Final Extension Report

Project Title: Soybean Crop Rotation for Manitoba Farmers (ARDI 12-1139)

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Background and Objectives:

Soybean is gaining popularity among farmers in Manitoba due to the development of improved short season varieties, its ease of production, and its tolerance to wet spring conditions. This is evident from the 37 % increase in harvested area of soybean from 2013 to 2015 with a total of 0.66 million harvested ha in 2015 across the Prairies (Statistics Canada, 2016). In Manitoba, the 32 % increase in soybean harvested area between 2013 and 2015 resulted in a record soybean production of 1.4 million tonnes in 2015, which was 30 % higher than that in 2013 (Statistics Canada, 2016). Most of the previous crop rotation research in the Canadian Prairies has been conducted with cereals or canola in rotation with pulses, mainly field pea (Stevenson and van Kessel, 1996; Beckie and Brandt, 1997; Beckie *et al.*, 1997; Miller *et al*., 2002). Research studies in eastern Canada and northern US states involving soybean have focused on soybean- corn, or soybean –wheat or soybean –sorghum rotations (Ding *et al. ,* 1998; Swink *et al.,* 2007; Dayegamiye *et al.,* 2012). With the expansion of soybean in Manitoba, soybean are increasingly being grown in rotation with crops such as spring wheat and canola, that are not common in the crop rotation literature. It is an important time to be studying how to optimize soybeans in rotation in Manitoba.

In western Canada, farmers are looking for ways to get the most out of their soybean crop. An important component of the soybean plant's capacity for nutrient uptake comes from the relationships it forms with soil bacteria and fungi. For example, the ability of the plant to fix nitrogen from the atmosphere is a result of the mutually beneficial symbiosis it forms with nitrogen fixing bacteria. This ability to biologically fix nitrogen has large agronomic and economic benefits. Soybeans also depend on soil fungi, known as Arbuscular Mycorrhizal Fungi (AMF), for phosphorus uptake. AMF have tiny roots known as hyphae that access soil pores that are too small for soybean roots to enter. This provides the plant with phosphorus that would otherwise be unavailable. These two types of soil organisms are affected by a number of environmental, agronomic and management factors.

It is also important to consider how soybeans impact other crops in the rotation. Soybeans are a nitrogen fixing crop. The other pulse crops grown in western Canada can contribute nitrogen to subsequent crops and soil test recommendations often include a nitrogen credit when they are developing fertilizer recommendations for subsequent crops. However, soybean is a different type of legume. It is a long season plant that removes a lot of nitrogen in the harvested grain. Nitrogen credits for soybean crops have been found to vary widely with location, soil type, growing season conditions, succeeding crops. Bundy *et al.* (1993) reported that nitrogen credits of soybean crop ranged from -22 to 210 kg N /ha for the corn crop that followed. Vanotti and Bundy (1995) reported nitrogen credits of 75 kg N /ha to corn crop following soybean on silt loam soils of Wisconsin. Ding *et al.* (1998) reported nitrogen fertilizer replacement value (NFRV) of soybean to following corn crop was 41 and 59 kg N /ha in 1994 and 1995 on silt loam soil of Guelph Ontario. In mid-western states of the USA, a general guide recommended for producers is to adjust nitrogen fertilizer rate downward by 45 kg N /ha when growing maize following soybean compared with maize following maize (Kurtz *et al*., 1984; Bundy *et al.,* 1993; Gentry *et al.,* 2001). In eastern Canada, the nitrogen credits of 30 kg N /ha in central Ontario, 15 kg N

/ha for south western Ontario (Ding *et al*., 1998) and 25 kg N /ha for Quebec have been recommended for soybean to a succeeding corn crop (Dayegamie *et al.,* 2012).

In Manitoba, Przednowek *et al.* (2002) studied rotational effects of legumes on succeeding wheat crop and found that soybean provided relatively little nitrogen benefit to a succeeding wheat crop. In Kansas, Staggenborg *et al.* (2003) reported that winter wheat following soybean required approximately 21 kg N /ha less nitrogen fertilizer to maximize yields than wheat following sorghum. In eastern Canada, Dayegamiye *et al.* (2012) calculated N credits of soybean in soybean-corn, soybean-wheat-corn and corn –corn rotations ranged from 31 to 42 kg N /ha. This highlights that nitrogen credits of soybean can be specific to the type of crop grown after.

The first objective of this crop rotation project focused on soybean was to study the effects of crop sequence on soybean yield, mycorrhizal colonization, and biological nitrogen fixation (Experiment 1). The second objective of this study was to quantify the nitrogen benefit of soybean to a subsequent wheat crop relative to a canola crop (Experiment 2).

Experiment 1: Soybean crop sequence

Procedure and Activities:

Experimental Sites

The experiments were conducted between 2012 and 2014 at the Ian N. Morrison Research Farm in Carman Manitoba (49°29’59.0”N, 98°01’50.4W”) (Carman), Kelburn Farms near St. Adolphe, Manitoba (49°41’46.6”N, 97°06’54.5”W) (Kelburn), and the CMDC Research Farm in Portage la Prairie, Manitoba(49°57’32.4”N, 98°16’32.6”W) (Portage). The soil at Carman was a clay loam, Orthic Black soil of the Eigenhof series. The soil at Portage la Prairie was a silty clay loam, Gleyed Rego Black soil of the Gnadenthal-Neuhorst complex. The soil at Kelburn was a clay Gleyed Black Chernozem of the Scanterbury series. See Table 1 for soil nutrient status of each site year. Climate data was obtained from weather stations in Carman, Portage la Prairie, St Adolphe and Winnipeg (James Richardson International Airport). Climate varied between site years and is summarized in Table 2.

Experimental Design

Each trial consisted of a two year crop rotation. The first set of trials ran from 2012-2013 and the second set of trials ran from 2013-2014. The first year of the trial consisted of a treatment crop and in the second year the test crop was grown. Note that the 2012-13 experiment at Portage was abandoned in 2012 due to poor crop establishment, thus there was only one experiment completed at the Portage site. The four different treatment crops were canola, corn, soybean and wheat. In the second year soybeans were grown on these different crop stubbles. The experiment design was a randomized complete block design (RCBD) with four blocks per site. The length of plot was 8 m at all site but the plot width varied depending on seeder width. Plot width was 7.5m at Portage, 6 m at Kelburn and 6.5m at Carman.

Table 1: Soil pH, soil nitrate (N), Olsen P, Potassium (K) and sulfate (S) at each experimental site in 2013 and 2014.

Research Site	Soil Depth (cm)	pH	N*	P**	K	S***
				ppm		
Carman 2013	0-15	6.6	3.3	10.3	287	6.9
	15-60		4.5			7.9
	60-90		2.9			
	90-120		1.8			
Carman 2014	0-15	6.7	5.3	9.3	240	
	15-60	7.7	6			
	60-90		3.6			
	90-120		3.1			
Kelburn 2013	0-15	6.9	10.4	12.9	356	9.5
	15-60		9.9			7.8
	60-90		3.8			
	90-120		3.2			
Kelburn 2014	0-15	7.3	7.9	29.3	367	
	15-60	7.6	6.1			
	60-90		3.1			
	90-120		3			
Portage 2014	0-15	8	9.3	9.8	249	
	15-60	8.1	5.9			
	60-90		3.1			
	90-120		2.4			

*nitrate-N, **Olsen-P, ***sulfate-S

Table 2: Mean monthly growing season temperature and precipitation at Carman, Kelburn and Portage research sites in 2013 and 2014 and long term averages (LTA) at each site.

Research Site	May	June	July	August	Growing Season
Temperature (°C)					
Carman 2013	10.5	17.7	18.7	18.9	16.5
Carman 2014	11.3	16.6	18.2	18.7	16.2
LTA	11.6	17.2	19.4	18.5	16.7
Kelburn 2013	11.6	18.4	19.3	19.8	17.3
Kelburn 2014	12	17.4	18.5	19.4	16.8
LTA	11.6	17	19.7	18.8	16.8
Portage 2013	10.9	17.8	19.2	19.5	16.9
Portage 2014	11.3	16.8	19	19.2	16.6
LTA	11.9	17.1	19.3	18	16.6
Precipitation (mm)					
Carman 2013	116	50.6	49.2	59.7	275.5
Carman 2014	30.9	116.7	47.5	122.4	317.5
LTA	69.6	96.4	78.6	74.8	319.4
Kelburn 2013	87.3	60.8	90.3	75.4	313.8
Kelburn 2014	66.8	157.1	40.3	91.8	356
LTA	56.7	90	79.5	77	303.2
Portage 2013	90.6	68.6	99.8	65.2	324.2
Portage 2014	49	135.3	20.2	92.3	296.8
LTA	58.4	90	78.4	68.3	295.1

Experimental Management

Wheat, canola, and soybeans were planted with an air seeder while corn was seeded with a row-crop planter. Seeding rates were based on recommendations from Manitoba Agriculture Food and Rural Development Initiative (MAFRD). See Tables 3 and 4 for treatment and test crop seeding dates, seeding rates and harvest dates. Test crop soybeans were seeding at rates between 445 000 and 545 000 plants/ha depending on site year (Table 4). All plots were cultivated once in the spring with shanks prior to seeding at Carman and Portage but not at Kelburn. In the fall, crop residue for soybean, wheat, and canola were incorporated using a cultivator after harvest. Corn plots were disced twice in the fall to incorporate the higher amounts of crop residue and received a finishing pass with a cultivator in the spring prior to soybean test crop planting at Carman and Portage. At Kelburn all tillage ocured in the fall due to heavy soils and not tillage occurred prior to spring planting of the wheat test crop.

