover crop treatment. In the fall 2016, the drought conditions and minimal precipitation prevented soil solution collection, therefore values are indicated as NA. Standard errors are in parenthesis.

	Year 1 (3 Jan 2017 – 27 May 2017)			
	Fall-Winter†	**Winter-Spring‡**	**Spring-Summer§**	**Total**
		cm		
Control	NA	6.9 (0.07)	9.7 (0.08)	16.6 (0.00)
Radish	NA	7.0 (0.04)	9.7 (0.17)	16.7 (0.21)
Rye	NA	7.8 (0.02)	11.2 (0.08)	19.0 (0.10)
Radish+Rye	NA	7.8 (0.02)	11.2 (0.08)	19.0 (0.10)
	Year 2 (31 Oct 2017 – 3 Jun 2018)			
	Fall-Winter¶	**Winter-Spring#**	**Spring-Summer††**	**Total**
		cm		
Control	4.2 (0.21)	7.9 (0.06)	11.2 (0.09)	23.3 (0.08)
Radish	6.4 (0.02)	8.0 (0.04)	11.0 (0.07)	25.3 (0.10)
Rye	5.3 (0.27)	9.6 (0.00)	14.7 (0.11)	29.6 (0.2)
Radish+Rye	5.3 (0.27)	9.6 (0.00)	14.7 (0.11)	29.6 (0.2)

† Fall-Winter = 1 Oct 2016 – 20 Dec 2016
‡ Winter-Spring = 21 Dec 2016 - 20 Mar 2017
§ Spring-Summer = 21 Mar 2017 - 5 Jun 2017
¶ Fall-Winter = 1 Oct 2017 – 20 Dec 2017
# Winter-Spring = 21 Dec 2017 - 20 Mar 20178
†† Spring-Summer = 21 Mar 2018 - 5 Jun 2018

Table 1.9. Mean NO_3^--N concentrations (mg L^{-1}) in soil solution (at 60 cm) by season and overall means by cover crop species. In the fall 2016, the drought conditions and minimal precipitation prevented soil solution collection, therefore values are indicated as NA. Standard errors are in parenthesis and values with different letters are significantly different at P<0.05.

	Year 1 (3 Jan 2017 – 27 May 2017)			
	Fall-Winter†	**Winter-Spring‡**	**Spring-Summer§**	**Mean**
		mg NO_3^--N L^{-1}		
Control	NA	23.3 (4.8) b	17.8 (3.0) a	20.5 (2.8) a
Radish	NA	44.8 (5.8) a	8.7 (3.4) ab	20.0 (4.2) a
Rye	NA	6.0 (2.3) c	0.1 (0.0) b	2.7 (1.1) b
Radish+Rye	NA	7.1 (2.0) c	0.6 (0.2) b	3.7 (1.1) b
	Year 2 (31 Oct 2017 – 3 Jun 2018)			
	Fall-Winter¶	**Winter-Spring#**	**Spring-Summer††**	**Mean**
		mg NO_3^--N L^{-1}		
Control	22.4 (6.0) a	33.9 (2.7) a	14.4 (1.5) a	20.9 (1.8) a
Radish	3.0 (1.1) c	4.9 (0.6) b	3.1 (0.6) b	3.5 (0.5) b
Rye	16.7 (3.3) ab	0.7 (0.5) b	0.2 (0.2) b	3.2 (1.0) b
Radish+Rye	9.2 (1.6) bc	0.1 (0.0) b	0.0 (0.0) b	1.6 (0.5) b

† Fall-Winter = 1 Oct 2016 – 20 Dec 2016
‡ Winter-Spring = 21 Dec 2016 - 20 Mar 2017
§ Spring-Summer = 21 Mar 2017 - 5 Jun 2017
¶ Fall-Winter = 1 Oct 2017 – 20 Dec 2017
# Winter-Spring = 21 Dec 2017 - 20 Mar 20178
†† Spring-Summer = 21 Mar 2018 - 5 Jun 2018

Table 1.10. Estimated NO_3^--N leaching losses (kg ha^{-1}) by season and total across seasons by cover crop treatment. In the fall 2016, the drought conditions and minimal precipitation prevented soil solution collection, therefore values are indicated as NA. For year 1, standard errors are in parenthesis, but no letter designation for significant differences were made because of non-significance. For year 2, standard errors are in parenthesis and values with different letters are significantly different at P<0.05.

Year 1 (3 Jan 2017 – 27 May 2017)				
	Fall-Winter†	**Winter-Spring‡**	**Spring-Summer§**	**Total**
		kg NO_3^--N ha^{-1}		
Control	NA	15.3 (7.5)	16.1 (9.3)	31.4 (16.8)
Radish	NA	19.0 (15.3)	13.7 (10.7)	32.7 (26.0)
Rye	NA	4.2 (3.4)	1.2 (1.1)	5.40 (5.0)
Radish+rye	NA	5.8 (0.9)	2.0 (0.3)	7.9 (1.2)
Year 2 (31 Oct 2017 – 3 Jun 2018)				
	Fall-Winter¶	**Winter-Spring#**	**Spring-Summer††**	**Total**
		kg NO_3^--N ha^{-1}		
Control	10.9 (3.8) a	25.3 (3.5) a	16.8 (4.0) a	52.9 (4.4) a
Radish	2.2 (0.9) b	3.3 (0.8) b	2.9 (1.4) b	8.4 (2.0) b
Rye	7.6 (2.0) a	1.7 (0.9) b	0.2 (0.2) c	9.6 (1.3) b
Radish+rye	5.7 (1.1) a	1.0 (0.2) b	0.0 (0.0) c	6.7 (3.1) b

† Fall-Winter = 1 Oct 2016 – 20 Dec 2016
‡ Winter-Spring = 21 Dec 2016 - 20 Mar 2017
§ Spring-Summer = 21 Mar 2017 - 5 Jun 2017
¶ Fall-Winter = 1 Oct 2017 – 20 Dec 2017
# Winter-Spring = 21 Dec 2017 - 20 Mar 20178
†† Spring-Summer = 21 Mar 2018 - 5 Jun 2018

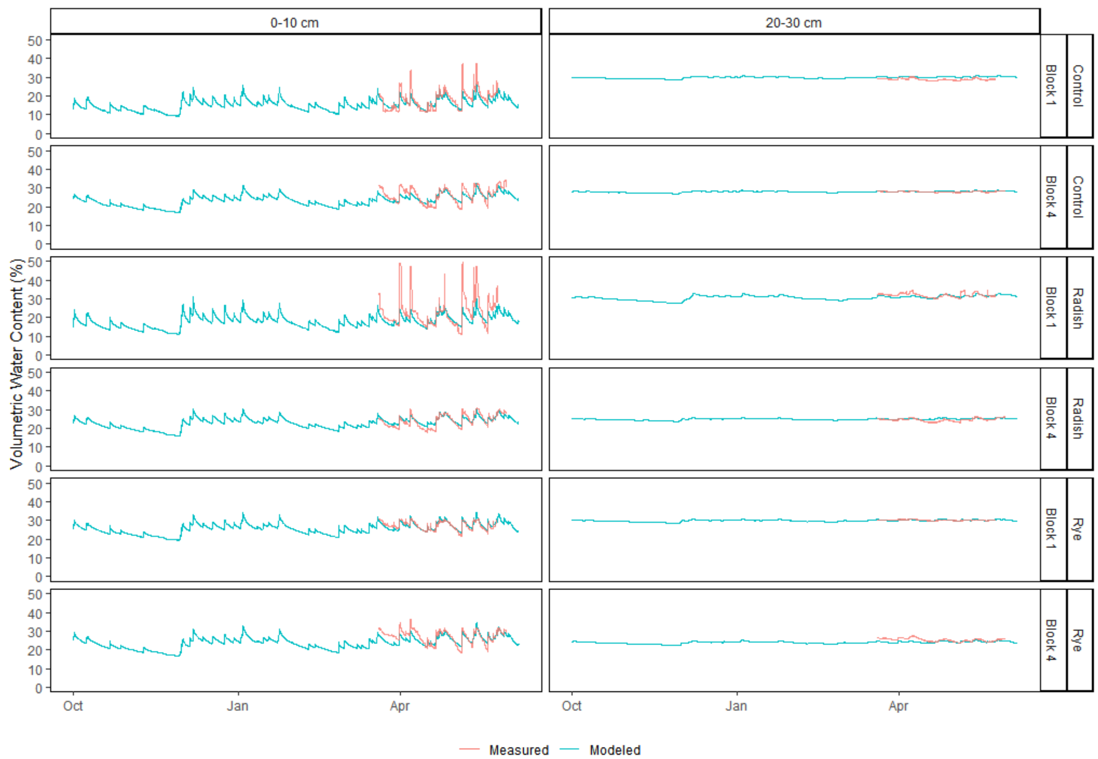


Figure 1.1. Volumetric water contents in surface soils (left: 0-10 cm and right: 20-30 cm) measured using Time-domain reflectometers (measured; red lines) and HYDRUS 1-D simulated volumetric water contents (modeled; blue lines) in 2016-2017 (Year 1) cover crop growing seasons in no cover crop control, radish, and rye plots

