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Intersowing Cover Crops into Standing Soybean in the US Upper Midwest

Alan T. Peterson, Marisol T. Berti * and Dulan Samarappuli

Department of Plant Sciences, North Dakota State Univ., Fargo, ND 58104, USA; Alan.Peterson.2@ndsu.edu (A.T.P.); Dulan.Samarappuli@ndsu.edu (D.S.)
* Correspondence: Marisol.Berti@ndsu.edu; Tel.: 001-701-231-6110

Received: 21 March 2019; Accepted: 23 May 2019; Published: 25 May 2019

Abstract: Nutrient losses and soil erosion after soybean (*Glycine max* (L.) Merr.) harvest are common in the US Upper Midwest. Cover crops need to provide adequate growth and cover to prevent soil degradation throughout the winter and early spring months. The objective of this study was to determine the establishment of intersown cover crops and their impacts on a soybean-wheat rotation. Four cover crops—winter camelina (*Camelina sativa* (L.) Crantz), winter pea (*Pisum sativum* ssp. *arvense* (L.) Poir), winter rye (*Secale cereale* L.), and radish (*Raphanus sativus* L.)—were directly sown at the R4 and R6 stages of soybean at two locations, Prosper and Fargo, ND in 2016–2017. Cover crops above ground biomass in the fall ranged from 0.4 to 3.0 Mg ha^{-1} and N accumulation ranged from 28.7 to 73.2 kg ha^{-1}. Winter camelina and winter rye reduced subsequent spring wheat yield compared with the no cover crop treatment. Fall soil residual NO_3-N levels were lowest where cover crops were sown compared with the check. Spring NO_3-N levels were lowest in winter camelina and winter rye compared with all the other cover crops and the check. Results indicated intersowing cover crops have no impact on soybean yield, and show potential to mitigate soil nitrate losses in areas that grow soybean as a cash crop.

Keywords: winter camelina; winter pea; radish; winter rye; intersowing; nitrogen-accumulation: cover crop; soil nitrate

1. Introduction

Global food security depends on world food supply, which comes mainly from rainfed productions areas in temperate climates. The USA is the world's leading producer of many crops, including maize (*Zea mays* L.) and soybean. Most of these crops are grown using conventional tillage in areas where the soil is left exposed with no protection during the winter months, which can lead to soil erosion, by wind and water. If land productivity decreases, producers must increase inputs to acquire enough production to supply food, feed, and fiber for a growing global population [1].

The lack of soil coverage with left over plant biomass, or "residue", following a soybean harvest across the US Upper Midwest is a concern. Soybean does not produce adequate amounts of residue to cover and protect the soil from erosion especially when fall and winter precipitation in the form of snow is low. Soybean has been shown to lose up to 68% of its total biomass within 32 d after harvest [2]. Without adequate soil coverage, soil and nutrients are lost due to wind and water erosion. Precious lost topsoil will not be recoverable in the near future. It is estimated that left unprotected, topsoil losses can be anywhere from 6 Mg ha^{-1} to 18 Mg ha^{-1} annually [2]. If some course of action is not taken soon, soil losses will diminish land value, productivity, and sustainability [3].

Most research on cover crop intersowing or sowing after soybean has been done with rye [4]. This cover crop has proven to have many advantages. However, in the Upper Midwest many growers also grow wheat or other cereals after soybean in which sequence rye is not an alternative. Rye is difficult to terminate early in the season, since temperatures are low and many herbicides will not completely kill the rye. Any rye plants surviving in the wheat crop will be a grain contaminant, which will be docked at harvest, hurting the grower's profits.

In the Upper Midwest, soybean harvest starts in late September and continues into November. Cover crops sown following a soybean harvest have a very limited time to grow and provide cover before the first killing frost. Typically, in this region, the average first light frost (<0 °C) occurs by 20 September; however, this frost usually is not strong enough to kill cool-season cover crops, which resume growth when the temperature is above 0 °C. The first killing frost (<−6 °C) occurs by mid- to end-October, but varies from year to year. This would allow about 60–70 days of growth to cover crops intersown by mid-August. Thus, the alternative may lie in direct intersowing or broadcasting of cover crops into standing soybean.

There are several different strategies available for sowing a cover crop. Cover crops can be sown using a drill, broadcasted onto the soil surface, or broadcasted and incorporated into the soil. The method used to sow the cover crops will depend on the available equipment and the desired application timing. Cover crops can be sown with a drill before the main crop is planted, with the main crop during it being sown, or after the main crop is harvested. The advantages of drilling are that it requires up to 50% less seed and results in faster, more uniform emergence compared with broadcast sowing [5,6]. Cover crop stand establishment can also be up to 50% greater when cover crops are drilled into the soil rather than broadcasted, even if some form of incorporation is used afterward [6,7].