Both reference and test crop soybeans were inoculated with liquid and granular inoculant. Neither test nor reference crop of soybean received any fertilizer inputs. At Carman in 2012, wheat was fertilized with 70-20-0 at a rate of 203.3 kg/ha based on soil test recommendations. Corn and canola were fertilized with 90-20-0-15 at a rate of 292 kg/ha based on soil test recommendations. At Carman in 2013, corn was fertilized with 46-0-0 at a rate of 148 kg/ha and 11-52-0 at a rate of 29 kg/ha based on soil test recommendations.

Table 3: Treatment crop varieties, seeding dates, seeding rates, harvest dates and yields.

Site Year		Preceding Crop			
		Canola	Corn	Soybean	Wheat
Carman 2012	Variety	73-75 RR	DKC 26-79 RR	25-10 RR	Glenn
	Seeding Rate	1 100 000	69 000	540 000	311 000
	Seeding Date	04-May	May-17	16-May	04-May
	Harvest Date	16-Aug	03-Oct	27-Sep	09-Aug
	Yield (kg/ha)	1597	5521	2677	4775
Kelburn 2012	Variety	73-75 RR	DKC 26-79 RR	25-10 RR	Glenn
	Seeding Rate	1 100 000	69 000	540 000	311 000
	Seeding Date	10-May	15-May	10-May	10-May
	Harvest Date	27-Aug	03-Oct	24-Sep	08-Aug
	Yield (kg/ha)	2187	7549	2938	3545
Carman 2013	Variety	73-75 RR	DKC 26-79 RR	24-10 RR	Glenn
	Seeding Rate	1 100 000	69 000	540 000	311 000
	Seeding Date	May-17	10-Jun	23-May	17-May
	Harvest Date	03-Sep	24-Oct	01-Oct	26-Aug
	Yield (kg/ha)	2640	9480	2878	5090
Kelburn 2013	Variety	73-75 RR	DKC 26-79 RR	24-10 RR	Glenn
	Seeding Rate	1 435 000	69 000	675 000	379 000
	Seeding Date	29-May	05-Jun	29-May	29-May
	Harvest Date	16-Sep	21-Oct	02-Oct	06-Sep
	Yield (kg/ha)	2845	8783	3050	2532
Portage 2013	Variety	73-75 RR	DKC 26-79 RR	24-10 RR	Glenn
	Seeding Rate	1 100 000	69 000	540 000	311 000
	Seeding Date	06-Jun	07-Jun	06-Jun	06-Jun
	Harvest Date	17-Sep	29-Oct	09-Oct	17-Sep
	Yield (kg/ha	3010	7713	2279	5122

Table 4: Soybean test crop seeding dates, seeding rates, and harvest dates.

Site year	Seeding Rate	Plant Stand	Seeding Date	Harvest Date
Carman 2013	545 000	510 000	22-May	01-Oct
Kelburn 2013	445 000	340 000	05-Jun	02-Oct
Carman 2014	495 000	435 000	26-May	07-Oct
Kelburn 2014	445 000	545 000	30-May	14-Oct
Portage 2014	445 000	600 000	04-Jun	21-Oct

Weeds in both treatment and test crops were controlled using herbicide. The same herbicide regime was used at all site years. Soybeans and corn were sprayed with glyphosate at a rate of 0.67L/acre (540 g a.e.). Canola was sprayed with glyphosate at a rate of 0.33L/acre (270 g a.e.). Wheat was sprayed with Buctril M at a rate of 0.4 L/ac and Axial BIA at a rate of 0.48 L/ac.

As a result of harvest loss and pod shatter from soybean treatment crop harvest, volunteer soybeans emerged in the soybean test crop at Carman and Kelburn in 2013. These volunteers were removed from the plots at harvest and weighed for yield separately. They were identifiable due to the fact that the treatment crop soybean was RR 25-10 while the test crop was RR 24-10, and could be visually distinguished by the color of the stem. In the 2014 test crop, volunteer soybeans were removed at the cotyledon (VC) stage at Kelburn as they emerged before the test crop. At Carman and Portage both volunteers and test crop emerged at the same time and could not be differentiated therefore no volunteers were removed from these sites.

Measurements

Soil sampling was performed in the spring before the growing season. Three soil samples were taken from each plot with a dutch auger at four depths: 0-15 cm, 15-60 cm, 60-90 cm, and 90-120 cm. Samples were sent to Agvise Laboratories in Northwood, North Dakota and analyzed for soil nitrate, Olsen P, potassium, sulfur.

Both reference crops and test crops were harvested to quantify yield. Plots were harvested at the end of the summer as each test crop reached maturity (Tables 3 and 4). Two combine passes were taken from each plot using a plot combine and then weighed. Samples were also taken to measure grain moisture content.

Arbuscular Mycorrhizal Fungi colonization

Soybean test crop roots were sampled to rate Arbuscular Mycorrhizal Fungi (AMF) colonization when the plants were at the V3 growth stage. Three plants per plot were sampled. Once root samples were collected they were soaked in water and washed using a 2 mm and 500 um sieve to separate the roots from soil and debris. Roots were removed from the sieves using fine forceps and stored in centrifuge tubes containing FAA (18 parts ethanol to one part formaldehyde to one part glacial acetic acid) in order to preserve the roots.

To prepare roots for scoring, roots were placed in a beaker containing a 10% KOH solution and then cleared in an autoclave for fifteen minutes to allow a staining agent (Chlorazol Black E mixed with equal parts water, glycerol and acetic acid) to enter the root cortex. Roots were heated in the staining solution in an oven for 60 minutes at 90°C. The roots were then put in centrifuge tubes containing 50% glycerol and 50% water in order to allow the darkest stains to clear out from the roots. After destaining for one to two weeks the roots were placed on a petri dish in a solution of 90% glycerol 10% water. The roots were then scored under a microscope to determine the percent mycorrhizal colonization based on the presence of hyphae, arbuscules, or vesicles visible in soybean roots along the intersection points of a 1 cm by 1cm grid. When at least a hundred events have been counted, the total number of hyphae, arbuscules, and vesicles is added up. The number of doubles (an event where two or more types of mycorrhizal activity have been found) is then subtracted from this number. Then the number of negatives is subtracted. This number is divided by the total number of events to determine the total percent colonization as shown below:

Hyphae + Arbuscules + Vesicles – Doubles – Negatives / Total Number of Events = Percent Colonization

Samples from Carman and Kelburn in 2014 contained spores that were initially mistakenly identified as vesicles. These spores likely belong to a different fungal species (for example *Pythium*). The vesicles counted in samples containing these spores were removed and identified as a negative event (no mycorrhizal activity) as there are generally only a small number of actual vesicles per sample.

Biological Nitrogen Fixation estimations

The percentage of nitrogen in the soybean plant fixed biologically by *Bradyrhizobium japonicum* was estimated using the natural abundance method. This method of N analysis compares the d15n signature of a nitrogen fixing crop to a non N fixing reference crop, from which biological N fixation can be estimated. Strips of canola were seeded at the back of each soybean test plot so that both the soybean and canola plants would grow in a similar soil profile. Soybeans in these canola test strips were weeded out by hand. A mass spectrometer was used to quantify the d15n signature of dried and ground plant material sampled from the soybean treatment crop and canola test crop. In 2013, soybean biomass was sampled at the R5 and R6 stages. In 2014, soybean biomass was sampled at the V3 and R5 stages. Samples were taken by cutting the soybean plant at ground level and taking the entire above ground part of the plant. Samples were dried in an oven for 48 hours at 100°C and ground sequentially in a wiley mill and cyclone mill to the consistency of baking flour prior to analysis. Samples were encapsulated and sent to the University of Saskatchewan where they were analyzed for n15 content using a mass spectrometer. The following formula was used to determine percent nitrogen biologically fixed based on the samples d15n signature:

(d15n of reference crop (canola) – d15n of test crop (soybean)) / (d15n of reference crop + B value) *100

The 15n isotope of nitrogen is found in the atmosphere at a constant rate of 0.3668‰. The 15n isotope is also found in soil, at a higher rate than that in the atmosphere. This rate depends on soil conditions and location. The reference crop is used to estimate the 15n signature of the soil. As canola is a non N fixing plant, all of its N will be derived from the soil and its 15n signature will closely resemble that of the soil. A N fixing plant will have a lower 15n signature as a significant portion of its N is derived from atmospheric N fixed biologically, where the 15n signature is lower. By comparing the difference in values between the d15n signature of the two plants, the amount of N fixed biologically can be estimated. The B value is a correctional value that accounts for N in the roots, which are typically not sampled.

Statistical Analysis

Analysis of variance was conducted with the Mixed procedure of SAS to test for significant treatment differences in terms of soybean yield, mycorrhizal colonization, biological nitrogen fixation and soil nitrate and phosphorus. Analysis of covariance was conducted with Proc Mixed for analyses that contained a covariate, including soybean yield adjusted for plant stands and mycorrhizal colonization adjusted for soil phosphorus. Regression analysis was conducted for mycorrhizal colonization and soil P, mycorrhizal colonization and plant P, and biological N fixation and soil N. Assumptions of ANOVA were tested using Proc Univariate to test for normality. Proc univariate was also used to test for and remove outliers.

<u>Results and Discussion:</u>

Soybean Yield

Soybean yields in this study were influenced by crop rotation. However, growing season conditions and soil conditions as each site were also very influential. Crop rotation trends were not consistent across all site years (Table 5). There were no significant differences in soybean yield between crop sequences at Carman in 2014 and Kelburn in 2013. Corn-soybean and wheat-soybean sequences were the most consistent, yielding comparatively well across all five site years. The canola-soybean sequence yielded lower than wheat-soybean at Portage in 2014, and lower than soybean-soybean at Carman in 2013, but otherwise did not yield lower than other crop sequences.

