INTERSOWING COVER CROPS INTO STANDING SOYBEAN IN THE UPPER MIDWEST

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MASTER OF SCIENCE

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ABSTRACT

In order to reduce nutrient losses and soil erosion in the United States Upper Midwest following soybean \textit{[Glycine max (L.) Merr.]}, cover crops can be intersown into standing soybean. The objective of this study was to determine the establishment of intersown cover crops and their impacts on a soybean-wheat \textit{(Triticum aestivum L.)} rotation. Four cover crops, winter camelina \textit{[Camelina sativa (L.) Crantz]}, winter pea \textit{[Pisum sativum ssp. arvense (L.) Poir]}, winter rye \textit{(Secale cereale L.)}, and radish \textit{(Raphanus sativus L.)}, were directly sown into the ground at the R4 and R6 stages of soybean at two locations, Prosper and Fargo, ND in 2016-2018. Results indicated intersowing cover crops have no impact on soybean yield, can produce above ground biomass which ranged from 0.44 to 3.04 Mg ha$^{-1}$, and show potential to mitigate soil nitrate losses in areas that grow soybean as a cash crop.
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1. INTRODUCTION

The lack of soil coverage with left over plant biomass, or “residue”, following a soybean harvest across the Upper Midwest is a concern. Soybean does not produce adequate amounts of residue to cover and protect the soil from erosion especially when precipitation, in the form of snow, is low. In soybean, 68% of its total biomass degrades and is lost from the field within 32 days after harvest (Broder and Wagner, 1988). Without adequate soil coverage, soil and nutrients are lost due to wind and water erosion. Precious topsoil lost will not be recoverable in the near future. It is estimated that unprotected soil can see topsoil losses from 6 Mg ha\(^{-1}\) to 18 Mg ha\(^{-1}\) annually (Hansen et al., 2012). If some course of action is not taken soon, soil losses will diminish land productivity and sustainability. If land productivity decreases, producers must increase inputs to enable enough production to supply food, feed, and fiber for a growing global population (Doran, 2002).

In the Upper Midwest, soybean harvest takes place starting in late September and continues into late November. Cover crops sown following a soybean harvest have very limited time to grow and provide cover before the first killing frost. Thus, the alternative may lie in direct intersowing or broadcast seeding of cover crops into standing soybean. The majority of the research has focused mainly on broadcasting over the cash crop than on direct sowing into it. Research has been done involving intersowing cover crops into corn [\textit{Zea mays} (L.)], with no decrease in corn grain yield (Baributsa et al., 2008). Berti et al. (2017) reported winter camelina intersown into standing corn and soybean at multiple sowing dates. In this study, direct sowing winter camelina on the same day as corn or soybean led to a reduction in grain yield. Sowing dates after the V4 stage that were broadcasted did not reduce corn or soybean yield (Berti et al., 2017).
If cover crops are drilled in between rows when corn or soybean are still standing, cover crops can establish under the canopy. Early establishment will allow cover crops to maximize growth and soil coverage, making the best of the short growing season in the Upper Midwest. Several studies have been conducted to study the response of cover crops intersown into corn (Sandler et al., 2015; Belfry and Van Eerd, 2016; Blanco-Canqui et al., 2017), but fewer studies have focused on intersowing in soybean. Intersowing radish into soybean did not reduce soybean yield while increasing radish biomass when compared with radish sown after soybean harvest (Sadler et al., 2015). Similarly, Belfry and Van Eerd (2016) reported a cover crop mix biomass accumulation of 1116 kg ha\(^{-1}\) when sown into V4-V6 corn which was a 33% greater compared with the later sowing at V10-V12. Blanco-Canqui et al., (2017) stated that soil cover in corn increased from 24% cover with no cover crop to 65% cover in plots intersown with winter rye. The increased green cover from the growing cover crops provides protection against soil erosion due to wind or water (Blanco-Canqui et al., 2011).

Cover crops can also provide many soil health benefits, such as: improving soil structure, increasing water infiltration, soil microbial activity, providing wildlife habitat, and scavenging of nutrients otherwise lost by erosion or leaching (Blanco-Canqui et al., 2011). Radish has the ability to scavenge NO\(_3\)-N from the soil ranging from 19.7 to 202 kg ha\(^{-1}\) (Ruark et al., 2018), and winter camelina can sequester residual soil NO\(_3\)-N in the biomass throughout the fall and resume scavenging in the spring. Berti et al. (2017) indicated sequestered NO\(_3\)-N can vary from 24 and 59 kg N ha\(^{-1}\) in the above ground biomass (Berti et al., 2017). If cover crops are not established early enough, limited growth will result in no added benefits.
2. OBJECTIVES

I. To measure cover crop performance under the soybean leaf canopy at two different sowing dates.

II. To measure cover crop impact on soybean yield and subsequent spring wheat yield.

III. To determine cover crops scavenging ability of fall soil NO$_3$-N and subsequent spring NO$_3$-N.
3. LITERATURE REVIEW

The Upper Midwest in North America, where most of the annual crops in the USA are grown, requires resilient cropping systems to remain productive as climate change takes place (Unger and Vigil, 1998). Soil erosion due to wind in the United States Upper Midwest has been an issue for past centuries. Research has been conducted to quantify how much soil is lost in this region due to wind erosion, which can range from 5 to 18 Mg ha\(^{-1}\) yr\(^{-1}\) (Hansen et al., 2012). Soils are most susceptible to wind erosion in late winter and early spring when crops are not present, leaving soil bare and vulnerable to erosion by high winds (Blanco-Canqui et al., 2011).

Tillage practices can play a major role in increasing or decreasing the amount of soil lost from wind erosion. Long-term conventional tillage and cultivation has caused major loses not only of topsoil but also of nutrient availability of extractable P, extractable K, surface pH, total C, and total N. Reduced productivity of the land will lead producers to increase inputs to maintain crop yield and farm income. Farmers that utilize intensive cropping practices and apply reduced tillage management can improve soil quality and agricultural sustainability (Malo et al., 2005).

Crop rotations in the United States have slowly decreased in diversity starting from 1978 and continued in to 2019. From 2012 on, corn-soybean rotations became more prominent in the Upper Midwest. As improved genetics push the hybrids/cultivars further North with earlier maturity hybrids/cultivars, crop diversity is decreasing (Aguilar et al., 2015). Reduction in diversification in a cropping system compounded with poor soil management strategies can have many negative impacts, causing reduction in crop yield and in soil health (Malo et al., 2005; Hansen et al., 2012).
In a corn-soybean or wheat-soybean rotation, little residue is left after soybean harvest. In an experiment done by Alberts et al., (1985) in central Missouri, continuous soybean on average lost more soil to erosion than continuous corn. One method to reduce impacts of soil erosion and increase the diversification of crops is introducing cover crops into crop rotations (Blanco-Canqui et al., 2015) by intersowing into standing soybeans.

Cover crops can be defined in many ways. As the name states, it is a crop sown to cover the ground, in between seasons when no cash crop is present. Cover crops are defined as: grasses, legumes, and other forbs that are planted for erosion control, improving soil structure, moisture, and nutrient content, increasing beneficial soil biota, suppressing weeds, providing habitat for beneficial predatory insects, facilitating crop pollinators, providing wildlife habitat, and as forage for farm animals (USDA-NRCS, 2019). Furthermore, cover crops can provide energy savings both by adding N to the soil and making more soil nutrients available, thereby reducing the need to apply fertilizer. In the Upper Midwest, winter-hardy cover crops are sown directly after harvesting the main crop and are termed a winter cover crop (Jawad et al., 2013). The main focus behind cover crops is introducing additional crops to cropping systems within the same growing season which will increase soil cover, nutrient cycling, soil organic matter, and crop diversity (Blanco-Canqui et al., 2015).

According to the National Agricultural Statistics Service (2017), over 6.23 million ha nationwide were sown with cover crops; 393,804 ha in Iowa, 579,147 ha in Minnesota, and 404,267 ha in North Dakota with an increase of cover crops area of 156%, 41.9%, and 89.1%, respectively, compared with the cover crops area in 2012 in the same states (USDA, 2017). Cover crops can provide a great benefit to the soil by holding it in place and reducing wind and
water erosion. It has been shown that sediment loss from fields due to water erosion can be reduced from 40 to 96% when using cover crops (Blanco-Canqui et al., 2015).