When sowing with a drill is not feasible, broadcast sowing cover crops is still a viable option [8]. The main advantage of broadcast applications over sowing with a drill is that it is not restricted by the main crop's height and can be done at almost any time during the season. This is accomplished either aerially by plane, with high-clearance equipment for in-season establishment, or with spreading equipment after harvest. The main disadvantage of surface broadcasting is it leaves the seed exposed to the hot, dry environment at the soil surface and predation by insects, birds, and rodents [8,9]. Successful germination and seedling establishment depends largely on whether or not the cover crop receives adequate precipitation within about a week of sowing [9]. Including some form of light soil incorporation, broadcast sowing can protect the seeds from predators and desiccation, but this may be difficult to accomplish while the main crop is still standing. Timing the application to occur immediately before leaf drop of the main crop is an alternative way to protect the seeds, and can improve the chances of successful seedling establishment [10].

The majority of research in this topic has focused on intersowing into standing maize by either broadcasting or direct-sowing, without yield reduction [7,11–13]. Belfry and Van Eerd [14] have reported cover crop mix biomass accumulation of 1116 kg ha^{-1} when sown into V4–V6 stages of maize (4–6 leaf stage) which was 33% increased compared with the later sowing at V10–V12 (10–12 leaf stage). Blanco-Canqui et al. [15] stated that soil cover in maize increased from 24% cover with no cover crop to 65% cover in plots intersown with winter rye. The increased green cover from the grown cover crops provides protection against soil erosion due to wind or water [16].

Baributsa et al. [11] found that using reduced maize sowing densities could significantly increase intercrop biomass when using red clover (*Trifolium pratense* L.) and chickling vetch (*Lathyrus sativus* L.). When legume species were sown at the same time as maize, maize yields were reduced 43 to 69% by the legumes when weeds were not controlled, and 0 to 35% depending on the legume when weeds were controlled [13]. It was also found that legume production was 57% higher when weeds were controlled. Legumes did not suppress weed growth, showing that weed control is still essential when intersowing. This also makes intersowing a better option compared with sowing cover crops at the same time as a cash crop because there is time to control weeds before cover crops are sown [13]. Multiple reports have concluded that intersowing does not affect maize grain yield when done at later growing stages such as V5–V8, when side dressing fertilizer is common [7,11], but Ruffatti et al. [12] found a maize yield reduction of 7 to 22% in some environments when intersowing in early September. Direct sowing winter camelina the same day as maize or soybean has been found to lead to a reduction in grain yield. Sowing dates after the V4 stage that were broadcasted did not have reduced maize or soybean yield in both crops [17]. Sandler et al. [18] demonstrated intersowing radish into soybean did not reduce soybean yield. Radish that was intersown was able to increase overall biomass when compared with radish sown after soybean harvest [18].

Cover crops can also provide many soil health benefits, such as improving soil structure, increasing water infiltration, soil microbial activity, providing wildlife habitats, and scavenging of nutrients otherwise lost by erosion or leaching [16]. Radish has the ability to scavenge NO_3-N from the soil, ranging from 19.7 to 202 kg ha^{-1} [19], and winter camelina can sequester residual soil NO_3-N in the biomass throughout the fall and resume scavenging in the spring, and has been shown to accumulate from 24 to 59 kg N ha^{-1} in the above ground biomass [17]. If cover crops are not established early, limited growth will result and there are no added benefits. One way to obtain enough growth to cover the soil in the fall would be to sow cover crops into R4 and R6 soybean.

The objectives of this work were to determine the following aspects of cover crops intersown into standing soybean: (1) cover crop performance under the soybean leaf canopy at two different sowing dates, (2) effects of cover crops on soybean yield and subsequent spring wheat yield, and (3) cover crop scavenging ability of soil NO_3-N .

2. Materials and Methods

2.1. 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). The soil type in Fargo is Fargo silty clay (fine, smectitic, frigid Typic Epiaquerts) and the soil type in Prosper is Kindred-Bearden silty clay loam (Kindred: fine-silty, mixed, superactive Typic Endoaquoll; Bearden: fine-silty, mixed, superactive, frigid Aeric Calciaquioll) [20]. Daily temperature and rainfall was recorded by the North Dakota Agriculture Weather Network [21] at both sites (Figure 1).