Table 5: Effect of crop sequence on soybean yield at Carman (2013, 2014), Kelburn (2013, 2014) and Portage (2014).

	Carman		Kelburn		Portage
Treatment	2013	2014	2013	2014	2014
	----------Kg/ha----------				
Canola	3764 **b**†	3095 **a**	2844 **a**	3110 **a**	2185 **b**
Corn	3614 **b**	2995 **a**	2529 **a**	3042 **a**	2458 **ab**
Soybean	4158 **a**	3198 **a**	2807 **a**	2487 **b**	2299 **ab**
Wheat	3550 **b**	3086 **a**	2550 **a**	3211 **a**	2619 **a**

† Means within a column followed by the same letter are not statistically different at P<0.05 according to Fisher's LSD.

The soybean-soybean treatment in this study had the greatest variability in yield between sites and years. At Carman in 2013, the soybean-soybean sequence yielded significantly higher than the other three sequences. The reverse was seen at Kelburn in 2014 where the soybean-soybean sequence yielded significantly lower than the other three treatments. The wheat-soybean sequence yielded higher than canola-soybean at Portage in 2014. Manitoba provincial data available from Manitoba Agriculture Services Corporation (MASC) shows that from 2008-2012 soybeans grown on soybean stubble yielded 95 percent relative to the provincial average for soybean yield (Kubinec, 2014).

Variability in yields for the soybean following soybean treatment were influence by volunteer soybeans in the test crop. Volunteer soybeans emerged in several of the soybean-soybean sequence plots and sometimes impacted yield. The largest effect was at Carman in 2013. Volunteer soybeans contributed on average 1715 kg/ha to yield in soybean-soybean treatments at Carman in 2013. Soybean-soybean sequence plots yielded 4448 kg/ha including volunteers, but only 2733 kg/ha without volunteers. When volunteer yield was not included, the soybean-soybean treatment yielded significantly lower than the other three sequences at Carman in 2013. Volunteer soybeans contributed on average 320 kg/ha to yield at each soybean-soybean plot at Kelburn in 2013. Soybean-soybean treatments yielded 2832 kg/ha including volunteers, and 2512 kg/ha without volunteers. Volunteer yield did not significantly affect soybean-soybean sequence yield compared to other sequences at Kelburn in 2013. Volunteers were included in final yield calculations as their presence would likely negatively affect the yield of non-volunteer soybeans. A small number of volunteer soybeans emerged at Kelburn in 2014, which were removed by hand. There were no volunteers detected at Carman and Portage in 2014.

Each of the preceding treatment crops in this study had different amounts of residue and it was hypothesized that this might play an influential role in soybean plant stand establishment. Crop residues were incorporated with tillage in the fall after harvest and seed beds were tilled prior to soybean seeding at some locations (Carman, Portage) in the spring. Surprisingly, location and year were more influential factors on soybean plant establishment than crop rotation treatments (Table 6). Soybean

plant stands were only influence by crop rotation treatments at one site year at Carman in 2014. There was no significant effect of plant stand on yield in this study.

Table 6: Soybean plant stands based on preceding crop at Carman (2013,2014), Kelburn (2013, 2014) and Portage (2014).

	Carman		Kelburn		Portage
Treatment	2013	2014	2013	2014	2014
	----------1000's of Plants ha^{-1}----------				
Canola	481 **a**†	483 **a**	320 **a**	561 **a**	603 **a**
Corn	534 **a**	481 **a**	354 **a**	535 **a**	617 **a**
Soybean	525 **a**	409 **ab**	340 **a**	507 **a**	592 **a**
Wheat	503 **a**	636 **b**	358 **a**	571 **a**	608 **a**

† Means within a column followed by the same letter are not statistically different at P<0.05 according to Fisher's LSD.

Soybean Nitrogen Fixation

Crop rotation had a significant effect on biological nitrogen fixation. Even in this two year rotational study, there were significant differences in biological nitrogen fixation between different rotations (Table 7). Soybeans grown on corn stubble had significantly higher biological nitrogen fixation than the other three rotations. Soybeans grown on canola had significantly lower nitrogen fixation compared to the other rotations.

The reason behind these differences may be explained in part by the levels of residual nitrogen left behind by each crop residue. Corn crops often leave behind lower levels of residual nitrogen (Table 8). This is likely due to the high carbon to nitrogen ratio of corn residue that ties up residual soil nitrogen, and makes it slower to break down than many other stubble types. In contrast, canola has a lower carbon to nitrogen ratio, and often leaves higher levels of residual nitrogen. In a soil environment with high residual nitrogen, the soybean plant is less likely to be heavily dependent on the *Bradyrhizobium* bacteria for its nitrogen as it can simply take what it needs from the soil.

This thrift makes sense for the soybean plant as there is a cost associated with forming a relationship with *Bradyrhizobium*. The soybean plant must provide the bacteria with carbohydrates in exchange for nitrogen. In a soil environment with low residual nitrogen, the plant becomes much more dependent on *Bradyrhizobium* to acquire its nitrogen and in this situation, it makes sense for the plant to give up some carbohydrates in exchange for scarcer nitrogen (Figure 1). The characteristics of corn stubble, therefore, creates an environment that is conducive to biological nitrogen fixation, while the canola stubble creates a higher nitrogen environment that makes it easier for the soybean plant to simply acquire more nitrogen itself. That being said, biological nitrogen fixation still accounted for more than half of the nitrogen in soybeans grown on canola stubble, indicating that regardless of crop stubble, biological nitrogen fixation is still the main source of nitrogen for soybeans.

Table 7: Average and the range in percent nitrogen in soybean plants fixed biologically by *Bradyzhizobium japonicum* as influenced by previous crop sequence at the beginning of seed set (R5 growth stage) at Carman (2013, 2014), Kelburn (2013,2014) and Portage (2014).

	Carman			
Treatment	2013		2014	
	% N Fixation	Range	% N Fixation	Range
Canola	67.6 **b**†	60.1 - 77.2	39.0 **c**	34.6 - 44.5
Corn	73.4 **ab**	66.0 - 82.8	70.5 **a**	62.9 - 82.4
Soybean	80.3 **a**	71.7 - 91.2	53.3 **b**	48.1 - 61.9
Wheat	79.3 **a**	71.0 - 89.8	62.2 **a**	54.9 - 71.8
	Kelburn			
Treatment	2013		2014	
	% N Fixation	Range	% N Fixation	Range
Canola	60.9 **c**	55.4 - 67.4	58.7 **b**	53.6 - 64.9
Corn	73.1 **b**	67.8 - 79.3	71.7 **a**	68.7 - 81.8
Soybean	85.1 **a**	78.1 - 92.7	56.0 **b**	51.5 - 61.5
Wheat	64.7 **c**	59.3 - 71.4	56.2 **b**	51.7 - 61.6
	Portage			
Treatment	2014			
	% N Fixation	Range		
Canola	55.3 **b**	50.3 - 61.3		
Corn	77.6 **a**	70.5 - 86.1		
Soybean	45.9 **c**	41.4 - 51.5		
Wheat	59.1 **b**	53.5 - 66.1		

† Means within a column by site year followed by the same letter are not statistically different at P<0.05 according to Fisher's LSD.

Table 8: Spring soil test nitrogen from 0-60 cm before seeding soybean test crops as influenced by preceding crop at Carman (2013, 2014), Kelburn (2013, 2014) and Portage (2014).

	Carman		Kelburn		Portage
Treatment	2013	2014	2013	2014	2014
	----------Soil N kg /ha----------				
Canola	43 **a**†	73 **a**	105 **a**	46 **a**	67 **a**
Corn	33 **ab**	30 **c**	85 **b**	55 **a**	29 **b**
Soybean	28 **b**	38 **bc**	35 **c**	41 **a**	67 **a**
Wheat	30 **b**	45 **b**	98 **ab**	68 **a**	54 **a**

† Means within a column followed by the same letter are not statistically different at P<0.05 according to Fisher's LSD. Note that nitrogen fertilizer applied to preceding treatment crops (corn, canola, and wheat) based on soil tests recommendations. No fertilizer was applied to soybean treatment and test crops.

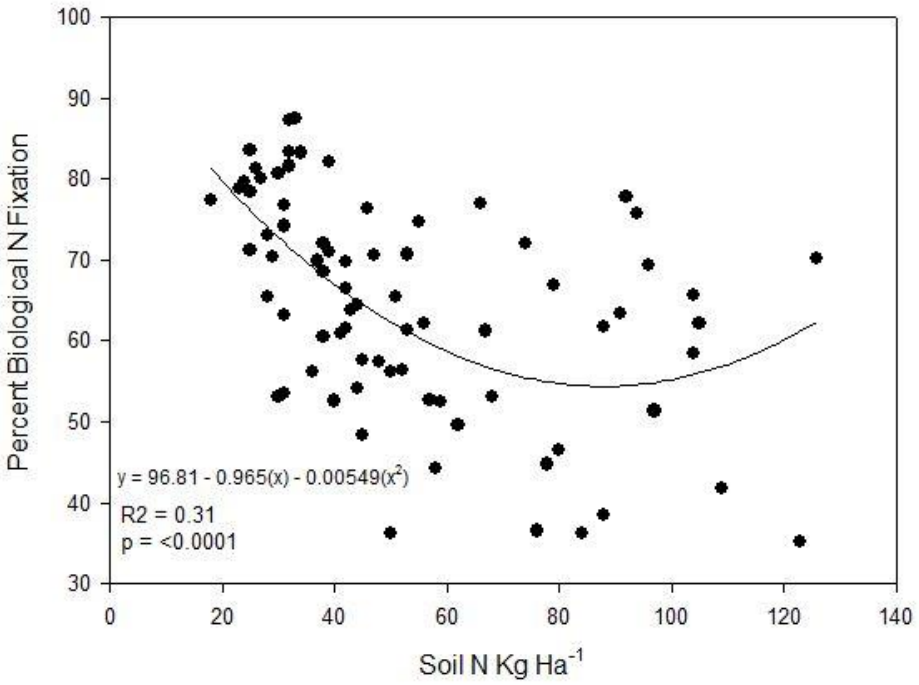

Figure 1: Effect of residual soil N on biological N fixation in soybean averaged across five site years. The regression equation and coefficient of regression are indicated.

