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Effects of broad-leaf crop frequency in various rotations on soil organic C and N, and inorganic N in a Dark Brown soil

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ABSTRACT

The objective of this study was to determine the impact of frequency of broad-leaf crops canola and pea in various crop rotations on pH, total organic C (TOC), total organic N (TON), light fraction organic C (LFOC) and light fraction organic N (LFON) in the 0 - 7.5 and 7.5 - 15 cm soil depths in autumn 2009 after 12 years (1998-2009) on a Dark Brown Chernozem (Typic Boroll) loam at Scott, Saskatchewan, Canada. The field experiment contained monoculture canola (herbicide tolerant and blackleg resistant hybrid) and monoculture pea compared with rotations that contained these crops every 2-, 3-, and 4-yr with wheat. There was no effect of crop rotation duration and crop phase on soil pH. Mass of TOC and TON in the 0 - 15 cm soil was greater in canola phase than pea phase in the 1-yr (monoculture) and 2-yr crop rotations, while the opposite was true in the 3-yr and 4-yr crop rotations. Mass of TOC and TON (averaged across crop phases) in soil generally increased with increasing crop rotation duration, with the maximum in the 4-yr rotation while no difference in the 1-yr and 2-yr rotations. Mass of LFOC and LFON in soil was greater in canola phase than pea phase in the 1-yr, 2-yr and 3-yr rotations, but the opposite was true in the 4-yr rotation. There was no consistent effect of crop rotation duration on mass of LFOC and LFON. The N balance sheet over the 1998 to 2009 period indicated large amounts of unaccounted N for monoculture pea, suggesting a great potential for N loss from the soil-plant system in this treatment through nitrate leaching and/or denitrification. In conclusion, the findings suggest that the quantity of organic C and N can be maximized by increasing duration of crop rotation and by including hybrid canola in the rotation.

Keywords: Broad-Leaf Crops; Canola; Frequency; Light Fraction Organic C; Light Fraction Organic N; Pea; Total Organic C; Total Organic N

1. INTRODUCTION

Crop rotation (the growing of different crops in the same field in a planned sequence) is often practiced to mitigate pests that often become unmanageable in monocultures (cultivation of the same crop year after year on the same field) [1,2]. In western Canada, research has indicated that rotations balanced between broad-leaf crops (canola and pea) with cereals (wheat and barley) tended to have less pest problems and lower production risk than rotations that were heavily cereal or broad-leaf based [3]. In many years, canola and/or pea provide the best economic return to producers compared to other field crops grown in western Canada. For this reason production of canola or pea is often intensive, meaning it is grown more than once every four years on the same field.

Research has shown that organic C in soil can be maintained or enhanced by combining reduced tillage or no-tillage with proper crop rotations that increase input of crop residues to soil and/or decrease their decomposition [4-8]. However, the magnitude of change in organic C in soil may vary with crop type/species/diversity/intensity, rooting characteristics (layer/volume/mass) of each crop, soil type and climatic conditions [9,10]. Our previous paper has discussed the impacts of frequency of broad-leaf crops canola and pea in various crop rotations on accumulation and distribution of nitrate-N and extractable P in the soil profile after 8 years [11]. The objective of this study was to determine the impact of frequency of broad-leaf crops canola and pea in various crop rotations (grown in monoculture or in rotation with wheat) on total organic C (TOC), total organic N (TON), light fraction organic C (LFOC), light fraction organic N (LFON) and pH in the 0 - 7.5 and 7.5 - 15 cm soil depths in autumn 2009 after 12 years (1998-2009) on a Dark Brown Chernozem (Typic Boroll) loam at Scott, Saskatchewan, Canada.