In 2015 and 2016, the previous crop in Fargo was oat (*Avena sativa* L.) and the previous crop 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. Soil samples were taken at the 0 to 15 cm depth and tested for soil pH, organic matter, P [22], and K with the ammonium acetate method [23] using a Buck Scientific Model 210 VGP Atomic Absorption Spectrophotometer (Buck Scientific, East Norwalk, CT, USA). Soil sample analysis for NO_3-N was analyzed from 0 to 60 cm depth according to the Vendrell and Zupancic [24] method (Table 1).

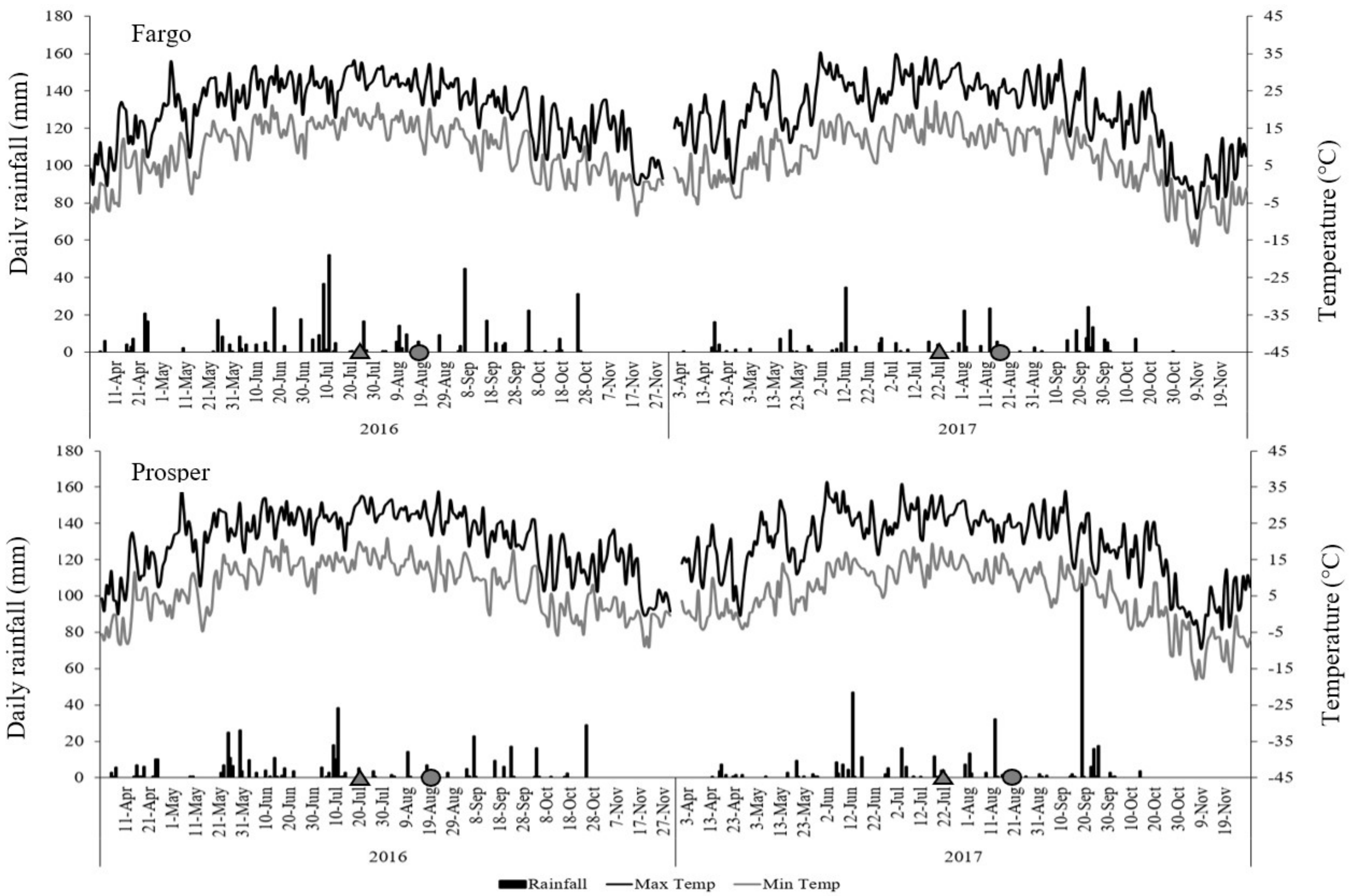

Figure 1. Daily rainfall, maximum temperature, and minimum temperature at Fargo and Prosper, ND, from April 2016 to November 2017. The R4 cover crop sowing dates are represented with an △, and the R6 cover crop sowing dates are represented with an ●.