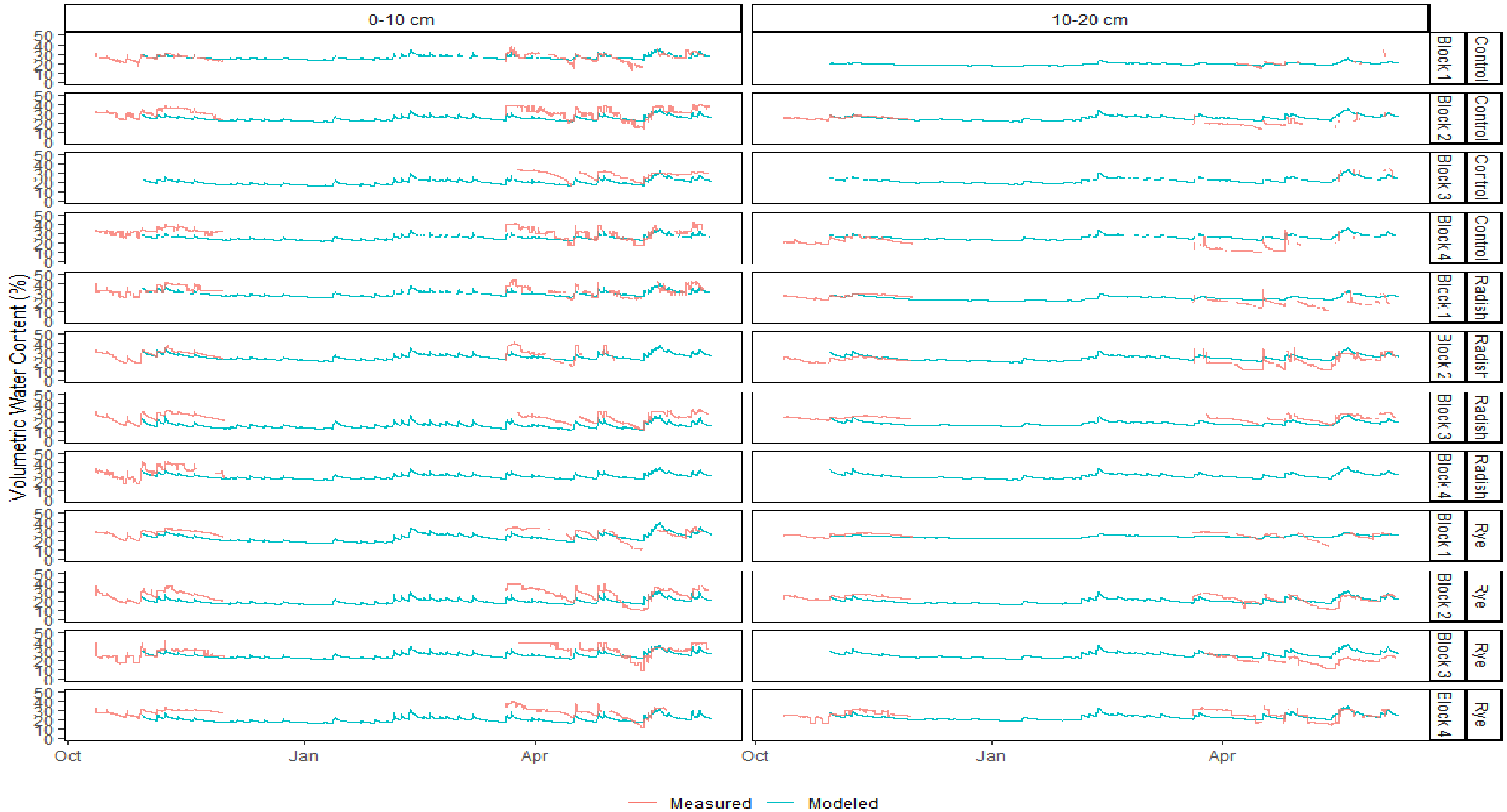


Figure 1.2. Volumetric water contents in surface soils (left: 0-10 cm and right: 10-20 cm) measured using Time-domain reflectometers (measured; red lines) and HYDRUS 1-D simulated volumetric water contents (modeled; blue lines) in 2017-2018 (Year 2) cover crop growing seasons in no cover crop control, radish, and rye plots.

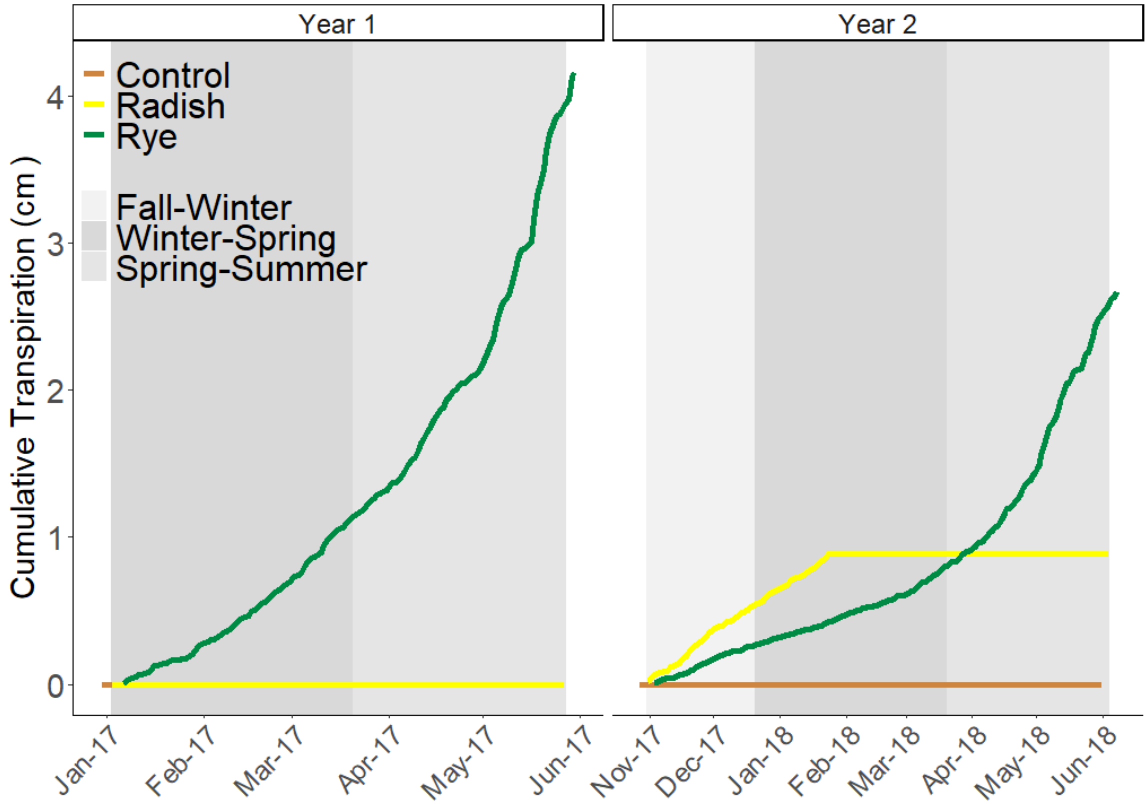


Figure 1.3. Estimated transpiration (cm) for control (brown), radish (yellow), and rye (dark green) for each study year. These data were simulated using HYDRUS 1-D.

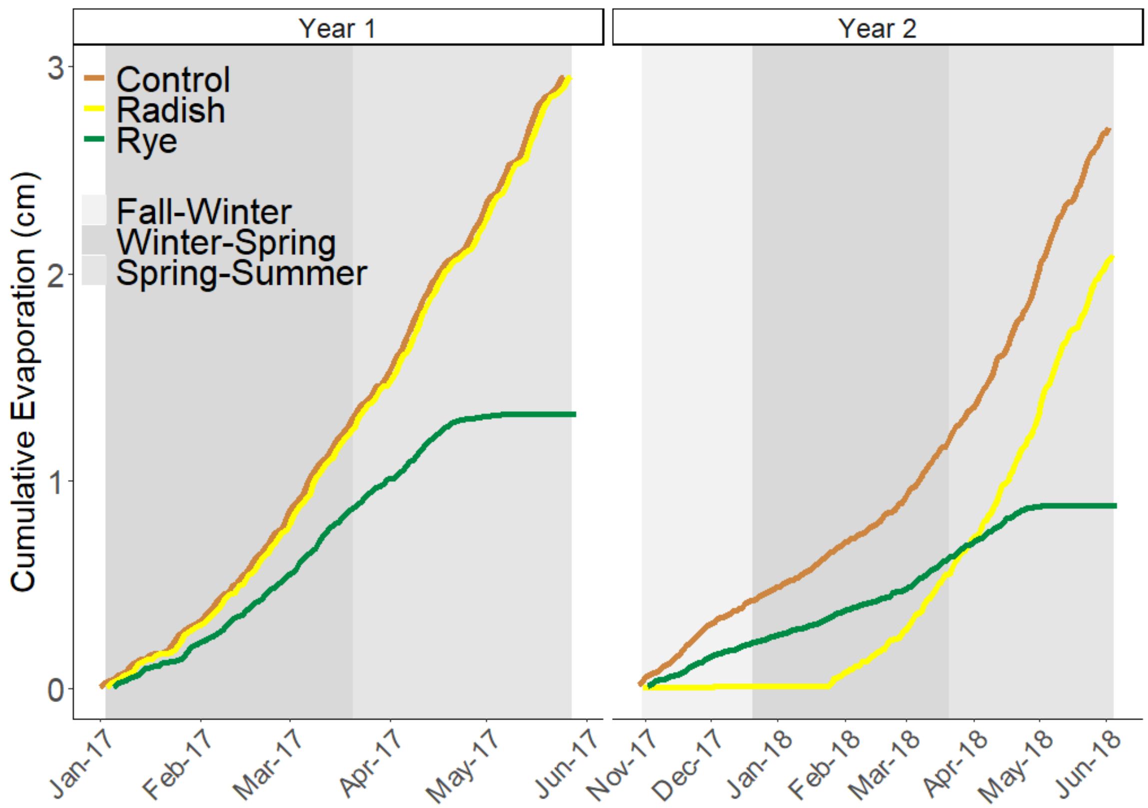


Figure 1.4. Estimated evaporation (cm) for control (brown), radish (yellow), and rye (dark green) for each study year. These data were simulated using HYDRUS 1-D.