Camelina has not been studied much for its use as a cover crop. Camelina is part of the Brassicaceae family and is believed to have originated in the regions of southeast Europe and southwest Asia (Berti et al., 2016). Camelina is typically used to produce seed meal and oil, which have many uses, including feedstock for animals, humans, and biofuels (Berti et al., 2016; Zanetti, et al., 2017) and now is of growing interest to use as a cover crop. Camelina has shown to have both spring and winter annual biotypes. The winter annual biotype has become more appealing in the Upper Midwest due to winter camelina being very winter hardy, making it capable of surviving harsh winters (Gesch and Archer, 2013; Gesch et al., 2014; Berti et al., 2017).

The ability of the winter biotype surviving the winter allows for an increase in biodiversity in the crop rotation through double or dual cropping of winter camelina into soybeans (Gesch and Archer 2013; Gesch et al., 2014). Winter camelina is known to flower early in the Upper Midwest, peaking at the end of May, allowing for ecosystem services to pollinators and habitat for beneficial insects (Berti et al., 2016). Winter camelina can also reduce the soil nitrates and decrease the likelihood of leaching and runoff. Berti et al. (2017) has shown winter camelina to acquire up to 24 and 59 kg N ha⁻¹ in the above ground biomass. In addition, winter camelina after soybean and before corn or wheat is a viable winter-hardy broadleaf crop option available. Winter rye is currently one of the few cover crops that survive North Dakota winters but when sown in the fall before corn can reduce its yield and before wheat can cause grain contamination (Moyer and Blackshaw 2009; Krueger et al., 2011).
Winter pea is a winter annual legume that can survive the winter in Kansas and some parts of the Pacific Northwest (Chen et al., 2006; Holman et al., 2018), but less research has been done in winter pea in the Upper Midwest. In general, winter pea available cultivars do not survive most winters in North Dakota. Winter legumes sown, as a cash crop, in the fall can fix N; research has shown winter pea to provide between 99 and 123 kg N ha$^{-1}$ credit to the following crop. It also provides water-stable aggregates that improve water infiltration (McVay et al., 1989).

Chen et al. (2006) demonstrated that winter pea planted early in the fall after a small grain can produce a significantly higher amount of biomass the following year when compared with a spring sown pea. Not only do legume cover crops fix N, they also scavenge excess nutrients in the soil. Research has demonstrated that the N tied up in winter pea biomass decreases C:N ratio. A low C:N ratio can lead to biomass readily broken down, promoting microbial activity and enhancing nutrient availability (Liang et al., 2014).

Like winter camelina, radish is part of the *Brassicaceae* family. Research has been done on radish as a cover crop, mainly looking at how the roots improve soil structure and their ability to scavenging nutrients. Brassica cover crops like the radish are deep-rooted and help alleviate effects of soil compaction by providing root pathways through hard pans in the soil for succeeding cash crops, like soybean. This is especially beneficial during limited water availability, allowing the soybean to reach down into the subsoil water (Williams and Weil, 2004; Gruver et al., 2019).

Gruver et al., (2019) demonstrated the radish tap root has shown to grow up to 30-cm deep reaching 3-cm in diameter, but under favorable growing conditions the tap root can reach about 1-m deep into soil in 60 days. The tap root growth in the soil allows for greater infiltration
of water the following spring after radish is winter killed (Gruver et al., 2019). Radish also has the potential of scavenging excess nutrients from deep in the soil profile. Radish was reported to take up 19.7- to 202- kg NO$_3$-N ha$^{-1}$ from the soil (Ruark et al., 2018). After radish winter kills, the N in its biomass is released in March and April, which can be taken up by soil microbes or early-sown cash crops which could lead to potential increasing yield (Lounsbury and Weil, 2015).

Winter rye is a cool-season winter-annual in the grass family. This is a well know cover crop in the Upper Midwest due to how versatile it can be. One of the greatest strengths of winter rye is its high tolerance to cold temperatures. Winter rye has been demonstrated to survive harsh winters and continue growth in the spring (Applegate et al., 2017; Noland et al., 2018). With growth in the spring time winter rye can be used as a water management strategy to reduce excess water (Krueger et al., 2011; Appelgate et al., 2017). Along the same idea of reducing excess water, rye can be used to scavenge for excess nutrients susceptible to leaching or runoff, which could later impact water quality (Kasper et al., 2007; Krueger et al., 2011; Kladivko et al., 2014; Appelgate et al., 2017).

In addition, winter rye is known for its allelopathic affects that can be used as a weed management tool. In a no-till organic system, winter rye was able to provide effective management of weeds compared with normal weed control of tillage (Bernstein et al., 2014). Other research has demonstrated that the impact of the residue itself smothering the weeds can contribute to weed control (Barnes and Putnam, 1983). A study conducted in Canada, found that winter rye significantly decreased broadleaf weed populations between 44% to 72% and grass weed populations by 43% to 88%, when compared with a bare soil control (Flood and Entz, 2019). Without proper termination management, rye in the spring can lead to a decrease in the
following crop yield due to decreases in soil water, nutrients locked up in the biomass and allelopathic effects on the main crop (Moyer and Blackshaw 2009; Krueger et al., 2011).

Intersowing cover crops into winter wheat in the spring has been done in Sweden, allowing early growth of winter wheat without competition from the cover crops compared with a spring cereal sown at the same time as cover crops. To reduce the use of synthetic fertilizer for a spring barley (*Hordeum vulgare* L.) cash crop, cover crops including red clover (*Trifolium pratense* L.), white clover (*Trifolium repens* L.), and perennial ryegrass (*Lolium perenne* L.) were intersown perpendicular to established winter wheat the previous year (Bergkvist et al., 2011). Cover crops sown into winter wheat will reduce the chances of NO$_3$-N leaching after N fertilizer is applied for the spring barley. The cover crop mix provided the greatest uptake of soil mineral N reducing NO$_3$-N leaching. Across all treatments, no negative effect on winter wheat grain yield was noticed. The surviving red clover after wheat can increase a succeeding spring barley yield by 2 Mg ha$^{-1}$ (Bergkvist et al., 2011).

In a study in southeast France, legume cover crops were sown into winter wheat using a manual centrifugal seed spreader at the end of wheat tillering, and no penalties were reported on grain yield when four legume species were sown into tillering winter wheat. Although there was no yield reduction in wheat after intercropping, some legume species reduced the protein content of winter wheat grain (Amossé et al., 2013). Undersowing cover crops at late tillering in winter wheat allows enough time for adequate establishment and greatest biomass yield of the cover crops before winter wheat harvest (Amossé et al., 2013).

Neugschwandtner and Kaul (2014) demonstrated that an oat (*Avena sativa* L.)-pea (*Pisum sativum* L.) intercropping showed no yield gains and generally had lower yield compared
with oat monoculture or pea monoculture. This was due to the oat out competing pea for nutrients and water.

Several studies have shown that intersowing cover crops after the V5 stage of corn do not decrease corn yield (Baributsa et al., 2008; Balde et al., 2011; Berti et al., 2017; Flores-Sanchez et al., 2013; Noland et al., 2018). However, Ruffatti et al. (2019) found corn yield reductions of 7% to 22% in some environments when intersowing cover crops at reproductive stages of corn. Baributsa et al. (2008) reported that intersown red clover into corn population of 75,000 plants ha\(^{-1}\) does not decrease corn yield. At a corn population of 75,000 plants ha\(^{-1}\); red clover was able to produce biomass that ranged from 3.10 to 6.05 Mg ha\(^{-1}\) when sampled in the following spring (Baributsa et al., 2008).

Baldé et al., (2011) reported no yield drag on corn at any of the intersowing dates of cover crops, including the earliest sowing date shortly after corn emergence. This is where the greatest amount of competition from cover crops was expected. Consequently, cover crop biomass was significantly lower when intersown versus grown as a sole crop. The decreased cover crop biomass when intersown could be influenced by reduced photosynthetically active radiation (PAR) and resources competition with corn. Cover crops sown on an earlier date produced greater biomass than those on later sowing dates (Baldé et al., 2011).