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

Environment	pH †	OM g kg^{-1}	P mg kg^{-1}	K mg kg^{-1}	NO_3-N (0-15 cm-depth) kg ha^{-1}	NO_3-N (15-60 cm-depth) kg ha^{-1}
2016						
Fargo	7.8	60	20.3	398.0		
Prosper	6.8	44	45.0	251.5	45.8	90.0
2017					27.5	94.2
Fargo	7.5	65	23.0	350.5		
Prosper	6.8	41	56.0	348.0	35.9	94.2

† pH; OM, organic matter; P, and K were analyzed from 0 to 15 cm soil depth.

The experimental design was a randomized complete block design (RCBD) with a split-plot arrangement with four replicates. The main plot was a 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, with each sub-plot separated by 0.56 m. Cover crops were sown into three inter-row spaces in each sub-plot to create a cover crops sown border and avoid interference with next plot's cover crop. Sub-plots were 7.6 m in length at planting and reduced to 6.1 m in length at harvest.

Soybean cultivar selected was glyphosate-tolerant (Roundup Ready 2 Yield®, Peterson Farms Seed, Prosper, ND, USA), with a bush type architecture, relative maturity group 00.8, and soybean cyst nematode (*Heterodera glycines* T.) resistance. A bush type architecture was selected to allow the cover crops to grow under the canopy, and the soybean cyst nematode resistance was selected due to the high 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, USA) at a row spacing of 0.56 m and a sowing depth of 2.5 to 3.8 cm for all sowing dates. The soybean sowing rate was 505,000 pure live seeds ha^{-1} in order to reach the target plant population of 432,100 plants ha^{-1}.

Five treatments were selected to be intersown between the soybean rows: winter pea (Austrian, 1000-seed weight 97.8 g); forage radish (Daikon type (variety not stated), 1000-seed weight 1.26 g); winter camelina (cv. Joelle, 1000-seed weight 0.9 g); winter rye (cv. Rymin, 1000-seed weight 3.41 g); and a mixture of all four cover crops. There was also a check treatment without cover crops. Cover crops were intersown at two later reproductive growth stages, R4 and R6. Previous studies have shown that best survival of cover crops intersown into soybean were at later stages of maturity [17]. This was the rationale to select R4 and R6 as intersowing dates, since in earlier stages the probability of failure to establish is much higher. In Fargo, for the R4 stage, the cover crops were sown on 25 July 2016 and 21 July 2017; at R6 cover crops were sown on 16 August 2016 and 21 August 2017. In Prosper, for the R4 stage cover crops were sown on 26 July 2016 and 21 July 2017, while at the R6 stage they were sown on 16 August 2016 and 21 August 2017. The sowing rates for winter pea and forage radish were 89 and 5.6 kg ha^{-1} of pure live seed (PLSE), respectively. Sowing rates for winter camelina and winter rye were 6.7 and 67.2 kg PLSE ha^{-1}, respectively. Treatment with 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. When sowing the mix treatment large seed and small seed were sown separately to reduce the chances of getting small seed only in one part of the sub-plot. Sowing by hand mimicked a new 15 cm high clearance twin-row planter (Amity Technology, Inc., Fargo, ND, USA) that can intersow cover crops at later developmental stages of soybean.

Spring wheat followed the soybean in the next growing season. 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 spacing planter (Great Plains, Salinas, KS, USA) at a target plant population of 3.7 million plants ha^{-1}. The spring wheat cultivar selected was Glenn, which had an average yield, was moderately resistant to head scab (*Fusarium graminearum*), and has medium- to early-maturity. Spring wheat plots were 2.2 m wide and 6.1 m long and were sown exactly where the cover crop treatments were the previous year. The same area with the four rows of soybean in each plot from the year before were therefore sown to wheat. There were 14 rows of wheat per plot.

Applications of glyphosate (N-(phosphonomethyl) glycine) at 1.1 kg active ingredient ha^{-1} were done prior to soybean sowing and post soybean emergence, and before the first cover crop sowing date at both locations in 2016 and 2017, for a total of three applications during the season. An 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.

Soybean and spring wheat were not fertilized with chemical fertilizer or manure in any environment. Not fertilizing was done to provide a better indication of how efficient cover crop treatments are at nutrient cycling.