Soybean Mycorrhizal Colonization

This study also found that crop rotation significantly affected the level of soybean root colonization by Arbuscular Mycorrhizal Fungi (AMF). Soybeans grown on corn or soybean stubble had significantly higher levels of AMF colonization than soybeans grown on wheat or canola stubble. This makes sense as corn and soybeans are strongly mycorrhizal crops that readily form symbiotic relationships with AMF. Thus, corn and soybean stubble would leave behind roots colonized by AMF that would serve as an excellent host, allowing AMF soil populations to remain relatively high. Although most plants are mycorrhizal to some degree, canola (and other members of the *Brassicaceae* family) does not readily form a relationship with AMF. In a field seeded to canola, the AMF population in the soil would have few roots available to colonize, and their population would start to decline. This study found that soybeans seeded on canola stubble had 10-20 % less mycorrhizal colonization than soybeans seeded on corn or soybean stubble after just one year (Table 9). Soybeans grown on wheat stubble also had lower mycorrhizal colonization. Although wheat is also capable of forming mycorrhizal symbiosis, it is generally less mycorrhizal than corn or soybeans.

The level of AMF colonization was also significantly influenced by soil phosphorus levels. In areas of higher soil phosphorus, AMF colonization tended to decline (Figure 2). This is similar to the relationship that soybeans have with *Bradyrhizobum* and soil nitrogen. If the plant can simply acquire nutrients itself, it will be less likely to expend energy and carbohydrates forming a relationship with AMF. In areas of low soil phosphorus, however, the plant becomes very dependent on AMF as a phosphorus source. Unlike residual soil nitrogen, however, there was no obvious correlation between crop stubble and soil phosphorus. Soil phosphorus levels had more to do with long term soil management and fertilization decisions at each of the experiment sites (Table 10).

Table 9: Total percent colonization by arbuscular mycorrhizal fungi of soybean roots at the V3 growth stage averaged across five site years (Carman 2013, 2014, Kelburn 2013, 2014 and Portage 2014).

	Type of mycorrhizal colonization		
Treatment	Percent Hyphae	Percent Arbuscules	Total Percent Colonization
Canola	29.7 **c**†	23.5 **a**	41.8 **b**
Corn	43.9 **a**	22.9 **a**	53.5 **a**
Soybean	44.0 **a**	24.0 **a**	54.0 **a**
Wheat	33.8 **b**	22.6 **a**	45.1 **b**

† Means within a column followed by the same letter are not statistically different at P<0.05 according to Fisher's LSD.

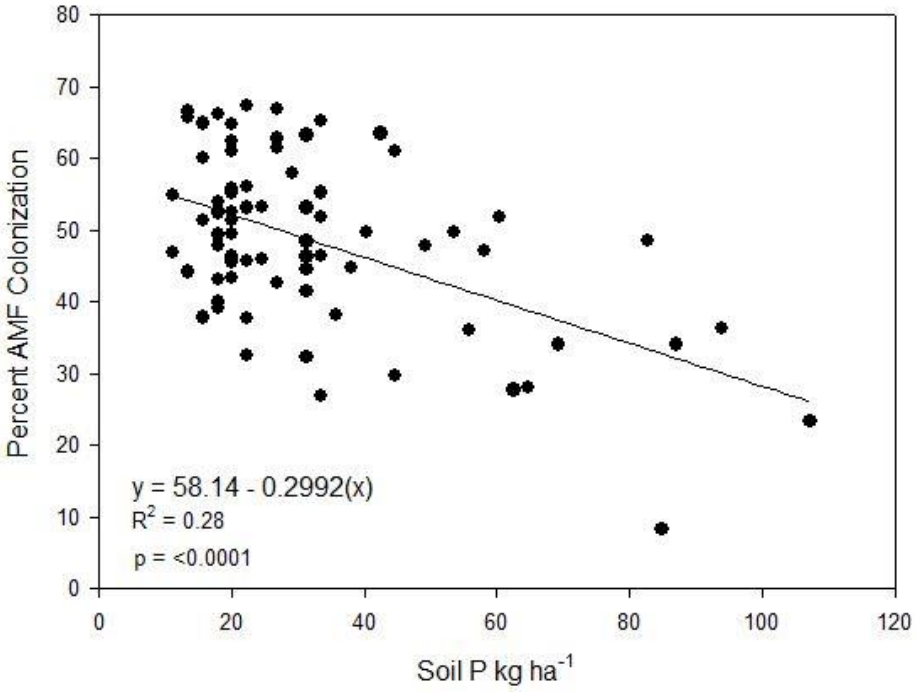

Figure 2: Effect of residual soil P on arbuscular mycorrhizal colonization of soybean roots across five site years. The regression equation and coefficient of regression are indicated.

Table 10: Soil P at 0-15 cm for soybean test crop plots based on preceding crop at Carman (2013, 2014), Kelburn (2013, 2014) and Portage (2014).

	Carman		Kelburn		Portage
Treatment	2013	2014	2013	2014	2014
			Soil P kg /ha		
Canola	26 **ab**†	27 **a**	31 **ab**	58 **b**	19 **ab**
Corn	27 **a**	22 **b**	32 **a**	60 **ab**	26 **a**
Soybean	18 **c**	19 **bc**	22 **b**	66 **ab**	17 **b**
Wheat	21 **bc**	16 **c**	30 **ab**	78 **a**	25 **ab**

† Means within a column followed by the same letter are not statistically different at P<0.05 according to Fisher's LSD.

Experiment 2: Soybean nitrogen credit

Procedures and Activities:

Experiment sites

Experiments were established at two of the same locations described for Experiment 1, the University of Manitoba Ian N. Morrison Research Farm in Carman (Carman) and Richardson International's Kelburn Farm (Kelburn). A two year experiment was set up at each location in both 2012 and 2013. In the first year of each experiment, treatment crops of soybean and canola were grown. Canola was selected as a non-nitrogen fixing reference to include as a treatment crop. In the second year of each experiment a wheat test crop was grown. Initial soil properties for each site are shown in Table 11. The nearest Environment Canada meteorological station to Carman and Kelburn were used to assess precipitation and temperature. Weather data from each growing season of test crop are reported in Figure 3 (Carman) and figure 4 (Kelburn).

Table 11 Experimental site soil characteristics in the top 0-15 cm of the soil profile (sampled in spring of 2012 and 2013) at the University of Manitoba Carman Research Farm (Carman) and Richardson International's Kelburn Research Farm (Kelburn).

			Crop rotation 2012-2013		Crop rotation 2013-2014	
			Carman	Kelburn	Carman	Kelburn
Sequence Year 1			***2012***		***2013***	
pH			6.7	-*	6.4	7.6
Olsen P (ppm)			8.0	-	19	81
K (ppm)			269	-	342	646
Organic Matter (%)			-	-	5.7	7.8
Cu (ppm)			1.16	-	0.89	2.34
Zn (ppm)			-	-	2.9	5.05
Soluble salts (mmhos /cm)		0-15 cm	-	-	0.27	0.4
		15-60 cm	-	-	0.7	0.6
Sulfur (ppm)		0-15 cm	9	-	14	15
		15-60 cm	60	-	360	39
Sequence Year 2			***2013***		***2014***	
Olsen P (ppm)	Canola		26	26	17	80
	Soybean		20	20	16	38
Sulfur (ppm)	Canola	0-15 cm	11	11	-	-
		15-60 cm	14	42	-	-
	Soybean	0-15 cm	12	24	-	-
		15-60 cm	13	78	-	-