In southwest Mexico, legumes intersown into corn did not have any effect on grain yield. The legumes served as a multifunction crop providing not only N\(_2\) fixation, but as catch crops, reducing nutrient losses due to leaching (Flores-Sanchez et al., 2013). However, decreases in yield have been seen when intersowing winter camelina into corn, where the only yield reduction to corn was observed when camelina and corn were sown the same day (Berti et al., 2017). Same day sowing of camelina with corn lead to a yield decrease of 14%, but when camelina was
intersown at corn stages V4-V5 or later, corn showed no reduction in yield (Berti et al., 2017). Belfry and Van Eerd, (2016), found similar results of no yield reduction when intersowing 17 different cover crop species along with different mixes into corn stages V4-V6. This study also demonstrated intersowing cover crops at the V4-V6 stage of corn growth increased cover crop biomass by 33% compared with the later sowing date of V10-V12. It is important to note that this study was done in corn seed production were where female plants were detasseled and male plants removed after polinization.

Noland et al. (2018) examined different sowing methods of cover crops into corn and silage corn including broadcasting, broadcasting with light incorporation, and direct sowing using high clearance equipment. Findings showed no reduction in yield with any method (Noland et al., 2018). The cover crops in this study, which were directly sown using a drill, had the highest biomass yield when compared with cover crops that were broadcasted (Noland et al., 2018). A study conducted with rye, radish, and a mixture of both were intersown in corn and fertilized in the fall before corn sowing and in the spring. The results indicated that corn grain yield was not reduced with the inclusion of cover crop treatments when fertilization was done in the fall; however, cover crops reduced corn grain yield by 7% when the fertilization was done in the spring (Ruffati et al., 2019).

While there is a large amount of research done looking at cover crops intersown into corn, limited research has been done looking at intersowing into soybean. Some research has been done broadcasting cover crops into soybean at different growth stages using radish (Sandler et al., 2015). Findings of this study indicate that radish broadcasted into standing soybean at different sowing dates did not reduce soybean yield when compared with a control without radish. This study also demonstrated how the environment can create variability in cover crop
establishment and biomass production. Radish biomass yield ranged from 25 kg ha\(^{-1}\) in a dry environment to 1488 kg ha\(^{-1}\) when adequate moisture was available (Sandler et al., 2015).

Ruffati et al. (2019) also examined broadcasting winter rye and radish into soybean showing no yield reduction in soybean grain yield when compared with a control without any cover crops present. Radish biomass produced a range of 145 to 807 kg ha\(^{-1}\) in the fall whereas rye produced 265 to 652 kg ha\(^{-1}\). Winter rye survived the winter in this study and produced a significantly higher biomass yield in the spring when compared with the rye fall growth ranging from 1034 to 2111 kg ha\(^{-1}\) (Ruffati et al., 2019).

Camelina intersown or broadcasted into soybean at multiple sowing dates was studied by Berti et al. (2017). Results indicated that sowing camelina on the same day as soybean was the only sowing date to reduce soybean grain yield by 9.5% when compared with the check without camelina (Berti et al., 2017). Results indicated that camelina performance as a cover crop did the best at the later sowing dates R1-R2 stage of soybean and when sown after soybean harvest. The V3-V4 soybean stage of intersowing camelina showed high competition from soybean and limited camelina growth and development throughout the experiment (Berti et al., 2017).
4. MATERIALS AND METHODS

4.1. Field establishment and experimental design

The experiments were conducted from 2016 to 2018 at two North Dakota State University (NDSU) experimental stations: Fargo, ND (46°89’ N, -96°82’ W, elevation 274 m) and Prosper, ND (46°58’ N, -97°3’ W, elevation 284 m). Soil type in Fargo is Fargo silty clay (Fine, smectitic, frigid Typic Epiaquerts) and soil type in Prosper is Kindred-Bearden silty clay loam (Kindred: Fine-silty, mixed, superactive Typic Endoaquoll; Bearden: Fine-silty, mixed, superactive, frigid Aeric Calciaquoll) (Soil Survey Staff, 2016). Daily temperature and rainfall were recorded by the North Dakota Agriculture Weather Network (NDAWN, 2016) at both sites (Fig. 1).

The previous crop in 2015 and 2016 in Fargo was oat and in Prosper was wheat. Fields were cultivated prior to sowing soybean. Baseline soil samples were taken before soybean sowing in 2016 and 2017 at both locations (Table 1). Soils samples were taken at the 0- to 15-cm depth and tested for soil pH, organic matter, P (Olsen, 1954), and K with the ammonium acetate method (Warncke and Brown, 1998) with a Buck Scientific Model 210 VGP Atomic Absorption Spectrophotometer (Buck Scientific, East Norwalk, CT). Soil sample analysis for NO$_3$-N was analyzed from 0- to 60-cm depth according to Vendrell and Zupancic (1990) method.

Table 1. Initial soil analysis for experimental sites Fargo and Prosper, ND, 2016 and 2017.

| Environment | pH † | OM | P | K | NO$_3$-N (0-15 cm-depth) | NO$_3$-N (15-60 cm-depth) |
|-------------|------|-----|---|---|--------------------------|--------------------------|
|             | g kg$^{-1}$ | mg kg$^{-1}$ | kg ha$^{-1}$ | kg ha$^{-1}$ |
| 2016 Fargo   | 7.8  | 60  | 20.3 | 398.0 | 45.8 | 90.0 |
| 2016 Prosper | 6.8  | 44  | 45.0 | 251.5 | 27.5 | 94.2 |
| 2017 Fargo   | 7.5  | 65  | 23.0 | 350.5 | 35.9 | 94.2 |
| 2017 Prosper | 6.8  | 41  | 56.0 | 348.0 | 30.3 | 117.7 |

† pH, organic matter (OM), P, and K were analyzed from 0- to 15-cm soil depth.
The experimental design was a randomized complete block design (RCBD) with a splitplot arrangement with four replicates. The main plot was soybean growth stage at which cover crops were sown, and the sub-plot was the cover crop treatment. Sub-plots consisted of four rows of soybean, each row spaced at 0.56-m apart, to total 2.24-m in width and each sub-plot was separated by 0.56-m apart. Sub-plots were 7.6-m in length at planting and reduced to 6.1-m in length at harvest. Increased experimental unit length during the growing season reduced the border effect on the cover crop treatments.

The soybean cultivar selected was glyphosate-tolerant, with a bush type architecture, relative maturity group 00.8, and soybean cyst nematode (*Heterodera glycines* T.) resistance. The bush type architecture was selected to allow the cover crops to grow under the canopy, and the soybean cyst nematode (SCN) resistance was selected due to the high SCN populations at the Prosper location. Soybean was sown on 18 May and 16 May 2016 in Fargo and Prosper, respectively, and on 11 May in Fargo and Prosper in 2017. The soybean was sown with a John Deere 1700 Maxemerge planter (John Deere, Moline, IL) at a row spacing of 0.56-m and a sowing depth of 2.5 to 3.8 cm for all sowing dates. Soybean sowing rate was 505,000 plants ha$^{-1}$ in order to reach the target plant population of 432,100 plants ha$^{-1}$

Four cover crops were selected to sow between the soybean rows: winter pea cv. Austrian, forage radish cv. Daikon, winter camelina cv. Joelle, winter rye cv. Rymin, a mixture of all four cover crops, and a check treatment without cover crops. Winter pea was chosen for prostate growth low to the ground. Winter camelina and winter rye were selected because winter biotypes will not bolt during the fall growth. Prostrate growth and not bolting will aid in harvesting the soybean. Radish was selected due to the benefits of the large tap root it produces. Cover crops were intersown at two later stages of soybean reproduction, the R4 and R6 stages.
Figure 1. Daily-rainfall, -maximum temperature, and -minimum temperature at Fargo and Prosper from April 2016 to November 2017. The R4 cover crop sowing date is represented with $\Delta$, and the R6 cover crop sowing date is represented with $\ominus$. 
In Fargo, R4 was sown on 25 July 2016 and 21 July 2017; R6 was sown on 16 August 2016 and 21 August 2017. In Prosper, R4 was sown on 26 July 2016 and 21 July 2017; R6 was sown on 16 August 2016 and 21 August 2017. The R4 and R6 growth stages were chosen in this study to see the cover crops response to limited light under the soybean canopy. At later growth stages, time to soybean senescence is shorter avoiding a long-term stress to cover crops due to limited light. As soon as soybean reaches physiological maturity leaves start falling off allowing light within the canopy which the cover crop can intercept and resume growth. Berti et al. (2017) demonstrated that cover crops broadcasted at soybean growth stages (R1-R2) had better stand establishment than earlier sowing dates, and a reason why later stages were chosen for this study.