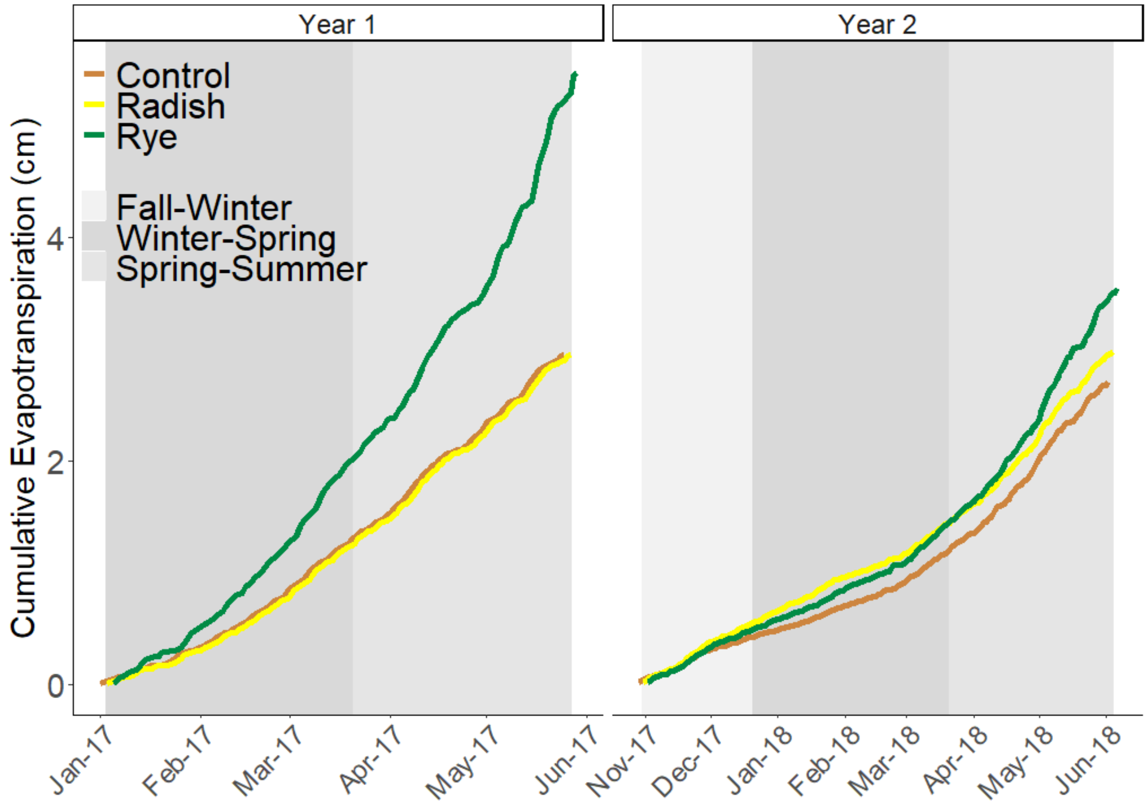


Figure 1.5. Estimated evapotranspiration (cm) for control (brown), radish (yellow), and rye (dark green) for each study year. These data were simulated using HYDRUS 1-D.

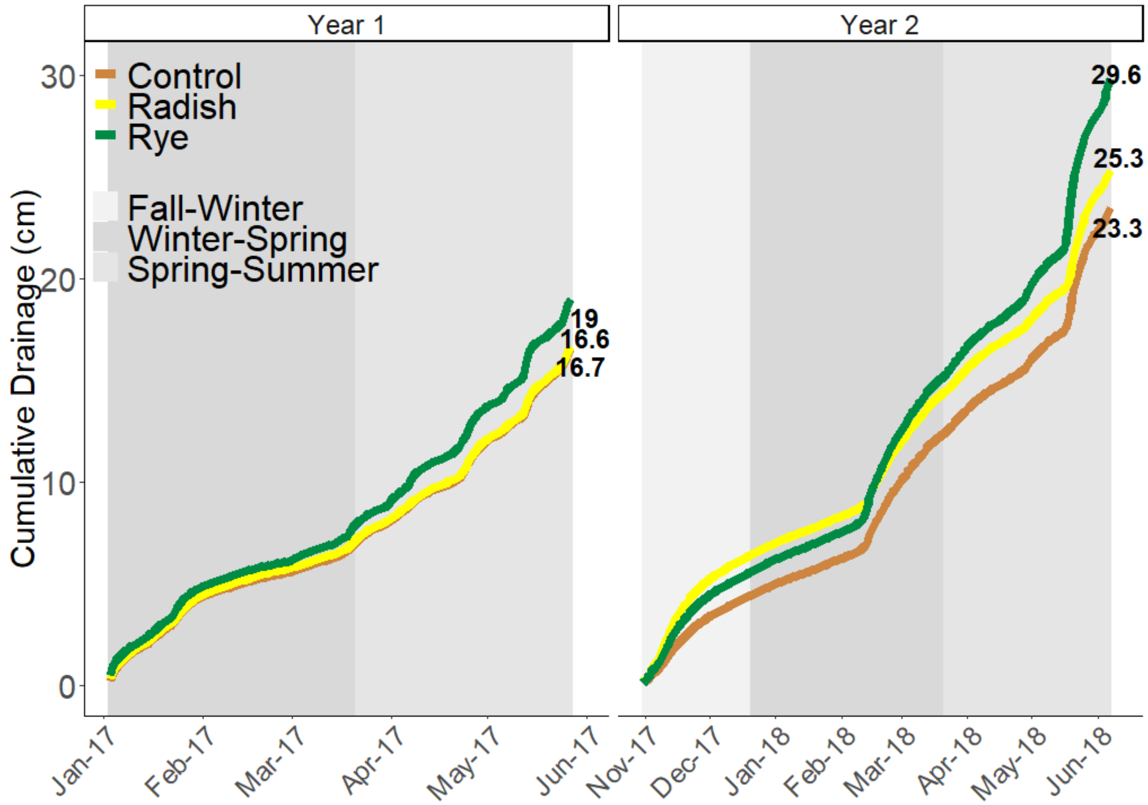


Figure 1.6. Estimated drainage (cm) past 60 cm soil depth from HYDRUS 1-D simulated drainage rates in year 1 (2016-2017) and year 2 (2017-2018) cover crop growing seasons in the no cover crop control (brown), radish (yellow), and rye (dark green) plots.

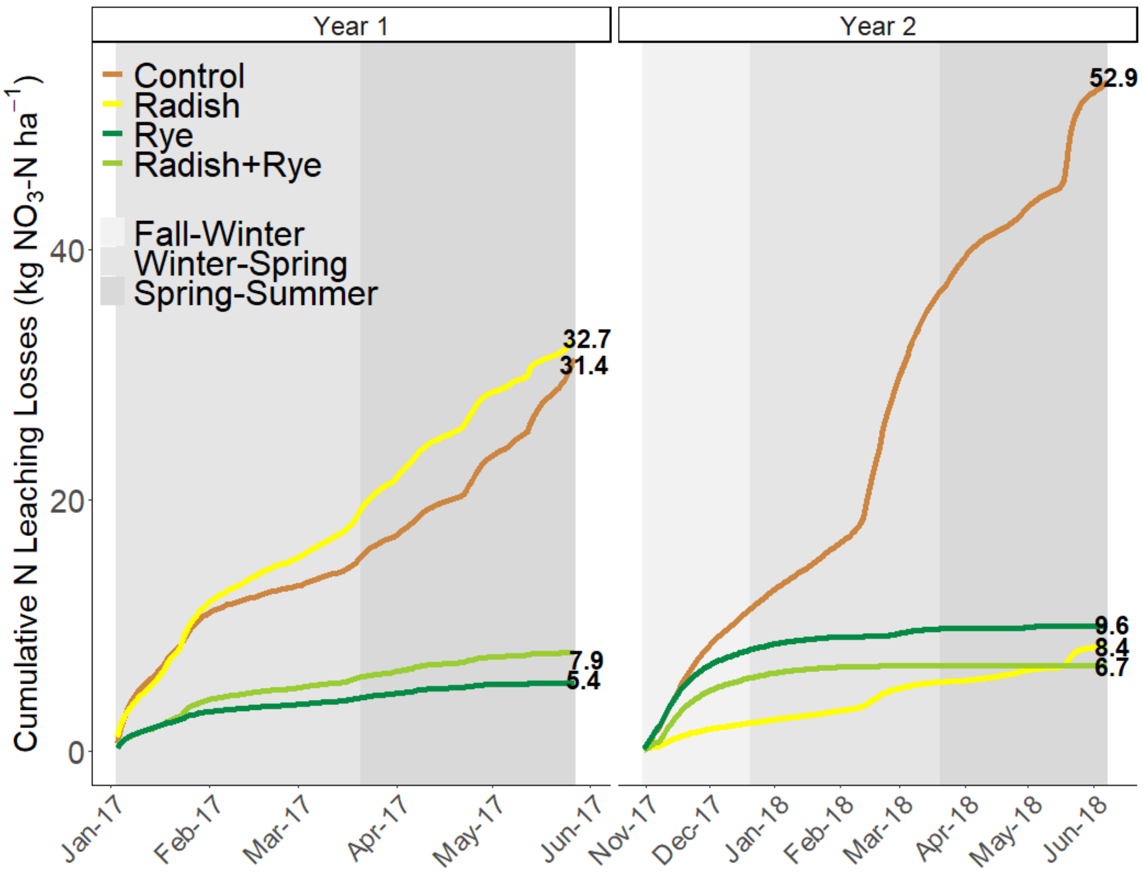


Figure 1.7. Estimated NO_3^--N leaching loss (kg ha^{-1}) for year 1 and year 2 with gray panels representing the different seasons. The left y-axis indicates the cumulative N leaching loss (kg ha^{-1}) for the no cover crop control (brown), radish (yellow), radish+rye (light green), and rye (dark green). Cumulative N leaching loss (kg ha^{-1}) were calculated using linearly interpolated soil solution concentrations collected from 60 cm soil depth, which were multiplied by modeled drainage fluxes calibrated against modeled volumetric water contents and scaled to kg ha^{-1}.

Chapter 2: Effect of cover crop mixtures on mineral soil N, cover crop biomass N, and N leaching

Introduction

About 42% of the nitrogen (N) loading into the Chesapeake Bay is attributed to N fertilizers applied to agricultural fields (Chesapeake STAT, 2019). Cover crops are an effective management tool for capturing residual N, since they have rapid growth in the fall or are winter hardy. In Maryland, 32% of farmed acres were planted with cover crops in the fall of 2017 (NASS, 2018; MDA, 2017). In the Mid-Atlantic, about half the annual rainfall occurs over the winter, and in the absence of cover crops, nutrients may be rapidly lost to ground and surface waters as residual N, primarily nitrate-N (NO_3^--N), which is water-soluble and readily moves through the soil profile (Jury and Nielson, 1989). A meta-analysis estimated that non-legume cover crops can reduce N leaching by 56% compared to systems without cover crops (Thapa et al., 2018a). Furthermore, the effectiveness of cover crops to suppress N leaching depends on cover crop planting dates and aboveground biomass produced, with greater effectiveness being observed with early planting dates, more aboveground biomass, and during cover crop growing seasons with relatively less precipitation (Thapa et al., 2018a).