* Data for these locations or nutrients not available

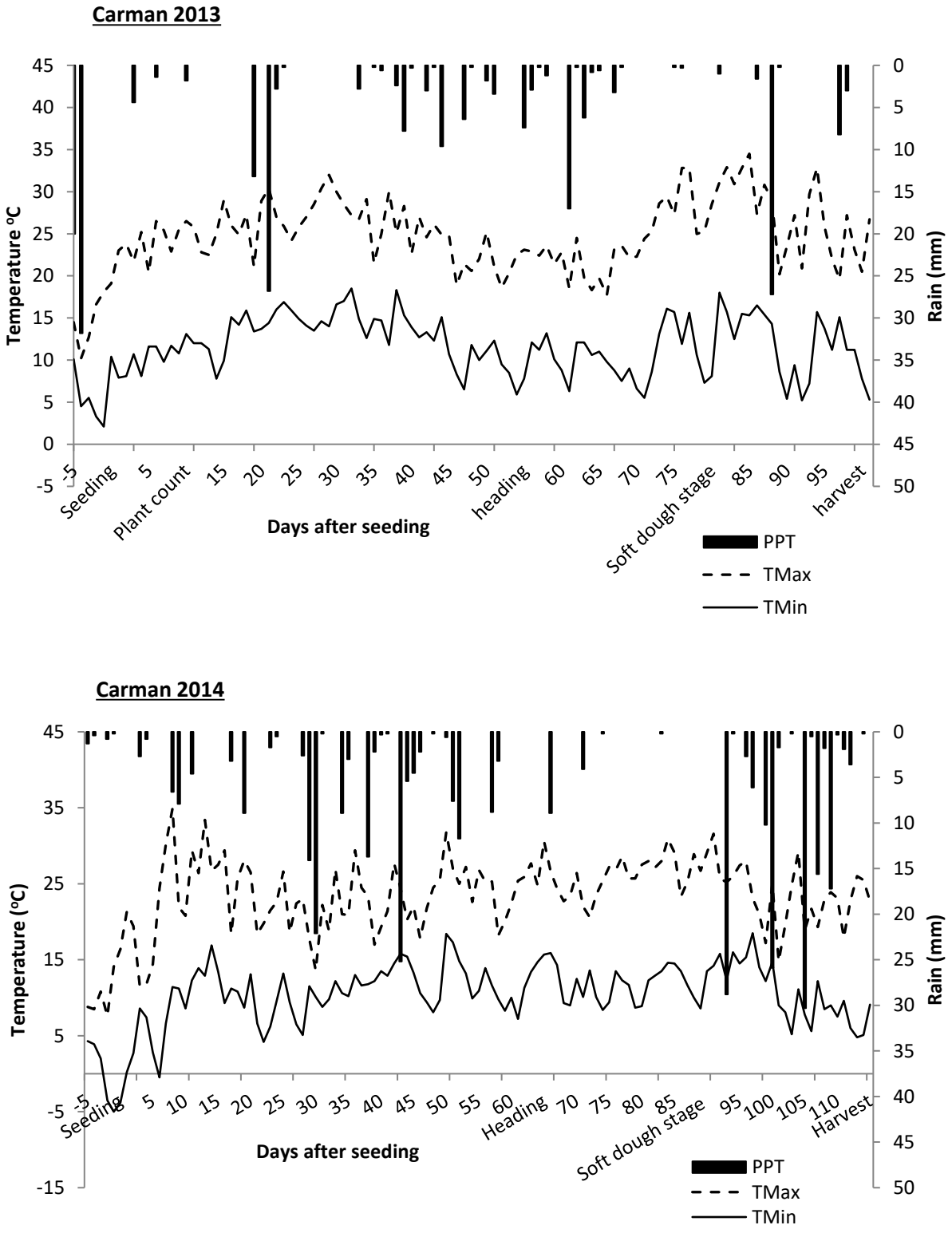

Figure 3: Growing season precipitation (PPT), maximum temperature (Tmax) and minimum temperature (Tmin) during the test crop growing season at Carman (2013 and 2014).

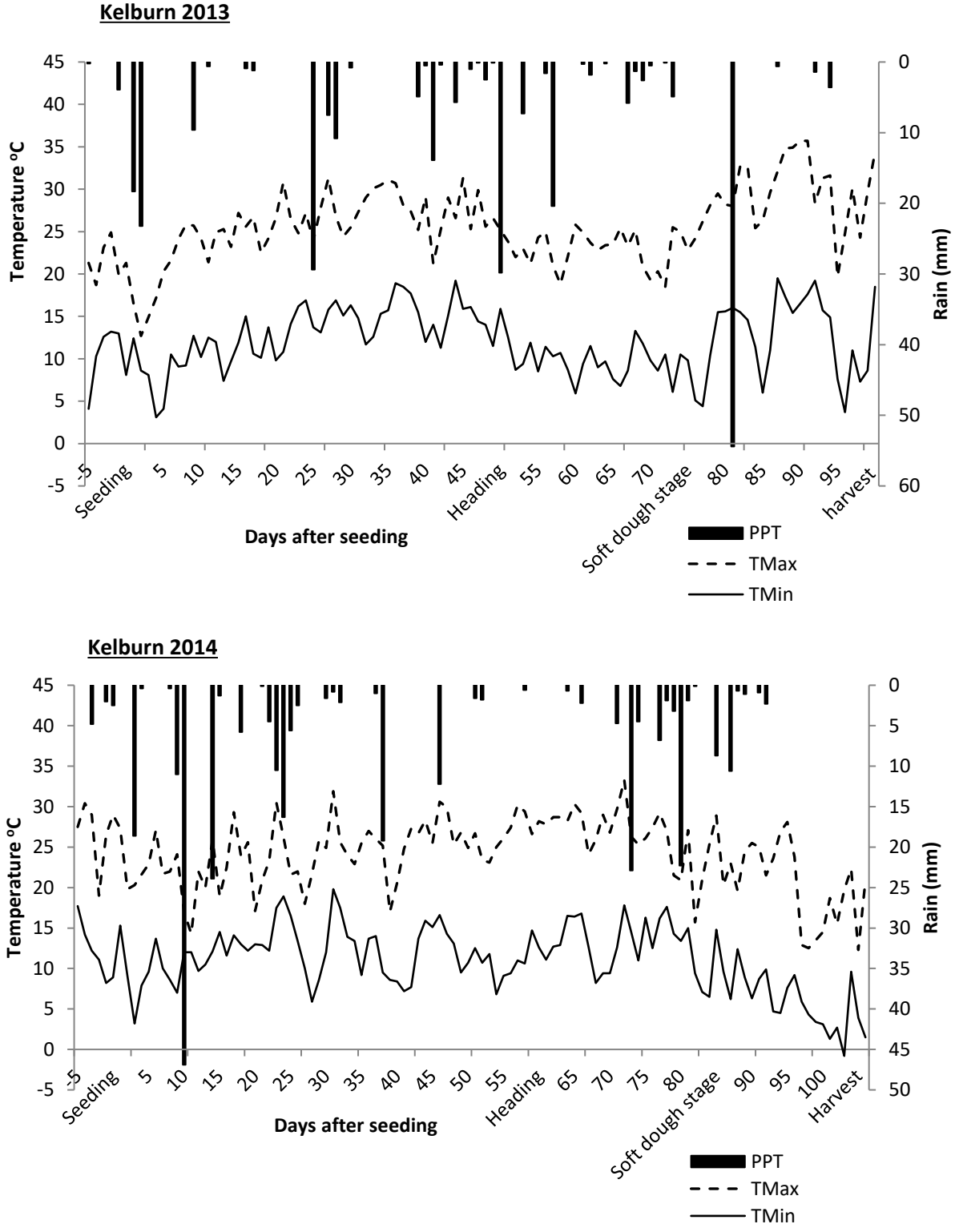

Figure 4. Growing season precipitation (PPT), maximum temperature (Tmax) and minimum temperature (Tmin) during the test crop growing season at Kelburn (2013 and 2014).

Experiment Design

This experiment had a nested design. In sequence year 1, the soybean and canola treatment crops were established at both experimental sites. The canola and soybean were seeded in large neighbouring unreplicated plots of 20 x 56 m at Carman and 17 m x 56 m at Kelburn. In sequence year 2, each large plot of soybean and canola was split into four replicated blocks. Each block were split into 5 plots for nitrogen fertilizer rate treatments (0, 30, 60, 90 and 120 kg N /ha) that were randomly assigned to each plot within a block. Subplot size was 4 m x 8 m at Carman and 3.4 m x 8 m at Kelburn. Thus, nitrogen fertilizer rate treatments for the wheat test crop were nested within each of the soybean and canola treatment crops.

Experiment Management

Sequence Year 1

In sequence year 1, both canola and soybean treatment crops were seeded on same day with a disc drills at both locations: Carman and Kelburn. See Table 12 for the dates of operations in each year. Soybean (Dekalb 25-10 RR) was seeded at the rate of 98 kg /ha to soil depth of 4 cm. Soybean seeds were inoculated with *Bradyrhizobium* using liquid (Optimize) and granular inoculant (Cell-Tech) at recommended rates. Canola (Dekalb 73-75 RR) was seeded at the rate of 6.7 kg /ha at soil depth of 1.5 cm. Nitrogen fertiliser was applied to canola crop as urea based on soil test recommendations for each site (Table 12). Plant counts were done in a 0.381 m^2 area at both of Carman locations using a metre stick. At Kelburn, weeds were controlled using glyphosate at the rate of 825 mL /ha. Canola was swathed approximately 10 days before harvest. Canola and soybeans were harvested using a small plot combine and yield was measured from a harvested area of 120 m^2 for canola and 91.44 m^2 for soybean. Both canola and soybean residues were spread evenly and plots were disced in the fall to incorporate crop residues. The following spring, soil samples were taken for soil moisture and nutrient testing within each soybean and canola block.

Sequence Year 2

In sequence year 2, the experiments were arranged in a randomised complete block design with four replicates within the canola and soybean treatment crop areas. A wheat test crop (cv *Glenn*) was grown across the entire experiment. Each block was subdivided into five nitrogen rate treatment plots (0, 30, 60, 90 and 120 kg N /ha). These nitrogen treatments were applied as urea mid-row banded at a soil depth of 5 cm at the time of wheat seeding. In 2014, there was a high risk of Fusarium Head blight (FHB) at Carman in 2014, when wheat was headed, therefore a fungicide (Porsaro) was applied at recommended rates. At Kelburn, fungicide was not applied as the crop was seeded later and was not flowering when conditions were favorable for the development of FHB.

Plant stand were counted in a 0.4m^2 area after 18-21 days after seeding at all sites except Carman 2013 where plant count was done 10 days after wheat seeding. Wheat biomass samples were taken at the soft dough stage from two 0.2 m^2 areas within each plot for dry matter yield and N uptake. Wheat was harvested using a small plot combine. After wheat harvest, all plots were soil sampled for nitrate N analysis. Three soil cores per plot were taken from three of the four replicates and were analysed separately. Soil samples were collected for in increments of 0-15 cm, 15-60 cm, 60-90 cm and 90-120 cm using a hydraulic soil corer or a hand auger.

Table 12: Management activities for the year 1 treatment crops (soybean, canola) and the year 2 soybean test crop at the University of Manitoba Carman Research Farm (Carman) and Richardson International's Kelburn Research Farm (Kelburn).