The sowing rates for winter pea and forage radish were 89 and 5.6 kg ha\(^{-1}\), of pure live seed (PLS) respectively. Sowing rates for winter camelina and winter rye were 6.7, and 67.2 kg ha\(^{-1}\), PLS respectively. Winter pea and radish 1000-seed weight was 113.3 g and 14.8 g, respectively. Winter camelina and winter rye was 32.1 g and 0.9 g, respectively. The cover crop mix had one quarter the rate of each individual sowing rate. The same sowing rates were used for both sowing dates. All cover crops were intersown by hand using a modified V-shaped-hoe to create two furrows 15-cm apart centered within the 0.56-m soybean rows. Cover crop seed was placed by hand within the furrow at a depth of approximately 1.3-cm for all cover crops, and then covered with soil. Sowing by hand mimicked a new 15-cm high clearance twin-row drill (Amity Technology, Inc., Fargo, ND) that can intersow cover crops at later developmental stages of soybean.

Spring wheat followed the soybean to evaluate the effect of fall-sown cover crops on the succeeding crop. Wheat was sown no-till on 2 May 2017 at both locations, and then on 1 May and 15 May 2018 in Fargo and Prosper, respectively. Sowing was done using a Great Plains 15-
cm row space planter (Great Plains, Salinas, KS) at a target population or 3.7 million plants ha\(^{-1}\).
The spring wheat cultivar selected was Glenn with an average yield, moderately resistant to head scab \(\textit{Fusarium graminearum}\), and medium- to early-maturity. Spring wheat plots were 2.24-m wide and 6.1-m long, sown exactly where the cover crop treatments were the previous year.

Applications of glyphosate \([\text{N-(phosphonomethyl) glycine}]\) at 1.1 kg a.i. ha\(^{-1}\) were done prior to soybean sowing and post soybean emergence, and before first cover crop sowing date at both locations in 2016 and 2017. A pre-plant application of glyphosate was done, a day prior to sowing spring wheat in both 2017 and 2018, to eliminate any weeds, and cover crops that may have overwintered from 2016 and 2017 cover crop sowing. Spring wheat was not fertilized, assuming this was a better indicator to understand how efficient cover crop treatments are at nutrient cycling, if at all.

4.2. Field sampling and processing

Cover crop biomass was collected after soybean harvest at both Fargo and Prosper on 28 October in both 2016 and 2017. Cover crops that survived the winter were harvested in the following spring, on 17 April 2017 at both locations and on 24 April 2018 in Prosper. Spring biomass in 2018 at the Fargo location was not harvested due to loss of plants. Biomass samples were collected by hand clipping 0.09 m\(^{2}\) from each cover crop area growing between the 2-center soybean rows. All above ground biomass was collected right above soil level; however, this did not include radish above ground enlarged hypocotyl protruding above the soil level. The below ground portion of each cover crop was left undisturbed and not sampled due the difficulty in retrieving all below ground biomass uniformly for all cover crops. All cover crop biomass samples were dried at 70°C until a constant weight. Dried samples were then ground by a Model 4 cutting mill (Eberbach Corporation, Ann Arbor, MI) to pass through a 1-mm size sieve. To
obtain the N and P content, ground samples were analyzed using a XDS near-infrared (NIR) rapid content analyzer (Foss, Denmark). With the NIR analysis, nutrient uptake by cover crops can be calculated using the formula N or P content times dry matter yield. Cover crop soil cover was taken visually in the fall and the following spring on a percent scale from 0 cover to 100% cover.

Soybean grain yield was harvested from the two-center rows, at 6.1-m in length, from each experimental unit using a Winstersteiger Classic plot combine (Wintersteiger, Salt Lake City, UT) on 30 September and 29 September 2016, in Fargo and Prosper, respectively. Soybean yield was collected on 22 September 2017 in Fargo and on 4 November in Prosper using a Hege 125B plot combine (Wintersteiger, Pullman, WA). Different combines were used due to equipment availability. Spring wheat grain yield was collected from the center-six rows from each 6.1-m plot on 24 August 2017 at both locations and on 2 August and 8 August 2018 in Fargo and Prosper, respectively, using a Hege 125B plot combine. Soybean grain protein and oil as well as wheat grain protein content were determined by XDS NIR rapid content analyzer. Soybean yield, protein and oil content were corrected to 13% moisture and spring wheat yield and protein levels were corrected to 15%.

Soybean plant stand counts were recorded from the 2-center rows of each experimental unit, taking 1 linear meter from each row prior to harvest. One of the two linear meter counts was harvested by hand to calculate total biomass and harvest index. Three separate soybean plant heights from soil level to the top node were recorded and averaged at physiological maturity or R8 before harvesting (Kandel, 2019).

Soil samples were collected in the fall 2016 and 2017 at the time when cover crop biomass was sampled. Soil samples were also taken the subsequent spring 2017 and 2018 before
spring wheat was sown and again in late fall after spring wheat harvest (Table 2). Soil samples were collected in between the middlemost cover-crop twin row, staying at least 5-cm away from a cover crop plant. Two samples were taken from each plot to create a composite sample from the 0- to 60-cm depth and analyzed for NO$_3$-N content.

Light measurements were collected throughout the soybean growing season. Light measurements were obtained using an AccuPAR LP-80 ceptometer built by Decagon Devices, Inc. in the USA. The ceptometer measures photosynthetically active radiation (PAR) (Decagon Devices, 2015). Three measurements below and above soybean canopy taken parallel to the ground and centered and parallel between soybean rows within the 2-center rows in every experiment plot. Measurements were taken every other week until soybean plants started to senesce, and then weekly. Measurements were taken when the PAR was at the highest during the day between 1200 and 1600 h. To calculate intercepted PAR by the soybean canopy, the PAR measured within the canopy was subtracted from the PAR recorded above the canopy with the equation:

$$\text{Intercepted PAR} = \frac{\text{PAR above canopy} - \text{PAR below canopy}}{\text{PAR above canopy}} \times 100$$

| Location/year | Fall after cover crop harvest | Before spring wheat sowing | After spring wheat harvest |
|---------------|-------------------------------|----------------------------|---------------------------|
| 2016          |                               |                            |                           |
| Fargo         | 6 Nov                         | -                          | -                         |
| Prosper       | 21 Oct                        | -                          | -                         |
| 2017          |                               |                            |                           |
| Fargo         | 30 Oct                        | 17 May                     | 19 Oct                    |
| Prosper       | 30 Oct                        | 17 May                     | 17 Oct                    |
| 2018          |                               |                            |                           |
| Fargo         | -                             | 15 May                     | 1 Nov                     |
| Prosper       | -                             | 15 May                     | 6 Nov                     |
4.3. Statistical analysis

Statistical analysis was conducted using standard procedure for a randomized complete block design with a split-plot arrangement. Locations in each year were analyzed separately and tested for homogeneity of variance. Each location per year combination was defined as an environment and was considered a random effect and if environments were homogeneous they were combined. Different growth stages (sowing dates) and cover crops were considered fixed effects. Analysis of variance and mean comparison was conducted using the procedure MIXED (method= type3) of SAS; if $F$ test was significant at $P \leq 0.05$, mean separation was performed using least square means paired differences, this only for fixed main effects or interactions. For significant interactions with random effects, only one LSD value was calculated for all possible mean comparisons in the interaction, with the error mean square value and corresponding degrees of freedom.
5. RESULTS AND DISCUSSION

5.1. Cover crop biomass yield

The analysis of variance was significant for the cover crop by environment and environment by sowing date by cover crop interactions for fall cover crop biomass yield, but not significant for cover crop spring biomass yield (Table 3). The significant interaction was the result of radish producing the highest amount of biomass overall, at 3.04 Mg ha$^{-1}$ in Prosper 2016 at the R4 sowing date, while in other environments and sowing dates, radish had the lowest biomass yield of all cover crops (Table 4). This may have been due to the fact that radish is more vulnerable to water deficit near the soil surface after sowing. Although it was drilled, radish seed is very small, so lack of rainfall after drilling for two- or three-weeks limited emergence. This is similar to Sandler et al., (2015) findings, where lack of rain in the early parts of establishment led to decreased biomass yield in radish intersown into soybean.