Different cover crop species have different physiological growth habits, which may influence how they acquire nutrients in both space and time. For instance, cereal rye (*Secale cereal* L.) has a fibrous root system that creates a vegetative mat, reducing the impact from raindrops, decreasing erosion, increasing water holding capacity in the topsoil, and contributing to soil organic matter when the roots decompose (Blanco-Canqui et al., 2015). Additionally, rye can accumulate N in its biomass from the fall through the spring (Meisinger et al., 1991), but must be terminated in the spring by herbicides, mowing, burning, or tilling into the soil before planting a cash crop. For this reason, there are farmers who prefer planting forage radish (*Raphanus sativus* L.) because it winter-kills (< -4°C for several nights), which saves labor, fuel, and agrochemicals in the spring (Dean and Weil, 2009). Unlike rye, radish has a taproot structure that reduces soil compaction and increases infiltration, a process known as bio-drilling (Chen and Weil, 2009). In addition, radish can efficiently capture N in the fall before it winter-kills, especially if planted early (Sandler et al., 2015; White and Weil, 2011). The biomass N from the radish is incorporated back into the soil after a winter-kill event and may be susceptible to N leaching as the ground thaws in the spring (Dean and Weil, 2009).

Cover crop mixtures may enhance ecosystem services provisioning (e.g. scavenging N; supplying N; providing root channels; mulching; improving soil structure and function) because they combine positive traits of two or more crop species (i.e. occupy different niches). For instance, cover crop mixtures can produce more total biomass and biomass N than monocultures, and thereby provide a greater N supply to the subsequent cash crop upon decomposition (Thapa et al. 2018b). Williams and Weil (2004) showed that a radish+rye mixture increased the yield of soybeans when provided with adequate precipitation as compared to no cover crop and rye monoculture. We hypothesize that during the fall, radish+rye mixtures will accumulate more N in its biomass than rye or radish alone, thereby reducing the mineral soil N to 60 cm soil depth. We also hypothesize that during the spring, the rye in the mixture will reduce the susceptibility of N leaching losses from the radish biomass N after its winter-kill. If this is true, we expect that a radish+rye mixture may produce greater biomass and accumulate more N than its monoculture counterparts, thereby enhancing ecosystem services

We investigated the soil-(cover) crop N cycle to determine cover crop's ability to accumulate biomass N, reduce mineral soil N and estimated N leaching losses compared to a no cover crop control. Our soil systems approach will identify three key components of the soil-(cover) crop N cycle: (1) cover crop biomass N (tissue N); (2) mineral soil N (NO_3^--N + NH_4^+-N); and (3) estimated N leaching losses within four different cover crop treatments (forage radish, cereal rye, radish+rye, and a no cover crop control).

Methods

Study sites

This study was conducted near Laurel, Maryland on two adjacent fields over two growing seasons (2016-2018) at University of Maryland, Central Maryland Research and Education Center (CMREC; 39°01'40" N 76°50'14" W). The study site soils are a Russett-Christiana complex. Russett series surface soils are comprised of loam and sandy loam, mixed, semiactive, mesic Aquic Hapludults, while Christiana series surface soils are a silt loam, kaolinitic, mesic Aquic Hapludults. The bulk density of the soils ranged from 1.3 -1.8 g cm^{-3}

depending on the soil complex and depth. The mean annual precipitation at the site is 1046 mm, the mean minimum temperature is 6.8° C, and the mean maximum temperature is 19° C (USDA-ARS, 2019).

Experimental design

To determine the effect of cover crops and residual soil N levels on cover crop biomass N and mineral soil N, experimental plots were established in a randomized strip block design. Fall N treatments of calcium nitrate were applied in strips at three rates (0, 70, 140 kg-N ha^{-1}) prior to cover crop planting to mimic low, medium, and high levels of residual soil N after a corn cash crop. Each strip was divided into five sub-plots into which the cover crop treatments were planted. The cover crop treatments include: (1) forage radish (radish); (2) cereal rye (rye); (3) triticale (*Triticosecale* Witt); (4) radish+rye mixture; and (5) no cover crop control. Each treatment combination was replicated four times and subplots were 3.1 x 10.7 m with a 0.3 m buffer zone between each subplot in a strip. Cover crops were planted at 17.8 cm spacing using a no-till grain drill. Cereal rye and triticale were seeded at 126 kg ha^{-1}, and forage radish at 19.8 kg ha^{-1} in both years. The radish+rye mixture was seeded at 2.8/63 kg ha^{-1} (radish/rye) with one row of radish to one row of rye in year 1 and at 4.5/84 (radish/rye) kg ha^{-1} with one row of radish to two rows of rye in year 2.

Soil sampling and analysis

Soil cores (0-90 cm) were collected in mid-September (i.e. baseline soil collection) before planting cover crops and inorganic N fertilizer applications. Six cores were collected from each plot and segmented into six increments: 0-15, 15-30, 30-45, 45-60, 60-75, and 75-90 cm (Table 2.1). To quantify mineral soil N through time, soil cores (0-60 cm; 2.54 cm diameter) were collected in late October (after cover crop planting), mid-December (winter-kill event), and late March or early April (a common cover crop termination date; Table 2.1). We collected four cores from each sub-plot at four increments: 0-15, 15-30, 30-45, and 45-60 cm. Soils composited at each depth and were returned to the Agroecology Laboratory at the University of Maryland, air-dried, homogenized using a DynaCrush, and passed through a 2 mm sieve before further analysis. A sub-sample of soil was dried in the oven at 105° C for 3 d to determine soil moisture content on a gravimetric basis. A subsample of soil from each collection was extracted with 2 *M* potassium chloride (KCl), and filtered (Fisher Q2 2.5μm) prior to chemical analysis. Ammonium (NH_4^+-N) and NO_3^--N were analyzed colorimetrically on a LACHAT QuikChem 8000 series (HACH, Colorado, United States) using the sodium salicylate (US EPA, 1983) and cadmium-reduction (Fisherman and Friedman, 1989) methods, respectively. Soil particle size was determined on the baseline soils (0-90 cm) using the hydrometer method (Bouyoucos, 1962) for each year. Soil N concentrations (mg-NO_3^--N g^{-1} and mg-NH_4^+-N g^{-1}) were summed and scaled to mineral soil N (kg-N ha^{-1}) using bulk density (g cm^{-3}) and sampling depth (15 cm). Mineral soil N at each depth increment was summed at the subplot-level to obtain the total mineral soil N from 0-60 cm at subsequent collection dates.

Biomass sampling and analysis

Cover crop biomass was sampled both years in December (radish winter-kill), late March in year 1, and early April in year 2 (common cover crop termination date). We randomly placed two 0.125 m^2 quadrats in December or one 0.125 m^2 quadrat in March/April in the middle of each plot, collected all biomass within that area, and transported bulk samples to the Agroecology Laboratory at the University of Maryland for processing (Table 2.1). Cover crop biomass samples were oven-dried, to a constant weight; all reporting was done on a dry-weight basis. All plant tissue was dried at 50° C for 2 weeks and then ground through a 2 mm mesh (Wiley Mill, Swedesboro, New Jersey). Total tissue C and N content was determined using elemental analysis with helium as the carrier gas (LECO CN628, LECO Corporation, St. Joseph, Michigan). All biomass data were scaled to kg ha^{-1}. Biomass N was calculated by multiplying biomass (kg ha^{-1}) by the total N content in the tissue.

Estimating N leaching losses

Estimated N leaching losses were calculated by multiplying linearly interpolated soil solution concentrations (collected by pourous cup lysimeter) by drainage rates simulated in HYDRUS 1-D, and then scaled to kg-N ha^{-1}. Once we estimated the rate of N leaching losses, the values were summed over each cover crop growing season for each study year. In year 1, N leaching rate was summed from 24 October 2016 (lysimeter installation) through 20 December 2016 (winter-kill) and 21 December through 20 March 2017 (common cover crop termination date). For year 2, the N leaching rate was summed from 28

September 2017 (lysimeter installation) through 20 December (winter-kill) and 21 December through 20 March 2018 (common cover crop termination date).

Statistical approach

Nutrient data is frequently non-normally distributed, and data were transformed using Box-Cox transformations to satisfy the assumption of homogeneity of variance for model testing when necessary (Box and Cox 1964). All data were analyzed in the R environment for PC (R Core Team 2018). In order to determine the effect of cover crop species on mineral soil N (kg ha^{-1}) in 30 March 2017 (year 1), we used a linear model (LM) to run an analysis of variance (R package *lme4*, Bates et al. 2013) with cover crop species as the main effect and block as a random effect. We only examined the effect of cover crop species on mineral soil N (kg-N ha^{-1}) in the high fall N treatment since soils were collected in those plots only. During year 2, a linear model was used to test the effect of cover crop species and fall N treatment (main effects) and block (random effect) on mineral soil N (kg-N ha^{-1}) since soil samples were collected in every plot.

A linear model with cover crop species and fall N treatment as the main effects and block as a random effect was used to determine their effect on cover crop biomass (Mg ha^{-1}) and cover crop biomass N (kg-N ha^{-1}) for each individual collection date.

To test the effect of cover crop species on estimated N leaching loss we used a linear model with cover crop species as the main effect and block as the random effect for the winter-kill and common cover crop termination date for each study year. In all cases, we used Tukey *post hoc* tests to examine pairwise comparisons among cover crop treatments, fall N levels, or both where significance was established at P<0.05 (*multcomp*; Hothorn et al., 2008).

Results

Effect of cover crop in the high fall N treatments

On 20 Dec 2016 (year 1), total biomass was similar among cover crop treatments (P<0.05; Table 2.2; Figure 2.1C). Plots with radish had more cover crop biomass N than plots with radish+triticale (P<0.05), while there were no differences among the other cover crop treatments (Table 2.2; Figure 2.2C). Furthermore, there were no observable differences in mineral soil N among cover crop treatments (Table 2.2; Figure 2.3A). The minimal amount of rainfall from 24 Oct 2016 - 20 Dec 2016 led to no soil solution (Table 2.2).

On 30 Mar 2017, the same trends were observed, where there were similar amounts of total biomass among cover crop treatments (Figure 2.1C). Notably, there was no radish biomass due to the winter-kill event in mid- Dec 2016 (Table 2.1). However, we continued to sample both the soil and soil solution. In March 2017, there was significantly more mineral soil N to 60 cm (in kg ha^{-1}) in the radish and no cover crop treatments than the rye and radish+rye treatments (P<0.05; Table 2.2; Figure 2.3A). Similarly, estimated N leaching losses were higher under radish and no cover crop than under rye and radish+rye from 21 Dec 2016 - 20 Mar 2017, but due to a lack of statistical power, there was no significant differences detected among the treatments (Table 2.2).