Site characteristic or management operation		Crop rotation 2012-2013		Crop rotation 2013-2014	
		Carman	**Kelburn**	**Carman**	**Kelburn**
Sequence Year 1		***2012***		***2013***	
Seeding date of soybean and canola		May 23	May 18	May 23	May 30
Fertiliser applied before seeding	Canola	67.3 kg /ha of N and 34 kg /ha of P	52 kg/ha N, 46 kg /ha P and 21 kg /ha of S *84 kg/ha of N	90 kg /ha of N and 20 kg /ha of P and 15kg /ha of S	52 kg/ha N, 46 kg /ha P and 21 kg /ha of S *84 kg/ha of N
	Soybean	-	-	-	6.7 Kg /ha N; 22.4 kg /ha P 0.6 kg /ha S
Plant count		June 7		Jun 11	
Weed control			Glyphosate @825 mL /ha		Glyphosate @825 mL /ha
Harvest	Canola	Aug 16	Aug 19	Aug 23	Sep 26
	Soybean	Sep 27	Sep 25	Oct 3	Oct 9
Sequence Year 2		***2013***		***2014***	
Spring Soil Sampling		May 9	May 13	May 10	Jun 2
Seeding of wheat		Jun 4	May 29	May16	Jun 4
Plant count		Jun 14	Jun 17	Jun 03	Jun 25
Soft Dough stage biomass		Aug 21	Aug 13	Aug 14	Aug 28
Wheat Harvest		Sep 13	Sep 6	Sep 8	Sep 18
Soil sampling (fall)		Sep 24-25	Oct 3-4	Oct 7-10	Oct 15-16

* A second application of N fertiliser was applied in June prior to rain event

Statistical analysis

The experimental design was a randomised complete block design of five nitrogen fertilizer rates nested within previous crop and sites. ANOVA was performed for each site using the MIXED procedure of SAS on the effect of preceding crop and N fertilizer rate on wheat grain yield, wheat N uptake and wheat biomass. Site, previous crop and fertiliser rate were considered as fixed effect in 3-way ANOVA. Replicates and replicate interaction with the fixed effects were treated as random.

Least squares means were used to compare fertilizer treatments within each previous crop. For the fertilisation treatment means and the fertilisation treatments with each previous crop, a probability level (α) of 0.05 was used as the significant thresholds for the soil and plant variables. Descriptive statistics were used to test the data for normality and skewness (γ) using the Proc Univariate function of

SAS. Most crop variables showed normal distributions. However, weed biomass and weed N uptake for Kelburn 2013 and residual soil nitrate data for all sites were transformed (square root for weed biomass and weed uptake, and log for total soil nitrate N were used) for analysis. The data reported for these variables are back transformed from the analyzed data.

Regression analysis was conducted using the REG procedure of SAS to test for relationships between nitrogen uptake and nitrogen fertilizer rates, and wheat grain yield and nitrogen fertiliser rates within each previous crop. Responses were best described by the quadratic model. Regression analysis was performed on treatment means averaged over replications as recommended by Gomez and Gomez (1984).

Results and Discussion

Was the wheat test crop responsive to nitrogen fertilizer treatments?

Wheat yield and biomass did not respond to nitrogen fertilizer treatments in all site years (Figures 5 and 6). This lack of yield or crop biomass response to nitrogen fertilizer limits scope of conclusions that can be made from this study to identify the nitrogen fertilizer replacement value of soybeans. Only one of the four site years (Carman 2013) showed a characteristic yield response to nitrogen fertilizer for both soybean and canola treatment crops (Figure 5). The other four site years (Carman 2014, Kelburn 2013, Kelburn 2014) had wheat yields that were unresponsive to nitrogen fertilizer treatment for either or both soybean and canola treatment crops (Figure 5). Trends in wheat biomass and nitrogen uptake were more consistent across sites and years (Tables 14 and 15) as theer was no significant interactions with site. Both measures of crop growth were higher following canola than soybean (when averaged over all fertilizer treatments) and both increased with the increasing nitrogen fertilizer treatment (when averaged over treatment crops) (Tables 14 and 15).

A few factors could have contributed to the lack of wheat test crop response to nitrogen fertilizer. One factor is the high organic matter levels at the experimental sites (Table 11). For example, the soil organic matter level at Carman in 2014 was 5.7%. Organic matter alone could have supplied a significant portion of the nitrogen to meet the demand of the wheat crop. Assuming growing season rainfall at Carman was around 250 mm, then soil with an organic matter level of 5.7 % could result in nitrogen mineralization of 124 kg N/ha during the growing season. If 40 kg N/ha is required to grow 1 ton of wheat, 160 kg N /ha would be required for the wheat test crops grown in this experiment. Thus, a large portion of the nitrogen for the wheat grown in the low fertility treatments in the experiment could have come from organic matter mineralization. Another reason could be the well drained nature of these loamy textured soil. Heavy rainfall during the crop growth period in 2014 (Figure 3) could have resulted in leaching of the nitrogen fertilizer applied.

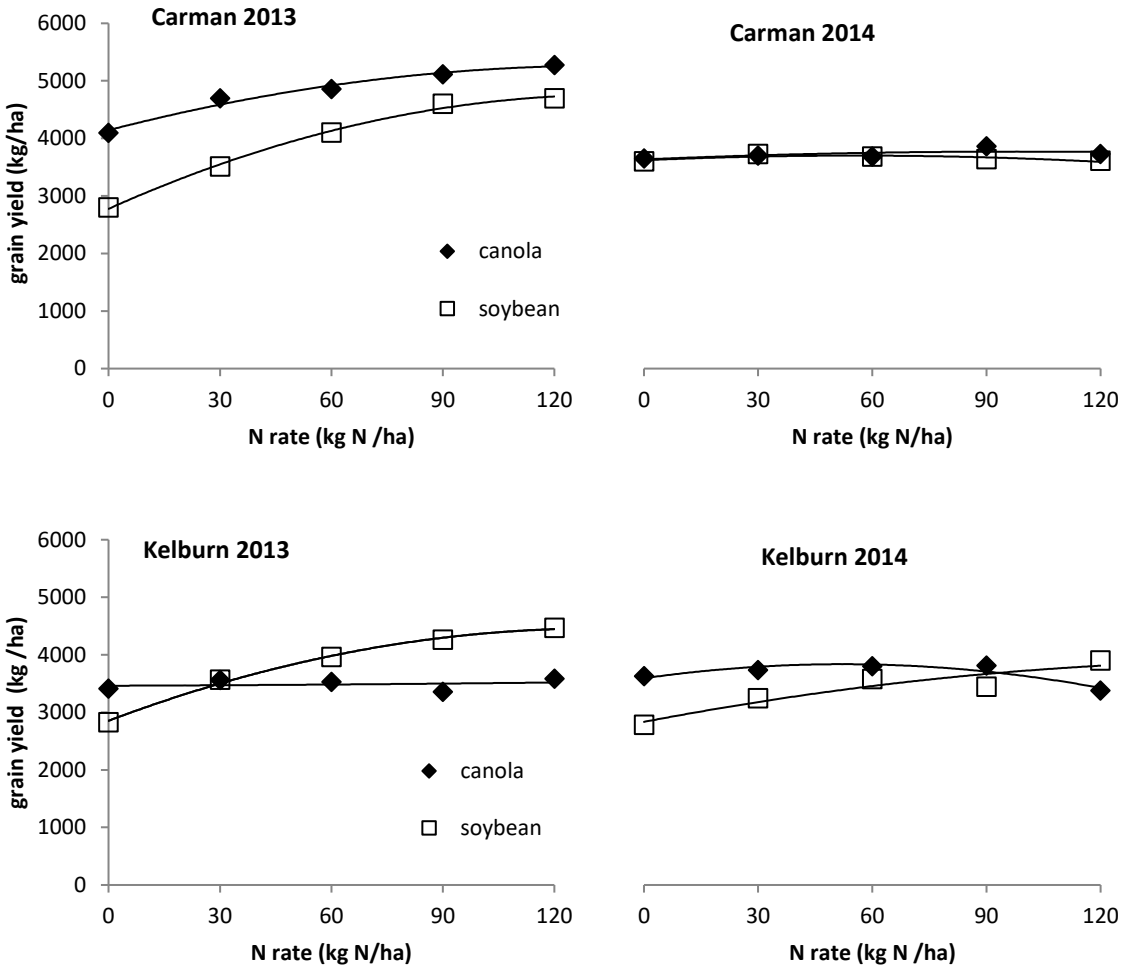

Figure 5: Wheat test crop yield response to nitrogen fertilizer rates following soybean and canola treatment crops at the University of Manitoba Carman Research Farm (Carman) and Richardson International's Kelburn Research Farm (Kelburn). See Table 13 for regression equation parameter estimates.