Low biomass yield in Prosper 2017 may be explained by water deficit after emergence and then excess water in September, with a rain event totaling just over 110 mm of rainfall (Fig. 1). This large rain event caused saturated field conditions for a prolonged period.

Winter pea response was more stable across environments, producing significantly higher biomass yield than at least one other cover crop treatment in four of the sowing dates in the four environments tested; three sowing dates in R4 showed a potential for greater biomass at the R4 sowing date. This may be attributed to larger seed, deeper sowing depth, and perhaps greater tolerance to shade, but this has not been reported before (Table 4). Winter pea was intersown successfully into switchgrass (*Panicum virgatum* L.) producing up to 2.7 Mg ha$^{-1}$ in Oklahoma (Sutradhar et al., 2017). Fall-sown winter pea in Kansas was able to produce an average biomass of 622 kg ha$^{-1}$ over four years (Holman et al., 2018).
Table 3. Analysis of variance and mean squares for five cover crops (CC) and two sowing dates (SD) for fall and spring: cover crop biomass, N and P accumulation, and cover crop biomass nutrient content across four environments (Env), Fargo and Prosper, ND, 2016 to 2018.

| SOV       | CC biomass | CC N accumulation | CC P accumulation | CC N content | CC P content | SOV       | CC biomass | CC N accumulation | CC P accumulation | CC N content | CC P content |
|-----------|------------|-------------------|-------------------|--------------|--------------|-----------|------------|-------------------|-------------------|--------------|--------------|
| Env       | 3          | 3                 |                   |              |              | Rep(env)  | 12         | 12                | 8                 |              |              |
| SD        | 1          | 0.6               | 1                 | 1026         | 9.7          | 0.39      | 0.0010     | 1                 | 0.3               | 1171.2       | 8.0          | 0.003        | 0.0001       |
| Env x SD  | 2          | 1.4               | 2                 | 2075         | 35.9         | 0.25      | 0.0001     | 2                 | 0.5               | 420.8        | 16.4         | 0.260        | 0.0024       |
| Error (a) | 8          | 0.9               | 8                 | 1055         | 15.6         | 0.21      | 0.0020     | 7                 | 0.2               | 225.5        | 4.6          | 0.225        | 0.0013       |
| CC        | 4          | 0.8               | 4                 | 2081         | 10.0         | 2.21      | 0.0111     | 2                 | 2.1               | 1859.3       | 57.8         | 1.616        | 0.0168       |
| Env x CC  | 9          | 0.6*              | 8                 | 583          | 10.6         | 0.58*     | 0.0108*    | 2                 | 0.5               | 485.9        | 14.6*        | 0.380        | 0.0001       |
| SD x CC   | 3          | 1.2               | 3                 | 986          | 19.3         | 0.34      | 0.0021     | 1                 | 0.1               | 1.6          | 0.6          | 0.037        | 0.0002       |
| Env x SD  | 6          | 0.6*              | 6                 | 767          | 12.1         | 0.19      | 0.0015     | 1                 | 0.7               | 347.8        | 15.3         | 0.494        | 0.0003       |
| Error (b) | 49         | 0.2               | 42                | 378          | 5.7          | 0.17      | 0.0016     | 13                | 0.2               | 261.5        | 3.8          | 0.084        | 0.0008       |
| CV, %     | 33.0       | 32.7              | 34.9              | 9.79         | 8.3465       | 29.9      | 29.3       | 29.9              | 6.5               | 5.7          |              |              |

* Significant at 0.05 probability level.
† Spring samples were only collected from three environments Fargo 2017 and Prosper 2017 and 2018.

Table 4. Mean fall cover crop biomass yield of two sowing dates (R4, R6) in four environments, Fargo and Prosper, ND, 2016-2017. Mean spring biomass averaged across sowing dates and environments.to 2018.

| Cover crop       | Fall R4 2016 | Fall R6 2016 | Fall R4 2017 | Fall R6 2017 | Spring† |
|------------------|--------------|--------------|--------------|--------------|---------|
| Winter camelina  | -            | -            | -            | -            | 0.73    |
| Winter pea       | 2.04 bc†     | 1.60 bc†     | 2.13 b       | 1.58 bc      | 1.13 cd | 1.17 cd |
| Radish           | 0.58 d       | 1.35 c       | 1.02 cd      | 3.04 a       | 1.52 bc | 1.00 cd | 1.40 bc |
| Winter rye       | 1.53 bc      | 0.57 d       | 0.97 cd      | 1.02 cd      | 0.82 cd | 1.28 cd | 0.72 cd |
| Mix              | 1.54 bc      | 0.96 cd      | 1.56 bc      | 1.53 bc      | 1.04 cd | 2.03 bc | 0.94 cd |

LSD(0.05) NS

† Fall means with different lowercase letter significantly different at $P < 0.05$ by the least square means test.
‡ Spring biomass means averaged across cover crop sowing dates and three environments Fargo 2017 and Prosper 2017 and 2018.
Although a winter annual in some environments, winter pea does not survive winters in North Dakota. Research in Kansas and states in the Pacific Northwest have shown winter peas to survive the winter and resume growth in the spring (Holman et al., 2018; Chen et al., 2006). Favorable conditions did not occur for winter camelina to establish in Fargo and Prosper 2016; no data was recorded. In Fargo and Prosper 2017, at the R4 and R6, respectively, winter camelina did establish and produce recordable biomass yield (Table 4) However, without adequate moisture after germination, winter camelina struggles to survive while under soybean (Berti et al., 2017).

Research also shows that larger camelina seed size attributes to faster emergence compared with smaller camelina seeds (Enjalbert et al., 2013; Zanetti et al., 2017). Other research has shown establishment of cover crops without moisture following sowing leads to a decrease in establishment and lower winter survival rate (Fisher et al., 2011; Wilson et al., 2013). Winter camelina and winter rye did not produce as high amount of biomass as winter pea, due to the fact that both species are winter annuals requiring vernalization to induce reproductive stage which limits fall growth.

The following spring, winter rye produced the highest amount of biomass of 1.74 Mg ha\(^{-1}\) (Table 4). These results for spring biomass are similar to other intersowing experiments including winter rye (Appelgate et al., 2017; Noland et al., 2018). Winter pea and radish did not survive the winter, so there was no recorded biomass in the spring. The cover crop mix mean averaged across four environments and two sowing dates was 0.94 Mg ha\(^{-1}\). The mix only consisted of surviving winter camelina and winter rye plants. Winter camelina biomass yield in the spring was 0.73 Mg ha\(^{-1}\) (Table 4). Winter camelina was able to survive winter where it established in the fall; this is similar to other work done in the Midwest (Gesch and Archer,
2013; Gesch et al., 2014). Other researchers have shown winter camelina to produce similar spring biomass yields as those observed in this study (Berti et al., 2017; Appelgate et al., 2017).

5.2. Cover crop nitrogen and phosphorus accumulation

The combined analysis of variance across all environments and sowing dates showed no difference among treatments for cover crop N or P accumulation in the fall ($P < 0.05$) (Table 3). The N accumulation in the above ground biomass in the fall ranged from 28.7- to 73.2-kg N ha$^{-1}$ (Table 5). The wide range of N accumulation is a reflection on biomass produced in the fall (Table 4). Previous researchers have looked at N accumulation of winter annuals; in the following spring, intersown winter rye accumulated 21.2 kg N ha$^{-1}$ and 21.7 to 26 kg N ha$^{-1}$ in studies by Applegate et al., (2017) and Noland et al., (2018), respectively.

In Berti et al., (2017), winter camelina intersown into corn and soybean accumulated 24 to 55 kg N ha$^{-1}$ in the spring. Other research that focused on radish intersown into soybean had N accumulations of 36.4 kg N ha$^{-1}$. The low amount was explained by dry weather in the fall (Ruffatti et al., 2019). Winter pea intersown into switchgrass had N accumulation of 42.1 kg N ha$^{-1}$ (Sutradhar et al., 2017). The results indicate when cover crops are well established into soybean, an acquisition of large amounts of N is present in the biomass, reducing the potential offsite dispersion of free N in the soil.