On 19 December 2017 (year 2), biomass was significantly greater in the radish treatment than rye, triticale, and radish+rye (P<0.05; Table 2.3; Figure 2.1F). Rye had significantly more biomass than radish+rye (P<0.05), while triticale had similar biomass levels to both rye and radish+rye (Table 2.3). The radish treatment had significantly more biomass N than any other treatment (P<0.05; Table 2.3; Figure 2.2F). Although the rye treatment had higher biomass levels than radish+rye, biomass N was similar in treatments with rye, triticale, and radish+rye (Table 2.3; Figure 2.2F). Additionally, there was significantly more mineral soil N (in kg ha^{-1}) to 60 cm soil depth in the no cover crop control compared to all other treatments (P<0.05; Table 2.3; Figure 2.3B). Estimated N leaching losses were significantly lower under radish than rye, radish+rye, and no cover crop from 1 Oct - 20 Dec 2017 (P<0.05; Table 2.3).

On 4 April 2018, total biomass was similar between cover crop treatments (Table 2.3; Figure 2.1F). Biomass N levels were similar in rye and triticale treatments, but had more biomass N than the radish+rye treatment (P<0.05; Table 2.3; Figure 2.2F). The no cover crop control had significantly more mineral soil N than rye, triticale, and radish+rye (P<0.05; Table 2.3; Figure 2.3B), but there were no differences in mineral soil N between radish and no cover crop (Table 2.3; Figure 2.3B). Mineral soil N declined steadily from October to April for every treatment except for radish (Figure 2.3B). Under radish, there was an increase in mineral soil N to 60 cm from January through April (Figure 2.3B).

Estimated N leaching losses were lower in the radish, rye, and radish+rye treatments than the no cover crop control (P<0.05; Table 2.3).

Effect of fall N treatments on N cycling

On 20 Dec 2016, there was no significant difference in in total biomass or biomass N between the three fall N levels (Figure 2.1A-C; Figure 2.2A-C). Cover crop biomass N was greater in the radish treatment than the other cover crop treatments in the low fall N level (P<0.05), but we found no differences in biomass N among cover crop treatments in the mid fall N level (Table 2.2; Figure 2.2A-B).

On 30 Mar 2017, monoculture treatments had significantly more biomass in the mid fall N level than the high and low fall N levels (P<0.05), while both radish+rye and radish+triticale were similar between fall N levels (Figure 2.1A-C). Rye and triticale biomass N levels were highest in the mid and high fall N levels (P<0.05), while radish+rye and radish+triticale were similar between fall N levels (Figure 2.2A-C).

On 21 Dec 2017, there was significantly more biomass in the mid and high fall N levels than the low fall N level (P<0.05; Figure 2.1D-F). Cover crop biomass N followed a pattern where, high > mid > low fall N level (P<0.05; Figure 2.2D-F). In the high and mid fall N level, mineral soil N to 60 cm was greater in the no cover crop control than the other cover crop treatments (P<0.05; Figure 2.3B; Figure 2.4A-B). Mineral soil N levels were similar among radish, rye, and radish+rye regardless of fall N levels (P<0.05; Table 2.3).

On 4 Apr 2018, radish+rye had the least amount of biomass in the low and mid fall N levels ($P<0.05$; Figure 2.1D-F). Radish+rye in the low fall N level had less biomass than any cover crop treatment ($P<0.05$; Figure 2.1D-F). Radish+rye also had the least amount of cover crop biomass N in every fall N level, while rye or triticale had the most cover crop biomass N ($P<0.05$; Figure 2.2D-F). Mineral soil N to 60 cm followed the pattern: high > mid = low for the fall N levels ($P<0.05$; Figure 2.3B; Figure 2.4A-B).

<u>*Discussion*</u>

Resiliency of grass-brassica mixtures in the high fall N treatment

In December 2016, we did not observe differences in biomass or biomass N among the cover crop treatments, which may have been caused by the extreme drought conditions in the fall (NIDIS). This is in contrast with our expectations that radish biomass would be larger in the fall than other cover crop treatments as they have been documented to establish quickly and scavenge N in the fall (Sandler et al., 2015; White and Weil, 2011). However, the ability of radish to accumulate biomass and biomass N while suppressing N leaching losses is greatly influenced by the amount of time radish has to establish (Sandler et al., 2015; White and Weil, 2011). Radish only had 52 days to establish in year 1, which resulted in only 0.6 Mg ha^{-1} of biomass with 26.7 kg-N ha^{-1} in monoculture and 0.16 Mg ha^{-1} of biomass with 7.3 kg-N ha^{-1} when grown with rye (Table 2.2). Furthermore, we found no differences in mineral soil N among treatments with cover crops, which was likely due to the minimal N uptake by all species because of the late planting date. In March 2017, there was an increase in the rye, radish+rye, and triticale biomass and biomass N. Since at least one crop in rye and radish+rye plots persisted through the winter we observed less mineral soil N and estimated N leaching losses compared to no cover crop and radish (Table 2.2). These results indicate that the rye and radish+rye mixture are more resilient than radish grown in monoculture when they are planted late in the fall.

In December 2017, the environmental conditions and planting date were more favorable for radish and rye cover crops, evinced by the large biomass and biomass N (Table 2.3). In the mixture, there were two rows of rye to one row of radish, however, we observed more radish biomass and biomass N in the mixture (1.0 Mg ha^{-1} of biomass with 32 kg-N ha^{-1}) than the rye in the mixture (0.8 Mg ha^{-1} with 23 kg-N ha^{-1}), which indicates that the radish was the dominant species before the winter-kill event (Figure 2.5). Despite the differences in biomass N between cover crop treatments, there were similar amounts of mineral soil N, which was likely due to the lower estimated N leaching losses in the radish treatment compared to other treatments in the fall-winter of 2017 (Table 2.3).

In April 2018, we observed similar amounts of biomass in the rye, triticale, and radish+rye treatments (Table 2.3; Figure 2.1C and 2.1F). Other studies have found an associated over-yielding effect with grass-legume mixtures compared to their monoculture counterparts (Thapa et al., 2018b), which we did not observe with the radish+rye mixture. Additionally, there was less cover crop biomass N in the radish+rye mixture, which can be attributed to the absence of radish after the winter-kill event. Despite there being less cover crop biomass N in the radish+rye mixture, there were similar amounts of mineral soil N in plots with rye, triticale, and radish+rye (Table 2.3). We observed significantly less mineral soil N in rye, triticale, and radish+rye treatments as compared to the no cover crop control, since there was a cover crop taking up N from the soil from 22 Sep 2017- 4 Apr 2018 (Table 2.3). However, mineral soil N increased from 19 Dec 2017 to 4 Apr 2018 in the treatments with radish likely due to the mineralization of the radish biomass N after the winter-kill event (Figure 2.5). Of note, mineral soil N levels in the spring were similar in the radish and no cover crop control treatments, as they are both essentially bare in the spring. All cover crops were able to suppress estimated N leaching losses from 21 Dec 2017- 20 Mar 2018, since plots with cover crops had significantly less N leaching losses compared to no cover crop plots (Table 2.3; Figure 2.5). These results indicate that under favorable growing conditions radish can suppress N leaching losses while maintaining comparable mineral soil N to a no cover crop control.

Effect of fall N levels on cover crop treatments

High biomass N, low quality biomass (high C/N), and small amounts of mineral soil N are common characteristics of a rye cover crop. Studies show that rye can immobilize N during the decomposition process because of its high C content (Hargrove, 1986), essentially competing with the cash crop for N (Throup Kristensen 2003; “preemptive competition”). In order to ameliorate competition for N, some studies have investigated increasing the fertilizer N rate for the cash crop (Hargrove and Frye, 1987; Wagger and Mengel et al., 1988; Holderbaum et al., 1990), earlier rye termination (Wagger et al., 1989; Otte, et al., *in prep.*), and increasing biomass N by mixing rye with other species (e.g. legume-grass mixtures; Mitchell and Teel et al., 1977; Ranells and Wagger, 1997; Rosecrance et al., 2000). We hypothesized that mixing rye with a radish cover crop would increase total biomass and biomass N (in radish+rye treatments). Instead, we found that radish+rye either had similar or less total biomass and biomass N than its monoculture counterparts in both the fall and the spring, regardless of how much N was left from the previous cash crop. However, the radish+rye mixture had similar levels of mineral soil N to each monoculture species (Table 2.3). Grass-legume mixtures have been shown to increase the mineral soil N compared to rye monocultures (Rosecrance et al., 2000). However, in grass-legume mixtures, both crops are killed at the same time, therefore, there is a simultaneous release of N into the system for the cash crop (Ranells and Wagger et al., 1996). In a radish-rye mixture, radish is killed by the cold winter temperatures (in December), and the rye may take up some of the N released by the radish in the spring. Thus, radish+rye mixtures were able to suppress estimated N leaching from the fall through the spring.

Synthesis

To optimize N cycling, radish must accumulate all of its biomass in the fall before the winter-kill event. We found that larger quantities of cover crop biomass corresponded to smaller mineral soil N and less estimated N leaching losses, which is supported by a meta-analysis that showed at least 2 Mg ha^{-1} of cover crop biomass is necessary to reduce N leaching losses compared to a no cover crop control (Thapa et al., 2018a). In year 2, radish accumulated significantly more biomass as compared to year 1, which prevented large quantities of N from leaving the system via leaching. A study conducted in similar soils showed that mineral soil N levels were significantly lower under rye and radish cover crops in the fall (about 5-10 kg NO_3^--N ha^{-1} to 90 cm) compared to a no cover crop control (over 55 kg NO_3^--N ha^{-1} to 90 cm; Dean and Weil, 2009). Previous studies have found that rapid fall N uptake by radish was followed with rapid mineralization of N in the spring after winter-kill (Dean and Weil, 2009; Mueller et al., 1989), which we also observed in both the mid and high fall N levels during year 2. Mineralization of N from the radish biomass was not observed in the low fall N treatment in year 2 since there was very little radish biomass N. Additionally, we did not observe mineralization of N under radish in the spring of year 1 due to a later planting date and smaller biomass N compared to year 2 (Figure 2.3A).

Mineral soil N levels under rye are typically lower than no cover crop controls due to plant uptake in the spring (Shipley et al., 1992; Ritter et al., 1998; Rosecrance et al., 2000; Restovich 2012), which we also observed (Figure 2.3A-B and 2.4B). Rye was largely unaffected by the difference in planting date since it is winter-hardy and has a longer growth period (fall to spring) compared to a radish monoculture (fall-winter). Rye had a similar amount of biomass and biomass N in both study years, which allowed rye to reduce estimated N leaching losses as compared to the no cover crop control (Figure 2.1C, F and 2.2C, F).