2.2. Sampling and Analysis

Cover crop biomass was collected after soybean harvesting 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 for any crop. Biomass samples were collected by hand clipping 0.09 m^2 from each cover crop area growing between the two-center soybean rows. All above ground biomass was collected; this did not include any above ground enlarged hypocotyl areas from radish. The below ground roots of the radish were left undisturbed. All cover crop biomass samples were dried at 70 °C until they reached a constant weight. Dried samples were then ground by a Model 4 cutting mill (Eberbach Corporation, Ann Arbor, MI, USA) to pass through a 1 mm size sieve. To obtain the N content, ground samples were analyzed using an XDS near-infrared (NIR) rapid content analyzer (Foss, Denmark). Selected biomass samples of cover crops were analyzed by wet chemistry proximal analysis and used to calibrate the NIR instrument. With NIR analysis, nutrient uptake by cover crops can be calculated using the formula N content times dry matter yield.

Soybean seed 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, USA) 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, Salt Lake City, UT, USA). Spring wheat grain yield was collected from the middle 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 and spring wheat grain protein content was determined via an XDS NIR rapid content analyzer.

Soil samples were collected in the fall of 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 cover-crop twin rows, 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.

Table 2. Soil sample dates of the two experiment locations, Fargo and Prosper, North Dakota.

Location/Year	Fall after Cover Crop Harvest	Before Spring Wheat Sowing	After Spring Wheat Harvest
Fargo 2016	6 Nov	-	-
Prosper 2016	21 Oct	-	-
Fargo 2017	30 Oct	17 May	19 Oct
Prosper 2017	30 Oct	17 May	17 Oct
Fargo 2018	-	15 May	1 Nov
Prosper 2018	-	15 May	6 Nov

2.3. Statistical Analysis

Statistical analysis was conducted using standard procedures for a randomized complete block design with a split-plot arrangement. Each location per year combination was defined as an environment and was considered a random effect. 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 ((version 9.4, SAS Institute Inc., Cary, NC USA); if the *F* test was significant at $p \leq 0.05$, mean separation was performed using least square means paired differences, but only for fixed main effects or interactions. For significant interactions with random effects (i.e., cover crops × sowing date × environment), only one least significant differences (LSD) value was calculated for all possible mean comparisons in the interaction, with the error mean square value and corresponding degrees of freedom.

3. Results and Discussion

3.1. Cover Crop Biomass Yield

The analysis of variance was significant only for the environment by sowing date by cover crop interaction for fall cover crop biomass yield, but not significant for cover crop spring biomass yield (Table 3). This significant interaction is 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 dates, it was the lowest yielding of all cover crops (Table 4). Cover crops sown in R4 in 2016 received rainfall every week after sowing (Figure 1) while in 2017 there were no significant rain events between mid-August and mid-September. Thus, lower biomass of radish in R4 in 2017 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, lack of rainfall after drilling for two or three weeks' limited emergence. This is similar to the findings of Sandler et al. [18] where lack of rain in the early parts of establishment led to decreased biomass yield in radish intersown into soybean. As explained above, low biomass yield in 2017 may be explained by lack of water after emergence and then excess water in September, with a single rain event totaling just over 110 mm of rainfall (Figure 1). This large rain event caused saturated field conditions for a prolonged period. Radish has a low tolerance for wet soils and performs poorly in soils prone to prolonged periods of wetness [25].

Different environments and sowing dates resulted in seven possible environments-sowing dates (ESD) scenarios. Only ESDs where at least one of the cover crops produced measurable biomass in the fall were considered in the analysis. None of the cover crops produced biomass in Fargo at the R4 sowing date. Hence, only seven ESDs had at least one cover crop species with measurable biomass, thus analyzed (Table 4).

Winter pea sown at either the R4 or R6 stage of soybean development established well and ranked first in five out of seven ESDs among all cover crops on fall biomass yield (Table 4). Chen et al. [26] have 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. Winter pea was intersown successfully into switchgrass (*Panicum virgatum* L.) producing up to 2.7 Mg ha^{-1} in Oklahoma [27]. Winter pea grown in Kansas was able to produce an average biomass of 622 kg ha^{-1} over four years [28].

Winter camelina did not establish in Fargo and Prosper 2016 and Fargo 2017 sown at R4; no data was recorded. In 2017, in Fargo, cover crops sown at R4 emerged in almost 100%. due to timely rain events of greater than 25 mm per event, one week and two weeks after being sown (Figure 1). However, 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 th eir leaves, increasing the solar radiation reaching the inter-row to about 90% photosynthetically active radiation (PAR) by 12 September. 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 for this sowing date for any of the cover crops. In addition, cover crop plants had acclimated to low light conditions of less than 20% PAR under the canopy, 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 have been converted into heat, explaining the desiccation and death of all emerged seedlings.