The P accumulation in the above ground biomass in the fall ranged from 4.9 to 8.6 kg ha$^{-1}$. This range is also a reflection of the biomass yield in Table 4. These results indicate that the cover crops used in this experiment are not as efficient at acquiring P as they are at taking up N which is similar to other researchers (Miller et al., 1994; Wendling et al., 2016).
Table 5. Mean fall and spring N and P cover crop biomass accumulation averaged across two sowing dates and three environments, at Fargo and Prosper, ND.

| Cover crop     | Fall N | P  | Spring‡ N | P  |
|----------------|--------|----|-----------|----|
| Winter camelina| 28.7   | 4.9| 36.4      | 4.9|
| Winter pea     | 71.5   | 7.4| -         | -  |
| Radish         | 73.2   | 8.6| -         | -  |
| Winter rye     | 47.2   | 6.2| 69.1      | 8.7|
| Mix            | 55.9   | 6.2| 43.2      | 4.4|
| LSD (0.05)     | NS     | NS | NS        | NS |

‡ Spring biomass only accounts for cover crops that survived the winter combined across three environments: Fargo 2017, Prosper 2017 and 2018, and two cover crop sowing dates.

5.3. Cover crop nutrient content

The combined analysis of variance across all environments and sowing dates showed no difference among treatments for cover crop N content in the fall, and N and P content in the spring (Table 3). The cover crop by environment interaction was significant for both N and P content (data not shown).

Nitrogen content in the fall was combined across four environments whereas fall P and spring N, and P were combined only across three environments. Fall N and P ranged from 23 to 44 g kg⁻¹ and 4.2 to 5.4 g kg⁻¹ respectively, whereas N and P in the spring had slightly higher content ranging from 42 to 50 and 4.9 to 6.9 g kg⁻¹, respectively (Table 6). Much of the research done has looked at N content in cover crops due to the higher chance of leaching compared with P where run off and erosion is a higher risk of offsite movement. Nitrogen and P content in the biomass in this study were similar to other research (Miller et al., 1994; Wendling et al., 2016).
Table 6. Mean fall and spring biomass nutrient content averaged across two sowing dates and four environments, Fargo and Prosper, ND, 2016 to 2018.

| Cover crop     | Fall N  | P   | Spring P | N  | P   |
|----------------|---------|-----|----------|----|-----|
| Winter camelina| 23      | 4.2 | 50       |    | 6.9 |
| Winter pea     | 43      | 4.4 | -        |    | -   |
| Radish         | 44      | 4.8 | -        |    | -   |
| Winter rye     | 42      | 5.4 | 42       |    | 5.1 |
| Mix            | 43      | 4.7 | 47       |    | 4.9 |
| LSD (0.05)     | NS      | NS  | NS       |    | NS  |

† Spring biomass only accounts for cover crops that survived the winter averaged across three environments, Fargo 2017, Prosper 2017 and 2018, and two cover crop sowing dates.

5.4. Cover crop soil coverage

The analysis of variance was significant for the cover crop main effect, cover crop by environment, and environment by sowing date by cover crop interactions for fall cover crop soil coverage, but not significant for spring cover crop soil coverage (Table 7). The significant interaction is the result of winter pea providing the highest soil coverage, at 78% soil cover in Prosper 2016 at both the R4 and R6 sowing dates, while in different environments and dates winter pea had the lowest soil cover (Table 8). Low soil cover in Prosper 2017 at the R4 sowing date can be explained by limited rainfall in the immediate weeks after sowing (Fig. 1) which likely caused water deficiency limiting establishment and posterior growth. These results are a direct reflection of biomass produced; if cover crops had low biomass yield (Table 4) it also has a low soil cover (Table 8).

The winter pea chosen for this study has a growth pattern different than that of a field pea, where winter pea grows closer to the ground in a prostrate manner. This growth pattern along with high biomass production seen in Table 4 can explain why winter pea had the highest soil cover.
Table 7. Analysis of variance and mean squares for five cover crops (CC) and two sowing dates (SD) for fall and spring cover crop soil cover in four environments (Env) at Fargo and Prosper, ND, conducted over 2016 to 2018.

| SOV            | df | Fall soil cover | df | Spring soil cover† |
|----------------|----|-----------------|----|-------------------|
| Env            | 3  | 3431.2          | 2  | 658.4             |
| Rep(env)       | 12 | 128.8           | 8  | 242.9             |
| SD             | 1  | 448.9           | 1  | 19.5              |
| Env x SD       | 3  | 465.3           | 2  | 709.3*            |
| Error (a)      | 8  | 491.0           | 7  | 126.7             |
| CC             | 4  | 2771.0*         | 2  | 1812.9            |
| Env x CC       | 9  | 393.5*          | 2  | 575.5*            |
| SD x CC        | 3  | 988.9           | 1  | 46.2              |
| Env x SD x CC  | 6  | 447.7*          | 1  | 934.6             |
| Error (b)      | 49 | 157.0           | 13 | 101.1             |
| CV, %          |    | 37.7            |    | 39.1              |

* Significant at 0.05 probability level.
† Spring soil cover only accounts for cover crops that survived the winter averaged across three environments: Fargo 2017, Prosper 2017 and 2018, and two sowing dates.

In the fall winter rye provided the next highest coverage at 55% soil cover averaged across environments. The mixed-cover crops ranged from 14 to 45% cover in the fall. Radish and camelina were among the lowest at providing soil cover in the fall. Radish only provided 5% coverage at the R4 stage in Fargo and Prosper 2016 and in R6 Fargo 2017 (Table 8). These results indicate radish intersown by itself may not provide adequate soil cover. Although, radish trended to provide more cover at the R6 sowing date versus the R4 averaged across environments.

Radish did have the highest biomass in R6 Prosper 2016 (3.04 Mg ha⁻¹) (Table 4), but only had 34% cover in that same environment and sowing date (Table 8). This is still significantly lower than winter pea in three environments; Fargo 2016 R4 sowing date and Prosper 2016, R4 and R6 sowing dates (Table 8).
Table 8. Mean fall and spring cover crop soil coverage at Fargo and Prosper, ND over 2016 to 2018.

| Cover crop       |          |          |          |          |          |          |
|------------------|----------|----------|----------|----------|----------|----------|
|                  | Fall     |          |          | Spring‡ |          |          |
|                  | Fargo 16 | Prosper 16 | Fargo 17 | Prosper 17 |
|                  | R4  R6   | R4  R6   | R4  R6   | R4  R6   |          |          |
| Winter camelina  | -        | -        | -        | 16 d     | 18 d     | -        |
| Winter pea       | 63 b†    | 58 bc    | 78 a     | 31 cd    | 5 d      | 50 bc    |
| Radish           | 5 d      | 15 d     | 5 d      | 34 cd    | 5 d      | 12 d     |
| Winter rye       | 34 cd    | 8 d      | 40 c     | 55 bc    | 13 d     | 40 c     |
| Mix              | 33 cd    | 21 d     | 40 c     | 45 bc    | 14 d     | 33 cd    |
|                  |          |          |          |          |          |          |
|                  |          |          |          | 13.4     |          |          |
|                  |          |          |          | NS       |          |          |

† Means with different lowercase letter significantly different at \( P < 0.05 \) by the least square means test.
‡ Spring soil cover only accounts for cover crops that survived the winter averaged across two cover crop sowing dates and three environments: Fargo 2017, Prosper 2017 and 2018.

This apparent disparity can be explained by the erect growth pattern of radish. Plants bolted and were about 60-cm tall at the end of the fall (data not shown) leaving gaps of uncovered soil between plants but still amounting to high biomass yield. Winter camelina provided some of the lower amounts of soil cover at 16% and 18% in Fargo 2017, R6 sowing and Prosper 2017, R4 sowing, respectively. The lower cover can be explained due to camelina not having a large amount of leaves to cover the ground in fall. Winter rye and the mix were similar in coverage where winter rye ranged from 5 to 55% and the mix ranged from 14 to 45%.

Spring soil cover show winter rye had the greatest cover at 39.2% and winter camelina had the lowest at 13.4% and the mix had 15% soil cover (Table 8), but they were not significantly different from one another. Similar results were reported in which camelina intersown in soybean in the previous year provided spring cover that ranged from 7-13% (Berti et al., 2017).
5.5. Soybean and spring wheat yield

The combined analysis of variance across all environments and sowing dates showed no differences among treatments for soybean grain yield; however, spring wheat yield was different among treatments ($P \leq 0.05$) (Table 9).