During each year the radish in the radish+rye mixture winter-killed around the same time, but was planted about a month earlier in year 2. Even so, the radish+rye mixture reduced estimated N leaching losses compared to no cover crop more consistently than the other cover crop treatments (Gaimaro et al., *In prep*). There was less biomass in the radish+rye mixture in the spring as compared to the rye alone due to the radish winter-kill event, however, this did not correspond to greater estimated N leaching losses. Additionally, in year 2, radish was able to accumulate more N with 3 Mg ha^{-1} biomass compared to rye with 3.1 Mg ha^{-1} and the rye in the radish+rye with 2.4 Mg ha^{-1} of biomass (Figure 2.1C, F). However, in the case of radish, we were able to sample the whole plant (tuber and leaves) in comparison to only the above ground biomass in the rye, which could partially explain why the biomass N was larger in the radish than the rye and radish+rye in year 2 (Figure 2.2C and 2.2F). The role of roots was beyond the scope of this study, but might help explain why we observed similar reductions in mineral soil N in the radish+rye mixture with less cover crop biomass N as compared to the radish and rye treatments (Kristensen and Thorup-Kristensen, 2004).

Conclusion

This research has shown that a radish+rye mixture and its monoculture counterparts are valuable tools for managing N leaching following even a poor corn cash crop (high fall N level) by accumulating N in their tissues and reducing mineral soil N compared to no cover crop control. Further, we show that a radish+rye mixture was dominated by the uptake of N by radish in the fall (if given enough time to establish) and dominated by the uptake of N by rye in the spring resulting in less total biomass and biomass N than monocultures, but overall comparable mineral soil N levels and similar (low) N losses to leaching. The ability of radish to accumulate biomass and biomass N in both the monoculture and the mixture was largely determined by the planting date and all efforts should be made to plant radish early in the fall. When radish is incorporated into a mixture with rye there is a reduced risk of N leaching losses, therefore mixing radish with a grass can serve as an effective tool for managing N losses in the Mid-Atlantic region.

Table 2.1. Field activities for each study year including timing of planting, sampling, installation of equipment, cover crop termination, and sorghum harvest.

Year 1 (2016–17)		**Year 2 (2017–18)**	
16 Sep 2016	Pre-treatment soil cores	*19 Sep 2017*	Pre-treatment soil cores
16 Oct 2016	Fertilize, then plant cover crops	*22 Sep 2017*	Fertilize, then plant cover crops
24 Oct 2016	Install lysimeters	*28 Sep 2017*	Install lysimeters
24 Oct 2016	Sample soils	*28 Sep 2017*	Install TDRs
Mid December	Radish winter-kill event	*27 Oct 2017*	Sample soils
20 Dec 2016	Sample soils	*19 Dec 2017*	Sample soils
20 Dec 2016	Sample cover crop biomass	*19 Dec 2017*	Sample cover crop biomass
14 Feb 2016	Install TDRs	*Mid January*	Radish winter-kill event
30 Mar 2017	Sample soils	*4 Apr 2018*	Sample soils
30 Mar 2017	Sample cover crop biomass	*4 Apr 2018*	Sample cover crop biomass

Table 2.2. Nitrogen leaching, mineral soil N (NH_4^+-N + NO_3^--N), and cover crop biomass N during year 1. The N leaching is the sum of losses over the fall-winter (24 Oct - 20 Dec 2016) and winter-spring (21 Dec 2016 - 20 Mar 2017). Values in parenthesis are standard errors and values with different letters indicate significant differences at P<0.05.

		Fall-Winter				*Winter-Spring*			
Fall N level	**Cover crop treatment**	**Cover crop biomass†**	**Cover crop biomass N†**	**Mineral Soil N†**	**N leaching (estimated)¶**	**Cover crop biomass‡**	**Cover crop biomass N‡**	**Mineral Soil N‡**	**N leaching (estimated)#**
		Mg ha^{-1}	*kg-N ha^{-1}*	*kg-N ha^{-1}*	*kg-N ha^{-1}*	*Mg ha^{-1}*	*kg-N ha^{-1}*	*kg-N ha^{-1}*	*kg-N ha^{-1}*
High	No cover crop	-	-	168.1 (15.5)	-	-	-	135.8 (16.8) a	15.3 (7.5)
	Radish	0.6 (0.2)	26.7 (7.8) a	169.7 (13.7)	-	-	-	93.2 (14.9) a	19.0 (15.3)
	Rye	0.6 (0.1)	21.5 (5.7) ab	124.6 (15.6)	-	2.1 (0.4) cf	73.9 (14.0) a	48.6 (9.0) b	4.2 (3.4)
	Triticale	0.5 (0.1)	17.1 (2.6) ab	-	-	1.5 (0.4) cf	39.6 (11.4) bc	-	-
	Radish+rye	0.4 (0.1)	16.5 (3.4) ab	140.3 (20.7)	-	1.3 (0.1) cf	39.2 (2.6) ac	53.8 (8.0) b	5.8 (0.9)
	Radish+triticale	0.4 (0.1)	13.8 (3.1) b	-	-	1.04 (0.1) df	35.9 (5.8) bc	-	-
Mid-	No cover crop	-	-	-	-	-	-	-	-
	Radish	0.7 (0.2)	29.9 (6.7)	-	-	-	-	-	-
	Rye	0.5 (0.1)	19.9 (2.9)	-	-	2.7 (0.1) ab	55.3 (3.2) ab	-	-
	Triticale	0.6 (0.1)	21.9 (2.0)	-	-	2.8 (0.2) a	58.9 (6.5) ab	-	-
	Radish+rye	0.4 (0.0)	15.5 (1.6)	-	-	1.4 (0.1) cf	36.0 (0.9) ac	-	-
	Radish+triticale	0.5 (0.1)	20.2 (3.5)	-	-	1.2 (0.2) cf	36.8 (1.9) ac	-	-
Low	No cover crop	-	-	-	-	-	-	-	-
	Radish	0.6 (0.1) a	22.1 (3.2) a	-	-	-	-	-	-
	Rye	0.4 (0.1) b	15.1 (2.2) b	-	-	1.5 (0.1) cde	29.1 (2.7) cd	-	-
	Triticale	0.4 (0.0) b	12.8 (0.4) b	-	-	1.6 (0.2) bcd	28.2 (3.8) cd	-	-
	Radish+rye	0.4 (0.0) b	14.7 (1.8) b	-	-	0.9 (0.1) ef	18.7 (3.2) d	-	-
	Radish+triticale	0.5 (0.1) ab	17.6 (1.8) b	-	-	0.9 (0.1) f	25.2 (2.1) cd	-	-

† Fall-Winter = 20 Dec 2016
‡ Winter-Spring = 30 Mar 2017
¶ Fall-Winter = 24 Oct 2016 – 20 Dec 2016
# Winter-Spring = 21 Dec 2016 - 20 Mar 2017

Table 2.3. Nitrogen leaching, mineral soil N (NH_4^+-N + NO_3^--N), and cover crop biomass N during year 2. The N leaching is the sum of losses over the fall-winter (31 Oct - 20 Dec 2017) and winter-spring (21 Dec 2017 - 20 Mar 2018). Values in parenthesis are standard errors and values with different letters indicate significant differences at P<0.05.

		Fall-Winter				*Winter-Spring*			
Fall N level	**Cover crop treatment**	**Cover crop biomass†**	**Cover crop biomass N†**	**Mineral Soil N†**	**N leaching (estimated) ¶**	**Cover crop biomass‡**	**Cover crop biomass N‡**	**Mineral Soil N‡**	**N leaching (estimated)#**
		Mg ha^{-1}	*kg-N ha^{-1}*	*kg-N ha^{-1}*	*kg-N ha^{-1}*	*Mg ha^{-1}*	*kg-N ha^{-1}*	*kg-N ha^{-1}*	*kg-N ha^{-1}*
High	No cover crop	-	-	203.7 (31.2) a	10.9 (3.8) a	-	-	174.2 (53.9) a	25.3 (3.5) a
	Radish	3.0 (0.2) a	95.0 (5.4) a	78.4 (12.2) bc	2.2 (0.9) b	-	-	90.7 (33.0) ab	3.3 (0.8) b
	Rye	2.3 (0.1) b	67.3 (5.0) b	78.4 (7.2) bc	7.6 (2.0) a	3.1 (0.2) a	62.0 (6.1) a	60.4 (25.0) b	1.7 (0.9) b
	Radish+rye	1.7 (0.1) c	55.0 (1.0) b	77.6 (3.3) bc	5.7 (1.1) a	2.4 (0.3) a	49.9 (4.4) b	55.5 (26.2) b	1.0 (0.2) b
	Triticale	2.0 (0.1) bc	55.7 (6.8) b	89.2 (13.4) bd	-	3.0 (0.1) a	64.0 (4.0) a	69.3 (26.7) b	-
Mid-	No cover crop	-	-	155.8 (32.1) ab	-	-	-	78.3 (31.8)	-
	Radish	3.1 (0.5) a	81.2 (5.2) a	76.9 (14.7) cd	-	-	-	77.3 (38.6)	-
	Rye	2.4 (0.2) ab	55.4 (3.9) b	80.3 (9.1) cd	-	2.3 (0.3) ab	39.0 (5.3) a	37.3 (19.6)	-
	Radish+rye	1.3 (0.2) c	32.5 (4.0) c	68.7 (8.4) cd	-	0.8 (0.1) d	16.0 (2.0) b	48.5 (25.6)	-
	Triticale	1.9 (0.1) b	40.0 (3.8) c	78.1 (9.1) cd	-	2.6 (0.3) a	47.1 (7.9) a	46.0 (29.1)	-
Low	No cover crop	-	-	66.7 (5.3) cd	-	-	-	58.2 (24.4)	-
	Radish	2.3 (0.6) a	46.4 (12.4) a	65.8 (9.8) cd	-	-	-	54.4 (21.2)	-
	Rye	1.4 (0.2) a	23.2 (6.5) b	59.8 (3.3) cd	-	1.0 (0.2) cd	18.5 (3.2) b	53.0 (16.9)	-
	Radish+rye	0.8 (0.1) b	13.4 (1.0) b	56.9 (1.4) cd	-	0.5 (0.0) e	9.8 (0.3) c	39.0 (16.3)	-
	Triticale	1.5 (0.2) a	22.5 (2.7) ab	61.3 (8.8) c	-	1.5 (0.1) bc	23.2 (1.9) a	46.7 (19.4)	-