In Fargo 2017 at the R6 and Prosper 2017 at the R4, respectively, winter camelina did establish and produce recordable biomass yield (Table 4). In Fargo 2017 the cover crops sown in R6 did not receive rain until 12 September (Figure 1), most of them emerging after this rain event when soybean had already dropped almost all their leaves, promoting cover crop growth. Without adequate moisture after germination, winter camelina struggles to survive while under soybean [17]. Research also shows that larger seed size leads to faster emergence compared with smaller seeds at the same sowing depth [29,30]. Other research has shown establishment of cover crops without moisture following sowing leads to a decrease in establishment and lower winter survival rate [8,9].

Another concern from farmers at the time of harvest is the possible interference of cover crops in the inter-row at harvest. Green leaves or material could clog the combine or increase the seed moisture. In this study, the cover crops species selection and sowing dates were chosen in order to avoid harvest interference. Three of the cover crops chosen were winter crops because they do not bolt and grow near the soil surface below the lowest soybean pod location. This reduces the chance of green material going into the combine at harvest. Radish is the only one of the cover crops studied which could interfere at harvest. Avoiding interference was also the reason for choosing later sowing dates. The shade soybean provides suppresses the cover crops' growth enough that they will not accumulate enough biomass to interfere with harvest. Soybean seed moisture was analyzed, finding no differences among treatments.

The following spring, winter rye produced the highest amount of biomass of 1.74 Mg ha^{-1} (Table 5). These results for spring biomass are similar to other intersowing experiments including winter rye [7,31]. Winter pea and radish did not survive the winter, so there was no recorded biomass in the spring. Although a winter annual in some environments, winter pea does not survive winters in North Dakota [32]. Research in Kansas and states in the Pacific Northwest have shown winter peas to survive the winter and resume growth in the spring [26,28].

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 5). Winter camelina was able to survive winter when it was established in the fall; this is similar to the results of other work done in the Midwest [33,34]. Other researchers have shown winter camelina to produce similar spring biomass yields as those observed in this study [17,31].

3.2. Cover Crop Nitrogen Accumulation

The combined analysis of variance across all environments and sowing dates showed no difference among treatments for cover crop nitrogen accumulation in the fall. 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. The 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. [31] and Noland et al. [7], respectively. In Berti et al. [17], winter camelina intersown into maize and soybean accumulated 24 to 55 kg N ha^{-1} in the spring. Other research focused on radish intersown into soybean has had N accumulations of 36.4 kg N ha^{-1}. This low amount may be explained by dry weather in the fall [12]. Winter pea intersown into switchgrass results show N accumulation of 42.1 kg N ha^{-1} [27]. The results that indicate when cover crops are well established into soybean, an acquisition of large amounts of nitrogen is present in the biomass, reducing the potential offsite dispersion of free NO_3-N in the soil to leaching and runoff.

Table 3. Analysis of variance and mean squares for five cover crops (CC) and two sowing dates (SD) for fall cover crop biomass, spring cover crop biomass, fall cover crop N accumulation, soybean yield, spring-wheat yield, fall soil NO_3-N , spring soil NO_3-N , and soil NO_3-N after spring wheat across four environments (Env) at Fargo and Prosper, ND, in 2016 to 2018.

SOV	df	Fall CC Biomass	df	Spring CC Biomass	df	Fall CC N	df	Soybean Yield	Wheat Yield	df	Fall Soil NO_3-N	df	Spring Soil NO_3-N	Soil NO_3-N NO3-N after Wheat
Env	3		2		3		3			2		3		
Rep(env)	12		8		12		12			9		12		
SD	1	0.6	1	0.3	1	1026	1	915	120	1	26.4	1	1.4	326.5
Env × SD	2	1.4	2	0.5	2	2075	3	13825	341442	1	407.5	2	1399.8	158.7
Error (a)	8	0.9	7	0.2	8	1055	8	245321	360822	5	432.8	8	511.8	275.1
CC	4	0.8	3	1.9	4	2081	5	86955	663396 *	5	613.5	5	886.4	27.3
Env × CC	9	0.6	2	0.5	8	583	12	29123	181466	8	405.0	12	595.4	140.1
SD × CC	3	1.2	1	0.1	3	986	3	29629	81480	3	44.6	3	198.8	64.6
Env × SD × CC	6	0.6*	1	0.7	6	767	6	104219	111508	2	434.5*	4	324.9	264.8
Error (b)	49	0.2	12	0.2	42	378	61	62146	141065	40	93.7	57	471.7	166.5
CV, %		33.0		31.5		32.7		8.4	14.5		32.0		35.6	31.7

* Significant at 0.05 probability level.