The results indicate intersowing cover crops at the R4 and R6 stages of soybean growth may be a potential time to intersow without impacting soybean grain yield (Table 10). Intersowing at later stages of soybean development may allow for greater advantage for the soybean over intersown cover crops. With the soybean already being established, cover crop growth is reduced due to limited incident solar radiation. Berti et al., (2017) found similar results when intersowing winter camelina into R1 and R2 without reducing soybean grain yield, but did see a yield reduction in soybean yield when winter camelina was sown the same day as the cash crop. In other studies, intersowing cover crops into soybean did not reduce soybean grain yield (Sandler et al., 2015; Ruffatti et al., 2019). One factor that could contribute to the non-reduction in soybean grain yield is the fact that soybean out competes the cover crops, since they are sown after the soybean critical period for competition. This has been proven through research of weed competition in soybean (Datta et al., 2017).

When compared with all the other treatments, including the check, spring wheat yield was significantly lower in plots with winter camelina and winter rye preceding wheat (Table 10). As winter camelina and winter rye survived the winter and resumed growth in the spring, these cover crops also acquired nutrients and water before wheat was sown. This, in turn, likely impacted the amount of available water for subsequent spring wheat growth, hindering development and decreasing yield. Krueger et al., (2011) found that winter rye terminated too close to corn sowing led to decreased soil water and crop yield.
Previous research has shown winter rye produces allelopathic compounds that reduces grasses growth, which can affect wheat (Moyer and Blackshaw, 2009) and corn (Krueger et al., 2011). Allelopathic compounds causing yield reduction may not have be the cause of yield reduction, due to the mix treatment having winter camelina and winter rye and did not show a reduction on the subsequent spring wheat yield. In addition, winter rye can keep the cycle of root diseases which can also contribute to yield decrease in corn (Bakker et al., 2016; Acharya et al., 2017). Moreover, reduction in soil N supply through N immobilization can negatively impact spring wheat yield following winter rye (Thomas et al., 2017).
Table 9. Analysis of variance and mean squares for five cover crop (CC) and two sowing dates (SD) for soybean grain yield, harvest index (HI), plant height at harvest, plant population, grain protein, and grain oil, and spring wheat grain yield and protein in four environments (Env), Fargo and Prosper, ND, 2016 to 2018.

| SOV            | df | Grain yield | HI    | Plant height | Plant population (x 10^6) | Grain protein | Grain oil | Spring wheat Yield | Grain protein |
|----------------|----|-------------|-------|--------------|----------------------------|---------------|-----------|-------------------|--------------|
| Env            | 3  | 915         | 0.0005| 0.0017       | 435                        | 0.75          | 0.04      | 120               | 0.12         |
| Rep(env)       | 12 | 138251      | 0.0005| 0.0022       | 783                        | 0.57          | 0.13      | 341442            | 0.77         |
| SD             | 1  | 245321      | 0.0006| 0.0058       | 823                        | 0.81          | 0.37      | 360821            | 1.61         |
| Env x SD       | 3  | 86955       | 0.0008| 0.0009       | 358                        | 0.09          | 0.04      | 663396*           | 0.45*        |
| Error (a)      | 8  | 29123       | 0.0008| 0.0045*      | 269                        | 0.16          | 0.03      | 181466            | 0.46         |
| CC             | 5  | 29629       | 0.0003| 0.0005       | 445                        | 0.33          | 0.08      | 81480             | 0.41         |
| Env x CC       | 12 | 104219      | 0.0003| 0.0027       | 257                        | 0.25          | 0.07      | 111508            | 0.47         |
| SD x CC        | 3  | 62146       | 0.0007| 0.0015       | 330                        | 0.27          | 0.06      | 141064            | 0.47         |
| Env x SD x CC  | 6  | 8.4         | 4.6   | 5.3          | 10                         | 1.52          | 1.33      | 15                | 5.02         |

* Significant at 0.05 probability level.
Table 10. Mean soybean and spring wheat grain yield for five cover crops and a no cover crop check averaged across two cover crop sowing dates and four environments at Fargo and Prosper, ND, from 2016 to 2018.

| Cover crop   | Soybean grain yield kg ha⁻¹ | Wheat grain yield kg ha⁻¹ |
|--------------|-----------------------------|---------------------------|
| Winter camelina | 2933                        | 2144                      |
| Winter pea   | 3008                        | 2708                      |
| Radish       | 3025                        | 2812                      |
| Winter rye   | 3025                        | 2174                      |
| Mix          | 2953                        | 2691                      |
| Check        | 2908                        | 2684                      |
| LSD (0.05)   | NS                          | 315                       |

5.6. Soybean and spring wheat characteristics

The combined analysis of variance across all environments and sowing dates showed no differences among treatments for soybean harvest index, plant height, plant count, and grain oil and protein content at the \((P \leq 0.05)\) (Table 9). However, the combined analysis of variance across all environments and sowing dates showed differences among cover crop treatments for spring wheat grain yield and protein content at the \((P \leq 0.05)\) (Table 9).

Results indicate that cover crops intersown into soybean does not affect any of the soybean plant characteristics measured in this study (Table 11). This also indicates the competition from the soybean is enough to keep the cover crops subdued until after harvest. Results were similar to studies done when intersowing camelina into soybeans at different sowing dates. Soybean oil or protein were unaffected by competition from intersown cover crops (Berti et al., 2017; Sadler et al., 2015).

Results show spring wheat protein levels were significantly higher for spring wheat that followed winter rye at 139 g kg⁻¹, compared with the other cover crops and the check treatment (Table 11). This can be explained by lower yields in wheat typically will have a higher protein value at the end of a season due to a concentration effect. Although not significant the trend of
lower yield and higher protein was also seen for spring wheat following winter camelina (Table 11).

Table 11. Mean soybean height, plant population (pop.), harvest index (HI), protein, oil, and spring wheat protein content for five cover crops and a no cover crop check averaged across two cover crop sowing dates and four environments at Fargo and Prosper, ND, from 2016 to 2018.

| Cover crop       | Height m | Plant pop. Plants x 10^6 ha⁻¹ | Soybean Protein g kg⁻¹ | Winter Wheat Protein g kg⁻¹ |
|------------------|----------|--------------------------------|------------------------|-----------------------------|
| Winter camelina  | 0.66     | 41.2                           | 57                     | 342                         | 177                         | 137.0                      |
| Winter pea       | 0.76     | 38.9                           | 57                     | 340                         | 184                         | 136.7                      |
| Radish           | 0.76     | 39.7                           | 57                     | 341                         | 183                         | 133.8                      |
| Winter rye       | 0.77     | 40.6                           | 56                     | 342                         | 184                         | 139.1                      |
| Mix              | 0.74     | 40.3                           | 57                     | 343                         | 182                         | 136.8                      |
| Check            | 0.73     | 39.1                           | 57                     | 340                         | 182                         | 133.6                      |
| LSD (0.05)       | NS       | NS                             | NS                     | NS                          | NS                          | 5.4                        |

5.7. Soil nitrate removal and replacement

The combined analysis of variance showed significance for the environment by sowing date by cover crop interaction for soil NO₃⁻-N in the fall (Table 12). The largest amount of residual soil NO₃⁻-N, 61.7 kg ha⁻¹, was seen in Prosper 2016 in the check plot without any cover crops (Table 13). The lowest soil NO₃⁻-N levels were seen with the mix, winter pea, and winter rye at 15.5, 20, and 20 kg ha⁻¹, respectively. The significant reduction in soil NO₃⁻-N in the cover crop plots can be related to the cover crop biomass N accumulation in Table 4. These cover crops show they have potential to scavenge and retain excess residual NO₃⁻-N. Previous research using winter rye as a cover crop was able to significantly reduce tile drainage discharge of NO₃⁻-N loads by 63% (Kaspar et al., 2007) and rye and annual ryegrass in a mix reduced discharge by 69-90% (Hanrahan et al., 2018). Winter rye has the ability to scavenge as much as 28 to 56 kg N ha⁻¹ (Chatterjee and Clay, 2016).

However, the analysis of variance combined across four environments and two sowing dates showed no significant difference for soil NO₃⁻-N in the spring before wheat sowing and
following wheat harvest (Table 12). The results show soil NO$_3$-N levels before wheat sowing were similar between the check compared with the plots with cover crops (Table 14), although the winter crops were numerically lower than the winter-killed cover crops.