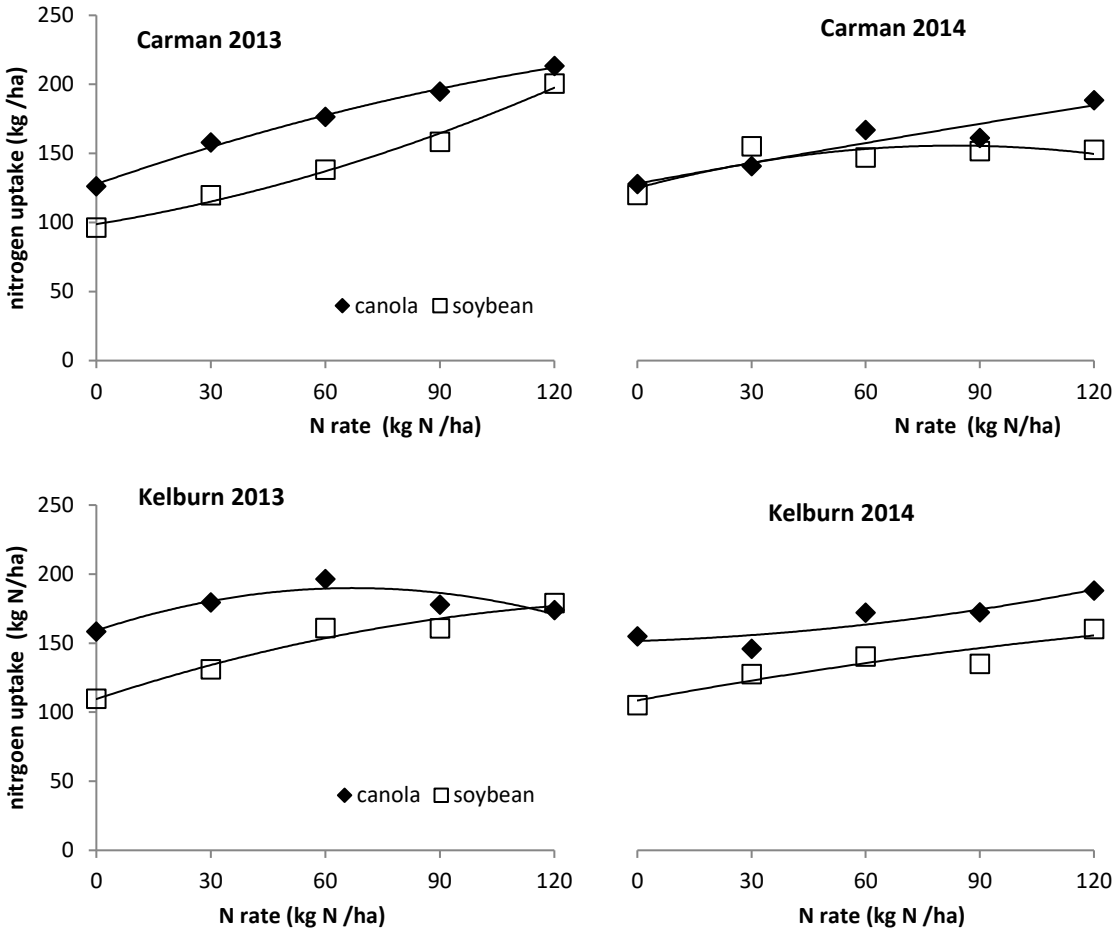

Figure 6 : Wheat test crop nitrogen uptake at the soft dough stage in response to nitrogen fertilizer rates following soybean and canola treatment crops at the University of Manitoba Carman Research Farm (Carman) and Richardson International's Kelburn Research Farm (Kelburn)in 2013 and 2014. See Table 13 for parameter regression equation parameter estimates.

Table 13: Regression equation parameters estimates (standard error in parentheses) describing the yield response and nitrogen uptake response of wheat test crops to nitrogen fertilizer treatments following soybean and canola treatment crops at the University of Manitoba Carman Research Farm (Carman) and Richardson International's Kelburn Research Farm (Kelburn) in 2013 and 2014.

Site	Crops	$[\$]a$	*b*	*c*	R2	Pr>F
Wheat N uptake (kg N/ha)						
Carman 2013	Canola	127.8 (3.03)	0.9563 (0.12)	-0.0021 (0.001)	0.9954	0.0046*
	Soybean	98.72 (5.74)	0.4544 (0.23)	0.0031 (0.002)	0.9882	0.0118*
Carman 2014	Canola	128.9 (10.14)	0.468 (0.40)	0.00005 (0.003)	0.8974	0.1026
	Soybean	125.4 (10.3)	0.726 (0.41)	-0.004 (0.003)	0.706	0.294
Kelburn 2013	Canola	178.2 (16.7)	1.39 (0.66)	-0.010 (0.005)	0.7092	0.2908
	Soybean	113.3 (14.6)	0.812 (0.578)	-0.001 (0.005)	0.9073	0.0927
Kelburn 2014	Canola	151.0 (8.86)	0.084 (0.350)	0.002 (0.003)	0.844	0.156
	Soybean	108.2 (9.44)	0.526 (0.373)	-0.001 (0.003)	0.8758	0.1242
Wheat grain yield (kg /ha)						
Carman 2013	Canola	4143 (92.4)	16.68 (3.7)	-0.062 (0.03)	0.9768	0.0232*
	Soybean	2776 (66.6)	28.9 (2.6)	-0.1054 (0.02)	0.9960	0.0040*
Carman 2014	Canola	3625 (104.7)	4.24 (4.1)	-0.033 (0.03)	0.3453	0.6547
	Soybean	3620 (42.8)	2.92 (1.7)	-0.0262 (0.014)	0.6675	0.3325
Kelburn 2013	Canola	3462 (129.2)	0.23 (5.1)	0.0021 (0.04)	0.0523	0.9477
	Soybean	2854 (55.4)	24.32 (2.2)	-0.092 (0.018)	0.9959	0.0041*
Kelburn 2014	Canola	3592 (92.2)	9.39 (3.6)	-0.0899 (0.029)	0.8507	0.1493
	Soybean	2834 (187.8)	12.62 (7.4)	-0.0374 (0.059)	0.8847	0.1153

[$]Quadratic function equation: $y = a + bx + cx^2$ where a = intercept, b =linear coefficient and c = curvilinear coefficient; y is grain yield (kg /ha) and x is fertiliser rate (kg N /ha);* significant at Pr<0.05

Table 14 : Table of treatment Means and ANOVA p-vales for the effect of previous crop (crop) and nitrogen fertiliser (fert) rates on wheat biomass at soft dough stage at four site years (site) including the University of Manitoba Carman Research Farm (Carman) and Richardson International's Kelburn Research Farm (Kelburn) in 2013 and 2014.

Previous crop		Site				Mean of all sites
		Carman 2013	Carman 2014	Kelburn 2013	Kelburn 2014	
		Biomass (kg /ha)				
Canola		11019	9301	9003	9554[a]	9719[a]
Soybean		9793	9021	8824	7712[b]	8837[b]
LSD (P<0.05)		s	s	s	343	187
fertiliser means						
0		8141[c]	8415	7797	7553[c]	7976[c]
30		10122[b]	9206	8597	8256[bc]	9045[b]
60		10631[ab]	9329	9594	8813[ab]	9592[ab]
90		10991[ab]	9494	9234	9072[ab]	9698[a]
120		12144[a]	9362	9344	9472[a]	10080[a]
LSD(P<0.05)		570	s	s	373	230
3-way ANOVA						Pr>F
Site	3					0.0004*
crop	**1**					**0.0027***
site *crop	3					0.1064
fert	**4**					**<.0001****
site *fert	12					0.1683
crop *fert	4					0.1363
site *crp*fert	12					0.544
rep (site *crop)	24					0.0112*
residual CV (%)						18.4

s LSD not reported due to no significant main factor effect; site and site interaction were considered as fixed effects
* significant at p<0.05
** highly significant at p<0.0001
a-d Mean values followed by same letter (within columns) are not significantly different

Table 15: Table of treatment Means and ANOVA p-vales for the effect of previous crop (crop) and nitrogen fertiliser (fert) rates on nitrogen uptake by wheat at soft dough stage at four site years (site) including the University of Manitoba Carman Research Farm (Carman) and Richardson International's Kelburn Research Farm (Kelburn) in 2013 and 2014.

Previous crop		Site				Mean of all sites
		Carman 2013	Carman 2014	Kelburn 2013	Kelburn 2014	
		N uptake (kg /ha)				
Canola		174[a]	157	177	167[a]	169[a]
Soybean		143[b]	145	148	134[b]	143[b]
LSD (p<0.05)		6	s	s	8	4
Fertiliser means						
0		111[d]	124[c]	134[b]	130[d]	125[d]
30		139[cd]	148[b]	155[ab]	135[cd]	145[c]
60		157[bc]	157[ab]	179[a]	156[ab]	162[b]
90		177[b]	156[ab]	169[a]	154[bc]	164[b]
120		207[a]	171[a]	177[a]	174[a]	182[a]
LSD (p<0.05)		10	7	12	8	5
3-way ANOVA						Pr>f
Site	3					0.2822
crop	**1**					**<.0001****
site *crop	3					0.4526
Fert.	**4**					**<.0001****
site *fert.	12					0.0665
crop *fert.	4					0.673
site *crop *fert.	12					0.3847
rep (site*crop)	24					0.0216*
Residual CV (%)						23

s LSD not reported due to no significant main factor effect; site and site interaction were considered as fixed effects
* significant at p<0.05
** highly significant at p<0.0001
a-d Mean values followed by same letter (within columns) are not significantly different

In one of the experiments, weeds played a role in the lack of wheat yield response to nitrogen fertilizer treatments. At Kelburn in 2013, there was significant weed pressure in the wheat test crop following canola and the wheat following the canola treatment crop did not have any response to N fertilizer. Weed biomass and nitrogen uptake was quantified and was found to be significantly higher in in the wheat crop following canola than wheat crop following soybean at soft dough stage of wheat (data not shown). Planter error at Kelburn 2014, also played a role as an accidental seeding into the plot area from a neighboring field resulted in the application of extra nitrogen at 50 kg N /ha in two of the four replicates. Fusarium head blight might have also played a role at Carman in 2014, however this was never quantified.

Was there a nitrogen credit from soybeans?