Table 4. Mean fall cover crop biomass yield in Fargo and Prosper, ND, in 2016 and 2017.

	Fargo 2016 Mg ha^{-1}		**Prosper 2016 Mg ha^{-1}**		**Fargo 2017 Mg ha^{-1}**		**Prosper 2017 Mg ha^{-1}**	
Cover crop	**R4**	**R6**	**R4**	**R6**	**R4**	**R6**	**R4**	**R6**
Winter camelina	-	-	-	-	-	1.13 cd	1.17 cd	-
Winter pea	2.04 bc†	1.60 bc	2.13 b	1.58 bc	-	1.52 bc	1.00 cd	1.40 bc
Radish	0.58 d	1.35 c	1.02 cd	3.04 a	-	0.82 cd	1.28 cd	0.72 cd
Winter rye	1.53 bc	0.57 d	0.97 cd	1.02 cd	-	1.09 cd	1.61 bc	0.44 d
Mix	1.54 bc	0.96 cd	1.56 bc	1.53 bc	-	1.04 cd	2.03 bc	0.94 cd

† Means with different lowercase letters are significantly different at $p \leq 0.05$ by the LSD test. Letters are to compare between means of all possible comparisons.

Table 5. Mean fall and spring cover crop biomass yield, N accumulation, 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, in 2016 to 2018.

Cover crop	Fall Biomass Yield Mg ha^{-1}	Spring Biomass Yield † Mg ha^{-1}	Fall Biomass N Accumulation † kg ha^{-1}	Soybean Seed Yield kg ha^{-1}	Spring Wheat Grain Yield ‡ kg ha^{-1}
Winter camelina	1.15	0.73	28.7	2933	2144b
Winter pea	1.61	-	71.5	3008	2708a
Radish	1.25	-	73.2	3025	2812a
Winter rye	1.03	1.74	47.2	3025	2174b
Mix	1.37	0.94	55.9	2953	2691a
Check	-	-	-	2908	2684a
LSD (0.05)	NS	NS	NS	NS	315

† Cover crop spring biomass and fall N accumulation is only accounting for above ground plant biomass. ‡ Means with different lowercase letters were determined to be significantly different at $p < 0.05$ using the LSD test.

3.3. Soybean and Spring Wheat Yield

The combined analysis of variance across all environments and sowing dates showed no differences among treatments for soybean seed yield. However, spring wheat did show differences among treatments ($p \leq 0.05$) (Table 3).

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 seed yield. 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. [17] found similar results when intersowing winter camelina into R1 and R2 without reducing soybean seed yield, but did see a yield reduction in maize grain when winter camelina was sown the same day as maize. Similarly, intersowing cover crops into soybean did not reduce soybean seed yield [12,18].

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 5). As winter camelina and winter rye survived the winter and resumed growth in the spring, these cover crops also acquired nutrients and water. This, in turn, impacts the amount of available water for subsequent spring wheat growth, hindering development and decreasing yield. Krueger et al. [35] found that winter rye terminated too close to maize sowing led to decreased soil water and crop yield. Previous research has shown winter rye produces allelopathic compounds that reduces other grasses growth which affect wheat [36] and maize [35]. In addition, rye can keep the cycle of root diseases which can also contribute to yield decrease in maize [37,38]. In addition, reduction in soil nitrogen supply can negatively impact spring wheat yield following winter rye [39].

3.4. Soil Nitrate Removal and Replacement

The combined analysis of variance showed significance for the environment by sowing date by cover crop interaction for soil NO_3-N in the fall. Soil NO_3-N in Fargo in 2016 is not shown, due to excessive rain in the fall which made sample extraction impossible. The largest amount of residual soil NO_3-N, 61.7 kg ha^{-1}, was observed in Prosper 2016 in the check plot without any cover crops (Table 6). The lowest soil NO_3-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 NO_3-N in the cover crop plots are related to the cover crop biomass N accumulation in Table 5. The initial soil NO_3-N from 0 to 60 cm in depth before starting the experiment ranged between 121.7 and 135.8 kg ha^{-1} (Table 1) and the N accumulation by cover crops in the fall ranged between 28.7 and 73.2 kg ha^{-1}, explaining in part the fate of soil NO_3-N at the beginning of the season. For example, the average baseline soil NO_3-N across the four environments was 133.0 kg ha^{-1}; radish average N accumulation in its biomass was 73.2 kg ha^{-1} and the residual soil NO_3-N at the end of the fall in radish plots across all environments was 25 kg ha^{-1}. This would explain the probable fate of about 90 kg ha^{-1} of soil NO_3-N, although N balance is much more complex since there are many N inputs and outputs in the N cycle. Legumes such as winter pea usually utilize the available NO_3-N first before N_2 fixation takes place, which may explain the similar N accumulation and soil residual NO_3-N in the fall winter pea compared to that of radish. These cover crops show they have potential to scavenge and retain excess residual NO_3-N. Previous research using winter rye as a cover crop was able to reduce tile drainage discharge of NO_3-N loads by 63% compared with the no cover control [40]. In addition, rye and annual ryegrass in a mix reduced discharge by 69–90% compared with the no cover control [41]. Winter rye has the ability to scavenge as much as 28 to 56 kg N ha^{-1} [42].