Table 12. Analysis of variance and mean squares for five cover crop (CC) and two sowing dates (SD) for soil NO$_3$-N in the fall, spring before spring wheat, and after wheat harvest at Fargo and Prosper, ND from 2016 to 2018.

| SOV          | df  | Fall NO$_3$-N$^\dagger$ | df  | Spring NO$_3$-N$^\ddagger$ | After wheat NO$_3$-N |
|--------------|-----|--------------------------|-----|-----------------------------|----------------------|
| Env          | 2   |                          | 3   |                             |                      |
| Rep(env)     | 9   |                          | 12  |                             |                      |
| SD           | 1   | 26.4                     | 1   | 1.4                         | 326.5                |
| Env x SD     | 1   | 407.5                    | 2   | 1399.8                      | 158.7                |
| Error (a)    | 5   | 432.8                    | 8   | 511.8                       | 275.1                |
| CC           | 5   | 613.5                    | 5   | 886.4                       | 27.3                 |
| Env x CC     | 8   | 405.0                    | 12  | 595.4                       | 140.1                |
| SD x CC      | 3   | 44.6                     | 3   | 198.8                       | 64.6                 |
| Env x SD x CC| 2   | 434.5*                   | 4   | 324.9                       | 264.8                |
| Error (b)    | 40  | 93.7                     | 57  | 471.7                       | 166.5                |
| CV, %        |     | 32.0                     |     | 35.6                        | 31.7                 |

* Significant at 0.05 probability level.
† Fall soil NO$_3$-N were averaged across three environments Fargo 2017 and Prosper 2016 and 2017. This was due to excess moisture in the fall in Fargo 2016 which unable to obtain samples.
‡ Spring soil NO$_3$-N averaged across four environments.

This is indicating that maybe cover crops, while able to scavenge soil NO$_3$-N, may not decompose and release the N in the biomass in a timely manner explaining the lack of significance observed. Even after wheat harvest, there were still no differences observed in soil NO$_3$-N levels (Table 14). The expected results were to see an increase of soil NO$_3$-N in cover crop treatments due to the cycling of N in the cover crop biomass from the previous growing season. These results are similar to a study done by Cicek et al., (2015) where radish biomass did not release the N fast enough to supply a subsequent wheat crop.
Table 13. Mean fall soil NO$_3$N from 0- to 60-cm in depth in Fargo and Prosper, ND, in 2016 and 2017.

| Cover crop | —Fargo 2016— | —Prosper 2016— | —Fargo 2017— | —Prosper 2017— |
|------------|--------------|----------------|--------------|----------------|
|            | R4           | R6             | R4           | R6             | R4           | R6             | R4           | R6             |
| Winter camelina | -            | -              | -            | -              | 20.7 b       | 32.0 ab       | -            |                |
| Winter pea  | -            | -              | 20.0 b†      | 23.7 b         | 32.5 ab       | 50.0 ab       | 27.0 ab      |                |
| Radish      | -            | -              | 26.0 ab      | -              | 34.5 ab       | 20.0 b        | 21.0 b       |                |
| Winter rye  | -            | -              | 31.3 ab      | 21.5 b         | 28.0 ab       | 26.7 ab       | 40.0 ab      |                |
| Mix         | -            | -              | 36.3 ab      | 22.8 b         | 28.5 ab       | 15.5 b        | 29.3 ab      |                |
| Check       | —            | —              | 61.7† a      | —              | 48.2 ab       | —              | 31.0 ab      | —              |

† Means with different lowercase letter significantly different at $P < 0.05$ by the least square means test.
‡ Values are compared only within each environment for both sowing dates.
Table 14. Mean soil NO$_3$-N levels in the spring before spring wheat sowing, and after spring wheat harvest for five cover crop treatments averaged across two cover crop sowing dates and four environments, Fargo and Prosper, ND, from 2017 to 2018.

| Cover crop   | Before wheat sowing | After wheat harvest |
|--------------|---------------------|---------------------|
| Winter camelina | 53.7 kg ha$^{-1}$  | 13.8 kg ha$^{-1}$  |
| Winter pea   | 65.8 kg ha$^{-1}$  | 43.8 kg ha$^{-1}$  |
| Radish       | 60.1 kg ha$^{-1}$  | 37.1 kg ha$^{-1}$  |
| Winter rye   | 44.1 kg ha$^{-1}$  | 48.3 kg ha$^{-1}$  |
| Mix          | 65.2 kg ha$^{-1}$  | 43.9 kg ha$^{-1}$  |
| Check        | 73.7 kg ha$^{-1}$  | 33.0 kg ha$^{-1}$  |
| LSD (0.05)   | NS                  | NS                  |

NO$_3$-N soil samples are totals from 0- to 60-cm depth

5.8. Photosynthetically active radiation (PAR) beneath soybean canopy

In many studies, limited photosynthetically active radiation has shown to influence cover crop by either suppression or failure of the cover crop growth (Wilson et al., 2013; Belfry and Van Eerd, 2016). A trend line of intercepted PAR by soybean canopy combined across four environments is shown in Fig. 2. In the R4 sowing date, which happened on mid-July at both locations, cover crops were sown before soybean had reached full canopy, however interception was already above 70% of above canopy PAR. The percent PAR intercepted by soybean canopy kept increasing until mid-August. These results show that cover crops had limited PAR that reached the soil to initiate growth, but by the first week of August, PAR interception had reached its maximum at above 80%. Cover crops at the R4 sowing date where then subjected to grow in very limited PAR for three weeks, which made plants etiolated and weak.

In 2017, in Fargo, cover crops own at R4 emerged in almost 100% (data not shown) due to timely rain events of greater than 25 mm per event, one week and two weeks after being sown (Fig. 1). After 11 August, significant rain (>5-mm/event) did not occur until mid-September causing severe water stress to recently emerged cover crops. By 25 August, soybean plants started to drop their leaves increasing the solar radiation reaching the inter-row to about 90%
PAR by 12 September (Fig. 3). This combination of water stress and exposure to almost full solar radiation literally desiccated the cover crops, hence no cover or biomass was recorded at the end of the season this sowing date. In addition, camelina plants had acclimated to low light conditions of less than 20% PAR, but soybean leaf drop happened rapidly in about 10 days which probably did not allow cover crop plants to adapt to higher incident PAR. Excess radiation would then had been converted into heat explaining the desiccation and death of all emerged seedlings.

Cover crops were sown in R6 the second and third week of August at the time where PAR intercepted by soybean canopy was the highest. The cover crops that germinated where subjected to low light conditions for only about a week. In Fargo, 2017 the cover crops sown in R6 did not receive rain until 12 September, most of them emerging after this rain event when soybean had already dropped almost all their leaves promoting growth, hence some coverage and biomass was observed at R6 at this environment. Averaged across all environments, by around the 25 August, PAR interception began to decrease rapidly (Fig. 2). By the second week of September, PAR interception was less than 10%.
Figure 2. Photosynthetic active radiation (PAR) interception of soybean canopy intersown with cover crops. Mean values were averaged across environments and sowing dates in Fargo and Prosper, ND, 2016 to 2017. R4 and R6 sowing in Fargo and Prosper are depicted as ← and ---, respectively in 2016. R4 and R6 sowing in Fargo and Prosper are depicted as ← and ---, respectively in 2017. Soybean harvest is shown as ▼.
6. CONCLUSIONS

Winter pea established well when sown at either the R4 or R6 stage of soybean development. Radish, when established in suitable growing conditions (rainfall after sowing), was able to produce over 3 Mg ha\(^{-1}\) amounts of biomass. Winter rye established well but tended to have lower fall biomass yield than pea and radish. Camelina struggled to survive under the soybean canopy, but if there was timely rainfall after sowing establishment was good.

Nutrient content of the cover crop treatments was not significantly different from one another and this was the same for overall nutrient accumulation in total biomass. Winter pea provided the highest soil cover out of all the cover crop treatments. When cover crops were sown into soybean at later reproductive stages, soybean outcompeted the cover crops and reduced the chances of soybean grain yield lag. This experiment demonstrated that intersown cover crops did not reduce soybean yield. Competition from soybean cover crops did not affect soybean plant height, harvest index, plant population, or oil content of the seed.

The cover crops were able to scavenge excess nitrates in the soil profile and accumulate them in their biomass. The N in the cover crops was not returned in time for the following spring wheat crop, and if not managed correctly, the negative impacts of decreased available water and allelopathy compounds from cover crops surviving the winter can lead to a wheat grain yield loss.
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