† Fall-Winter = 19 Dec 2017
‡ Winter-Spring = 4 Apr 2018
¶ Fall-Winter = 31 Oct 2017 – 20 Dec 2017
# Winter-Spring = 21 Dec 2017 - 20 Mar 2018

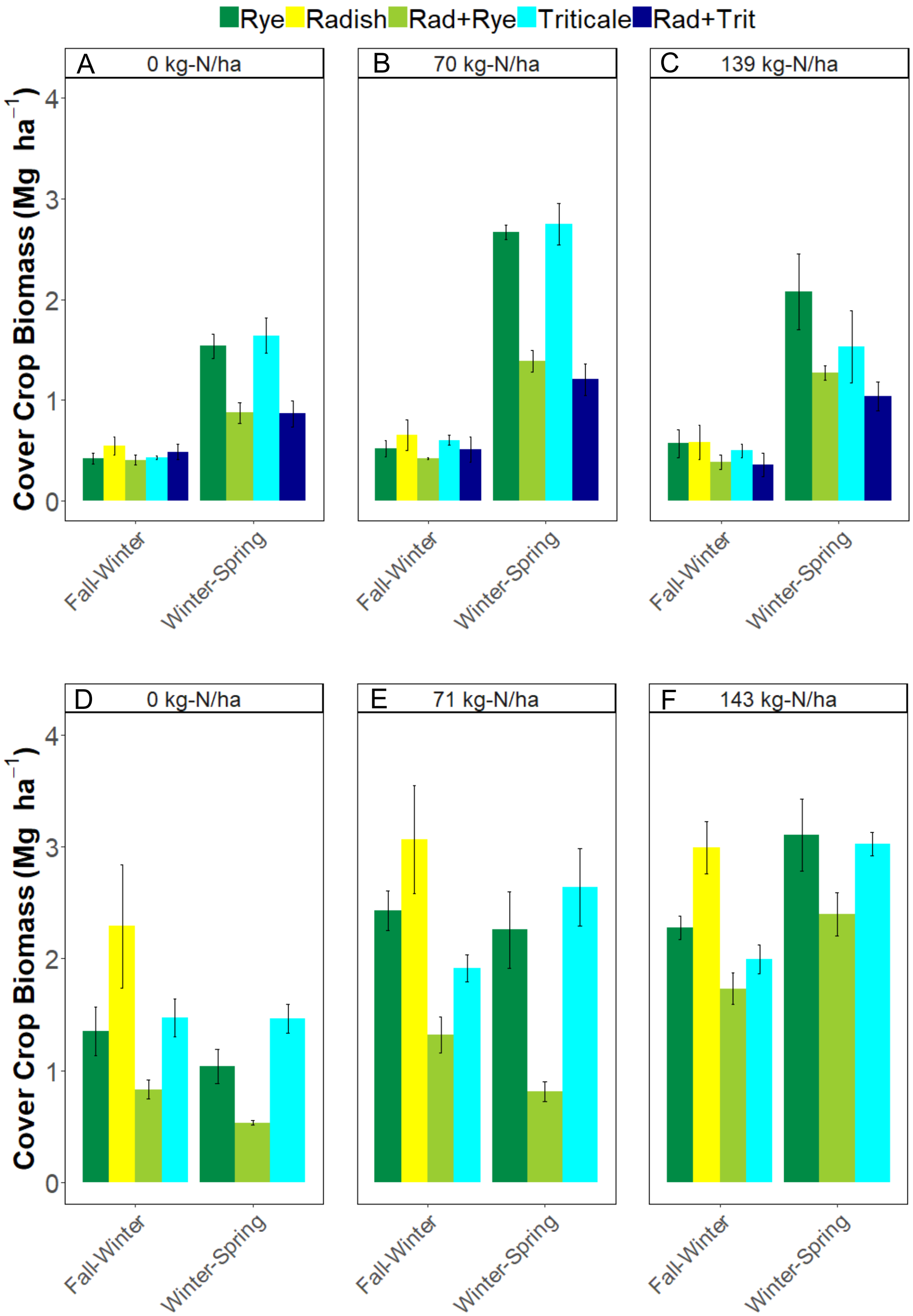


Figure 2.1. Cover crop biomass (Mg ha^{-1}) by collection date and treatment in (A-C) year 1 and (D-F) year 2 by (A, D) low, (B, E) mid, and (C, F) high fall N levels. Cover crop treatments include rye (dark green), radish (yellow), radish+rye (light green), triticale (cyan), and radish+triticale (dark blue). Vertical bars represent the standard error of the mean.

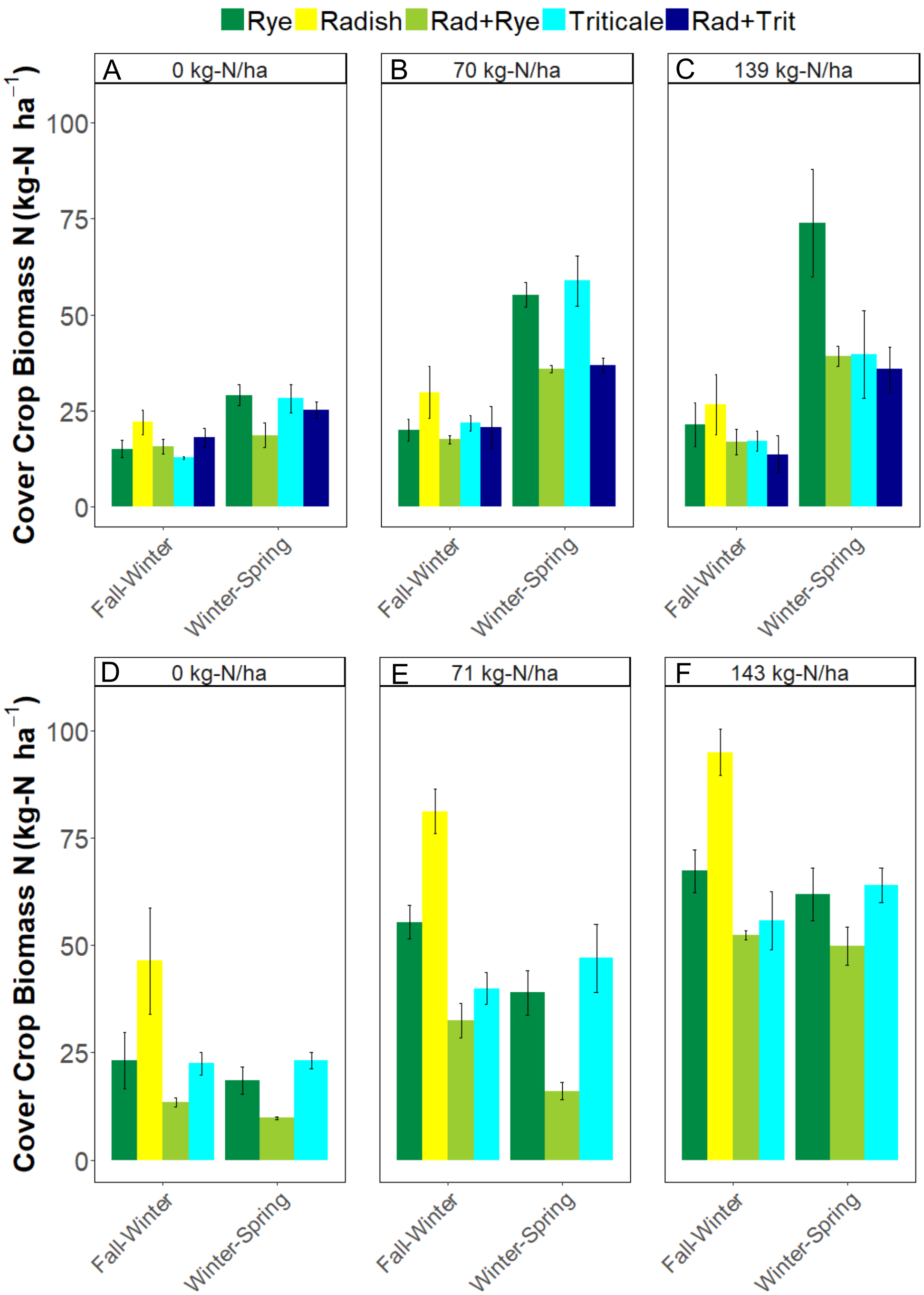
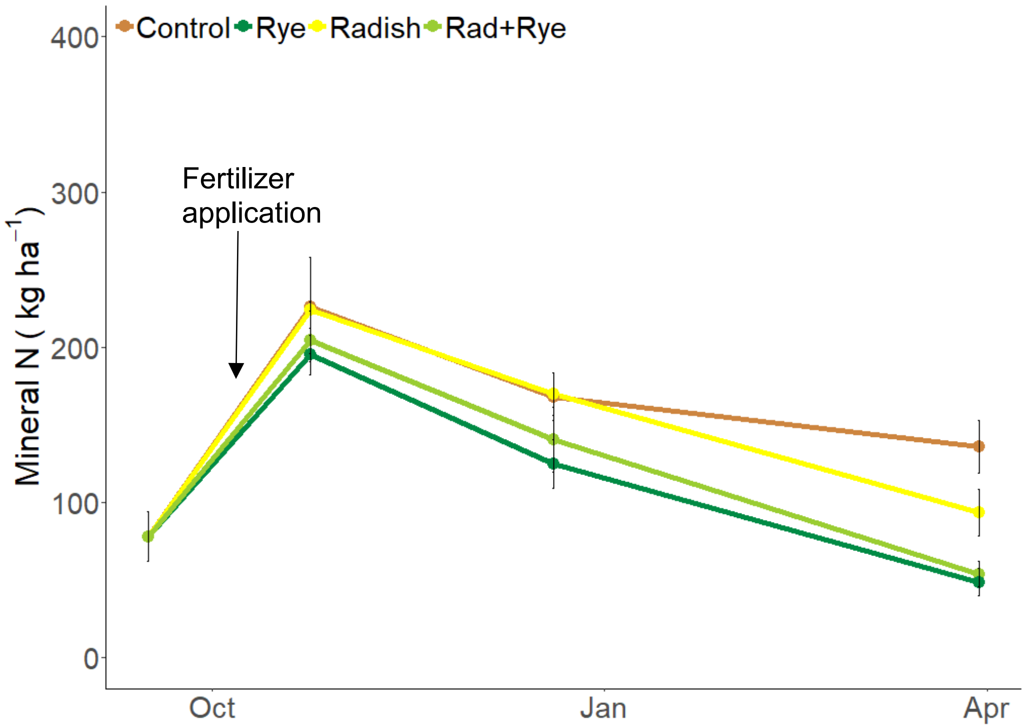


Figure 2.2. Cover crop biomass N (kg ha^{-1}) by collection date and treatment in (A-C) year 1 and (D-F) year 2 by (A, D) low, (B, E) mid, and (C, F) high fall N levels. Cover crop treatments include rye (dark green), radish (yellow), radish+rye (light green), triticale (cyan), and radish+triticale (dark blue). Vertical bars represent the standard error of the mean.