However, the analysis of variance combined across four environments and two sowing dates showed no significant difference for soil NO_3-N in the spring before wheat sowing and following wheat harvest (Table 7). The expected results were to see an increase of soil NO_3-N in cover crop treatments due to the cycling of the nutrients in the cover crop biomass from the previous growing season. These results are similar to a study done by Cicek et al. [43] where radish biomass did not release the nitrogen fast enough to supply a subsequent wheat crop.

Table 6. Mean fall soil NO_3-N from 0 to 60 cm in depth in Fargo in 2017 and Prosper, ND, in 2016 and 2017.

Cover Crop	Prosper 2016 kg ha^{-1}		Fargo 2017 kg ha^{-1}		Prosper 2017 kg ha^{-1}	
	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 letters were found to be significantly different at $p \leq 0.05$ via LSD test. Letters are to compare between means of all possible comparisons.

Table 7. Mean soil NO_3-N levels in the fall, before spring wheat sowing, and after spring wheat harvest for five cover crop treatments averaged across two cover crop sowing dates and four environments at Fargo and Prosper, ND, from 2017 to 2018.

Cover Crop	Fall Soil NO_3-N kg ha^{-1}	Before Spring Wheat Sowing Soil NO_3-N kg ha^{-1}	After Spring Wheat Harvest Soil NO_3-N kg ha^{-1}
Winter camelina	26.4	53.7	13.8
Winter pea	30.6	65.8	43.8
Radish	25.0	60.1	37.1
Winter rye	29.5	44.1	48.3
Mix		65.2	43.9
Check	47.0	73.7	33.0
LSD (0.05)	NS	NS	NS

Soil NO_3-N samples are totals from 0 to 60 cm depth.

4. Conclusions

Although there was a lot of variability on cover crop establishment and performance among environments and sowing dates due to the effect of rainfall or lack thereof after sowing, some general recommendations can be drawn from this study. Different environments and sowing dates resulted in seven possible environments-sowing dates where at least one of the cover crops produced measurable biomass in the fall.

Winter pea sown at either the R4 or R6 stage of soybean development established well and ranked first among all cover crops in five out of seven ESDs on fall biomass production. Radish ranked first in fall biomass production at only one ESD. Winter rye was able to establish under soybean but since it is a winter annual, fall biomass production was low. Winter camelina survived in only two ESDs producing about 1 Mg ha^{-1} of biomass. As in winter rye, winter camelina is a winter annual so it is expected to produce reduced biomass in the fall. The mix of four cover crops was ranked second or third in five out of seven ESDs and first in one ESD indicating than mixes usually have a better establishment and fall biomass production than sole crops.

In general, we could conclude that the mix outperformed radish in three ESDs and outperformed winter rye and camelina in most of the ESDs, but winter pea was superior to the mix in most ESDs. Only winter camelina and rye survived the winter and provided some soil cover in the spring.

Cover crops intersown into standing soybean did not decrease soybean yield and scavenged significant soil NO_3-N in the fall while providing soil cover, which is of great benefit to increase cropping systems sustainability. However, this study did not show a benefit to a succeeding wheat crop.

Author Contributions: A.T.P. conducted the experiments, analyzed data, wrote the manuscript and edited it further. D.S. collaborated on the execution of experiments and editing of manuscript. M.T.B. obtained the funding, wrote project proposal, designed experiments, assisted with statistical analysis, and corrected and edited the manuscript.

Funding: Coordinated Agricultural Program, award no. 2016-69004-24784, and the North Dakota Soybean Council.

Acknowledgments: This research was possible thanks to the funding provided by the North Dakota State University Agricultural Experimental Station project ND 01557, the USDA-NIFAAuthors also wish to thank all technical staff, visiting scientists, undergraduate, and graduate students that helped support these studies.

Conflicts of Interest: The authors declare no conflict of interest.

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