The nitrogen credit of a nitrogen fixing crop is determined by comparing the amount of nitrogen fertilizer required for a non-legume test crop that follows a non-legume reference crop to produce the same yield as the test crop grown after a legume (Mahler and Auld, 1989; McEwen *et al.* 1989). Thus, a legume crop provides a nitrogen credit to subsequent crops when test crop yields following the legume crop are higher than when following the non-nitrogen fixing reference crop. In three of the four site years, yields of the wheat test crop following soybeans was the same or lower compared to the wheat test crop following the canola reference crop. In the one site year of the study where the wheat test crop following both soybeans and canola responded to nitrogen fertilizer treatments, Carman 2013, the yield of wheat following canola was always higher than following soybeans across the range of fertilizer rates tested. It is also important to note that the one site year where wheat yield response was higher following soybean than following canola in Kelburn 2014, was the experiment with poor weed control in wheat test crop following canola. Thus, this patter was likely the result of wheat yield loss due to weeds following canola, rather than a wheat yield increase as a result of the soybean treatment crop.

Two factors are playing a role in the lack of nitrogen credit following soybeans. The first is the carbon to nitrogen ratios of canola and soybean residue. The carbon to nitrogen ratio of canola residue was lower than for soybean (Table 16) which would result in less immobilization. The other large contributing factor is the application of nitrogen fertilizer to the canola crop in the treatment crop year. No fertilizer was applied to soybean in the treatment crop year. These two differences were observed in the spring soil tests that were taken before wheat test crop planting (Table 17) Other studies have reported that soybean nitrogen credits, relative to cereal crops such as corn or wheat, are predominately the result of greater nitrogen release from soybean residues that are more rapidly mineralized and taken up by subsequent crop (Bundy *et al*., 1993; Vanotti and Bundy, 1995; Power *et al*., 1986).

Table 16: Soybean and canola treatment crop yield, biomass, harvest index (HI), crop residue carbon to nitrogen ratio (C:N), and nitrogen in crop residues at the University of Manitoba Carman Research Farm (Carman) and Richardson International's Kelburn Research Farm (Kelburn) in fall of 2012 and 2013. This data characterizes the residues that were incorporated from the treatment crops the fall prior to growing the wheat test crop in 2013 and 2014.

Crop	Site	Grain yield (kg /ha)	Biomass yield (kg /ha)	HI	C:N	Nitrogen in crop residue (kg/ha)
Canola	Carman 2012	1574	4014	0.25	54	33 (25-39) *
	Kelburn 2012	1289	7795	0.13	39	90 (79-100)
	Carman 2013	2663	3418	0.41	32	45(40-50)
	Kelburn 2013	3140	5913	0.34	42	61 (38-86)
Soybean	Carman 2012	2677	5012	0.39	65	34 (20-48)
	Kelburn 2012	2460	-	-	-	-
	Carman 2013	4524	4891	0.48	47	46 (40-50)
	Kelburn 2013	4822	3915	0.55	71	25 (21-32)

Table 17: Residual spring soil profile nitrate (0-120 cm) prior to winter wheat seeding and wheat test crop nitrogen update at the soft dough stage following soybean and canola treatment crops in the no nitrogen control treatment at the University of Manitoba Carman Research Farm (Carman) and Richardson International's Kelburn Research Farm (Kelburn) in 2013 and 2014.

Site	Previous crop		*Nitrogen balance
	Canola	Soybean	
	Total residual soil nitrate (0-120 cm) in ppm before wheat seeding		
Carman 2013	48	14	-35
Carman 2014	51	23	-29
Kelburn 2013	106	32	-75
Kelburn 2014	140	65	-76
	N uptake (kg N /ha)of wheat biomass at soft dough stage		
Carman 2013	126	96	-30
Carman 2014	128	120	-8
Kelburn 2013	158	110	-49
Kelburn 2014	155	105	-50

*Nitrogen balance is the difference between available soil N or N uptake by the wheat test crop following soybean in no N fertiliser treatment and wheat following canola in no N fertiliser treatment

Soil nitrogen status following soybean crops

One of the clear lessons learned from this study for farmers in Manitoba is that soil fertility recommendations for crops following soybeans should be approached differently compared to crops following canola in rotation. Soil nitrate N concentrations were lowest following soybean in all site years when compared to canola (Table 17). Other studies have reported that soybeans result in a net depletion of soil nitrogen (Zapata *et al*. 1987). This is driven by the high protein content of soybean seeds that is harvested relative to the amount of nitrogen that is fixed by the plant. Salvagiotti *et al*. (2008) found that soybean requires 8 kg of nitrogen to produce 100 kg of seed, of which 6 kg of that nitrogen is removed in the seed. In that same study, the source of nitrogen for soybeans was found to be 50-80 % derived from nitrogen fixation and 20-50% from soil mineral nitrogen. In comparison, to produce a 100 kg of seed, canola plant takes 6-7 kg of N, of which 3-4 kg is removed with the seed (Canola Council of Canada). Thus, more fertilizer may be required to grow canola than for soybean, but a greater proportion of the nitrogen may be left in the field through crop residues after harvest to be mineralized for subsequent crops. Differences will aslo be driven by fetilizer applied to canola crops compared to soybean crops. If this fertilizer is not all taken up by the canola crop during the growing season, it may accumulate in the soil profile.

Differences between canola and soybean are also a result of the nitrogen in the crop residues after harvest. The total amount of nitrogen in soybean crop residue was smaller than the amount in canola crop residue. The percent of nitrogen content of soybean residues was 0.68-0.90% compared to canola residue at 0.87-1.22%. Thus, nitrogen returned to the soil by soybean residues ranging between 25-46 kg N /ha while canola residues returned 33-90 kg N/ha (Table 20). Similar results had been reported in Alberta, where pea crop residues returned 22 kg N /ha while canola residue returned 50 kg N /ha (Soon and Clayton, 2002).

Table 18: Soybean and canola treatment crop yield, biomass, Harvest Index (HI), crop residue carbon to nitrogen ratio (C:N), and nitrogen in crop residues at the University of Manitoba Carman Research Farm (Carman) and Richardson International's Kelburn Research Farm (Kelburn) in fall of 2012 and 2013. This data characterizes the residues that were incorporated from the treatment crops the fall prior to growing the wheat test crop in 2013 and 2014.

Crop	Site	Grain yield (kg /ha)	Biomass yield (kg /ha)	HI	C:N	Nitrogen in crop residue (kg/ha)
Canola	Carman 2012	1574	4014	0.25	54	33 (25-39) *
	Kelburn 2012	1289	7795	0.13	39	90 (79-100)
	Carman 2013	2663	3418	0.41	32	45(40-50)
	Kelburn 2013	3140	5913	0.34	42	61 (38-86)
Soybean	Carman 2012	2677	5012	0.39	65	34 (20-48)
	Kelburn 2012	2460	-	-	-	-
	Carman 2013	4524	4891	0.48	47	46 (40-50)
	Kelburn 2013	4822	3915	0.55	71	25 (21-32)

*values in brackets are the range of N added to soil based on 2-4 samples or replicates

Differences beteween canola and soybeans are also driven by their contrasting maturity and harvest periods in the fall. Canola was harvested 37-42 days earlier than soybean ove the four site years of this experiment. Earleir harvest for canola results in that there is more time for decompositon of crop residues and mineratilization of nitrogen. The longer growing season of soybean extends the lengh of thime that it can take up nitrogen from the soil profile. At the time of canola harvest, soybean is still in one of the highest nitrogen demanding growth stage of mid-pod filling (stage R5) and the demand for nitrogen continues untill the end of maturity (stage R7) (Zapata *et al.* 1987). Thus, soil nitrogen was depleted in soybean plots for longer period of time than in canola plots where crop had already been harvested.

Conclusions

Soybeans are a profitable crop for farmers because they yield well with relatively few inputs. Although soil bacteria and fungi aren't necessarily at the top of the list of concerns for most farmers, this research shows that both *Bradyrhizobium* and Arbuscular Mycorrhizal Fungi play an important role in nitrogen and phosphorus acquisition, and are significantly affected by crop rotation decisions even after one field season. Farmers growing soybeans in low fertility areas could especially benefit by an increased awareness of management decisions that benefit *Bradyrhizobium japonicum* and Arbuscular Mycorrhizal Fungi to acquire valuable nutrients, such as nitrogen and phosphorus, that are needed by soybeans.

Soybean did not provide a nitrogen credit to the wheat test crops in this study, rather soybean showed negative nitrogen balance. Soybean treatment crops resulted in lower spring soil test compared to canola treatment crops. The total amount of nitrogen in soybean crop residue was smaller than the amount in canola crop residue. The percent of nitrogen content of soybean residues was 0.68-0.90 % compared to canola residues at 0.87-1.22 %. Thus, nitrogen returned to the soil by soybean residues ranging between 25-46 kg N /ha while canola residues returned 33-90 kg N/ha.

The carbon to nitrogen ratio of soybean residues were higher than expected. They ranged between 47-71, with an average carbon to nitrogen ratio of 61. The carbon to nitrogen ratio of soybean was higher than the carbon to nitrogen ratio of canola residue, which ranged from 32-54 and averaged 42. Larger carbon to nitrogen ratios result in increased immobilization of soil nitrogen as crop residues are decomposed by soil organisms. The growing season of canola was approximately 37-42 days shorter than that of soybean. Thus, canola residues had longer period to decompose than soybean residues before winter. Additionally, nitrogen immobilized as a result of canola residues in early autumn could be released before the onset of winters (Soon and Arshad, 2002), but this would be unlikely for nitrogen from soybean residues.

Extension of Research Project Results

Results from this research project were presented to farmers at several field days and production meetings including: the Manitoba Pulse and Soybean Growers (MPSG) 2015 Soybean Management and Research Transfer Day, the 2016 MPSG Getting It Right Workshop. News articles about his research have appeared in MPSG's Pulse Beat Magazine and in Top Crop Manager Magazine. Results were also presented to a scientific audience at the 2015 Canadian Society of Agronomy Annual meeting as part of the Botany 2015 conference held in Edmonton, AB and at the University of Manitoba 2014 Plant Science Graduate Students Symposia.

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