A



B

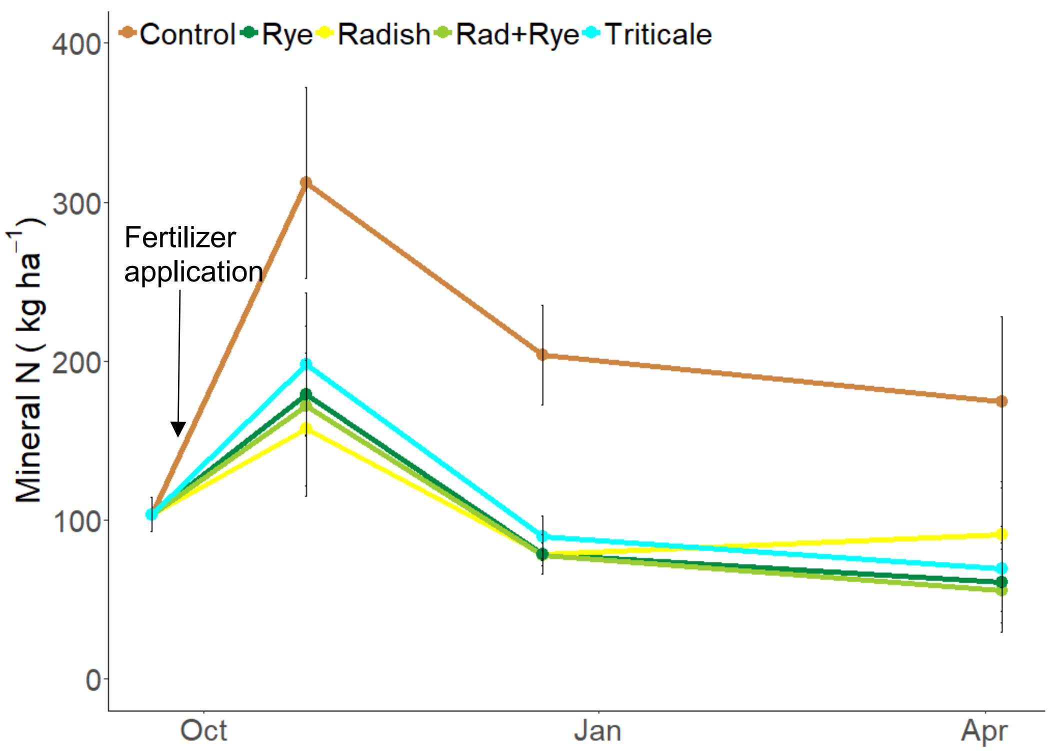


Figure 2.3. Mineral soil N (NH_4^+-N + NO_3^--N) in (A) year 1 and (B) year 2 for the high fall N level only. The cover crops treatments include: no cover control (brown), rye (dark green), radish (yellow), and radish+rye (light green), and triticale (cyan).

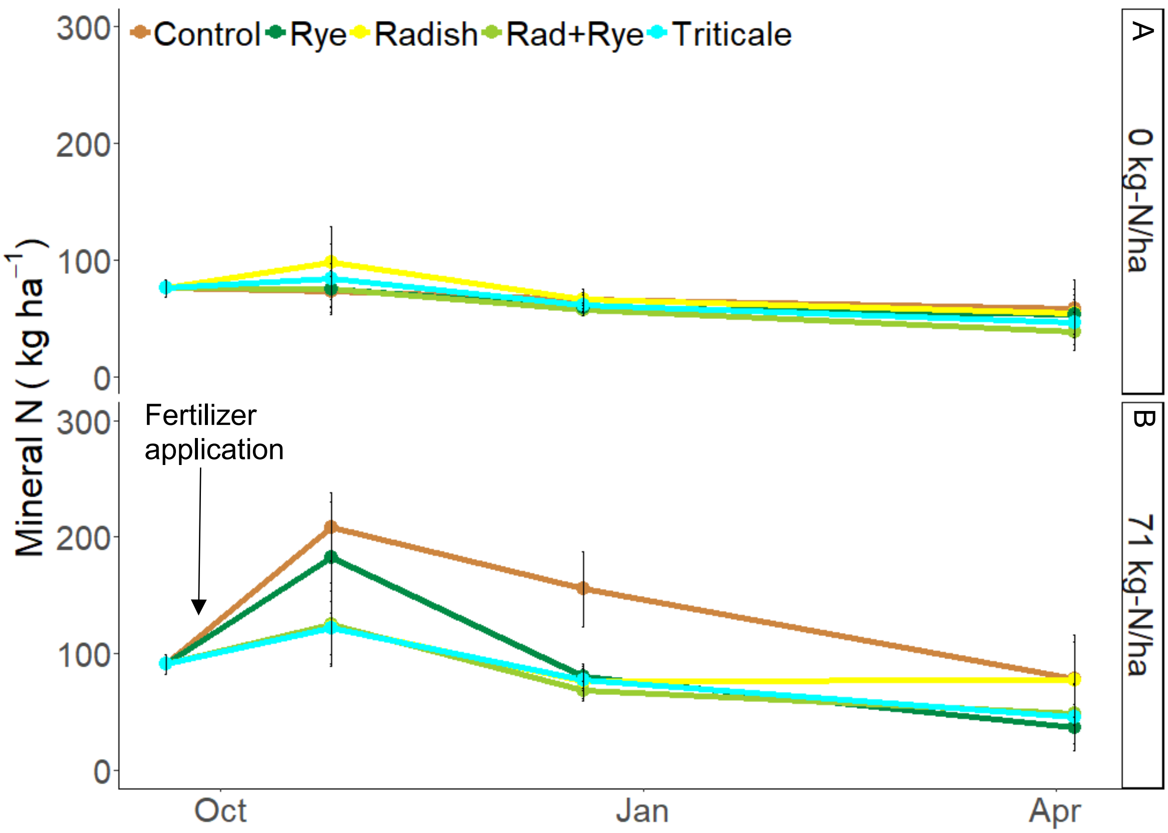


Figure 2.4. Mineral soil N (NH_4^+-N + NO_3^--N) from the low (0 kg-N ha^{-1}; A) and mid (71 kg-N ha^{-1}; B) fall N level during year 2. Cover crop treatments include control (brown), rye (dark green), radish+rye (light green), radish (yellow), triticale (cyan). Vertical bars represent the standard error of the mean

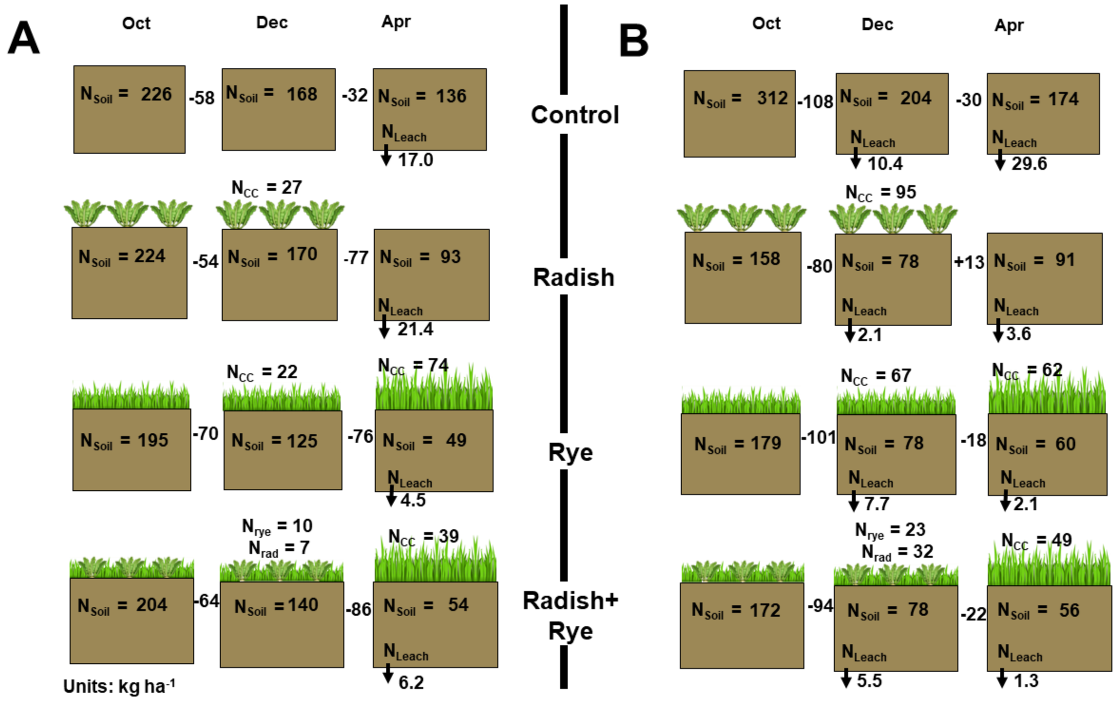


Figure 2.5. Mineral N (NH_4^+-N + NO_3^--N) and cover crop biomass N by cover crop treatment from 19 Dec 2016 - 30 Mar 2017 in year 1 (A) and 20 Dec 2017 - 4 Apr 2018 in year 2 (B) in the high fall N level. N leaching losses are from 1 Oct - 20 Dec and 21 Dec - 20 Mar for year 1 and year 2 in the high fall N level. All values are reported in kg ha^{-1}. Where N_{soil} is the mineral soil N, N_{cc} is the cover crop biomass N, and N_{Leach} is the estimated N leaching loss over that period.

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