2. MATERIALS AND METHODS

A 12-yr field experiment was conducted from 1998 to 2009 on a Dark Brown Chernozem (Typic Boroll) loam at Scott, Saskatchewan. The field experiment was designed as split-plot of 12 crop rotations (main plots) and two fungicide (treated and untreated) treatments applied to sub-plots (**Table 1**), with all phases of each rotation present every year in four replications. For this area, the long-term average precipitation in the growing season (from May to August) is about 210 mm. Growing season precipitation was substantially below average in 1998, 2001 and 2008, fairly below average in 2002, 2003 and 2004, slightly below average in 2000, 2006 and 2009, above average in 1999 and 2007, and very wet in 2005. All crops were seeded with a Versatile Noble hoe drill set at 23-cm row spacing. Seeding rate was 245 $kg \cdot ha^{-1}$ for pea, 7 - 9 $kg \cdot ha^{-1}$ for canola, 100 $kg \cdot ha^{-1}$ for wheat and 50 - 62 $kg \cdot ha^{-1}$ for flax. All plots received monoammonium phosphate seed placed to supply adequate amounts of P (15 $kg \cdot P \cdot ha^{-1}$ plus 7 $kg \cdot N \cdot ha^{-1}$) and nitrogen (N) as 46-0-0 midrow banded at the time of seeding at rates based on soil test recommendations (on average ranged from 11 to 60 $kg \cdot N \cdot ha^{-1}$, with no N fertilizer to pea) for optimum crop growth and yield. Appropriate herbicides were applied to control annual weeds. The crops were harvested for seed yield every year and straw was returned to each corresponding plot/treatment.

In the autumn of 2009, soil samples in selected treatments were obtained by taking 10 cores (about 2.4 cm diameter) from the 0 - 7.5, 7.5 - 15 and 15 - 20 cm layers. Bulk density of soil was determined by the core method using soil weight and core volume [12]. The soil samples were air dried at room temperature after removing coarse roots and easily detectable crop residues, and ground to pass a 2-mm sieve. Sub-samples were pulverized in a vibrating-ball mill (Retsch, Type MM2, Brinkman Instruments Co., Toronto, Ontario) for determination of TOC, TON, LFOC and LFON in soil. Soil samples used for organic C and N analyses were tested for the presence of inorganic C (carbonates) using dilute HCl, and none was detected in any soil sample. Therefore, C in soil associated with each fraction was considered to be of organic origin. Total organic C in soil was measured by Dumas combustion using a Carlo Erba instrument (Model NA 1500, Carlo Erba Strumentazione, Italy), and Technicon Industrial Systems [13] method was used to determine TON in the soil. Light fraction organic matter (LFOM) was separated using a NaI solution of 1.7 $Mg \cdot m^{-3}$ specific gravity, as described by Janzen *et al.* [6] and modified by Izaurralde *et al.* [14]. The C and N in LFOM (LFOC, LFON) were measured by Dumas combustion. Soil samples (ground to pass a 2-mm sieve) from the 0 - 7.5 and 7.5 - 15 cm layers were also monitored for pH in 0.01 M $CaCl_2$ solution with a pH meter.

In autumn 2009, soil samples were also obtained by taking 4 cores (using 4-cm diameter coring tube) from the 0 - 15, 15 - 30 and 30 - 60 cm layers. The bulk density of soil was determined by the core method using soil weight and core volume [12]. The soil samples were air dried at room temperature, ground to pass a 2-mm sieve, and then analyzed for ammonium-N [15] and nitrate-N [16] by extracting soil in a 1:5 ratio of soil and 2 M KCl solution.

The cumulative amounts of crop residue (CR) input from 1998 to 2009 growing seasons were estimated as: above-ground residue (AGR) + belowground residue (BGR) returned to soil. The AGR was determined from the straw yield of each crop from 1998 to 2009 growing seasons. The BGR was estimated from grain dry weight

Table 1. Description of crop rotations in a field experiment from 1998 to 2009 at Scott Saskatchewan.

Crop rotation name (duration)	Crop rotation	Input of C from crop residue in 12 years ($kg \cdot C \cdot ha^{-1}$)
Monoculture (1-yr)	Monoculture hybrid **canola**	20,847
Monoculture (1-yr)	Monoculture **pea**	9930
2-year rotation (2-yr)	Hybrid **canola**-wheat	26,697
2-year rotation (2-yr)	**Pea**-wheat	18,482
3-year rotation (3-yr)	Pea-hybrid **canola**-wheat	20,724
3-year rotation (3-yr)	**Pea**-hybrid canola-wheat	22,705
4-year rotation (4-yr)	Hybrid **canola**-wheat-pea-wheat	24,854
4-year rotation (4-yr)	Hybrid canola-wheat-**pea**-wheat	23,910

(GDW) and AGR, using the formula: BGR = a (GDW + AGR). The value of the constant "a" was 0.24 for wheat and 0.25 for canola [17], and 0.25 for pea. The amounts of crop residue C input were estimated by multiplying the concentration of total C by the amount of crop residue input in various rotation treatments. The estimated C concentration was 45% for wheat, 42% for canola [18], and 40% for pea [19]. The cumulative amounts of crop residue C were calculated as: CR-C = AGR-C + BGR-C; for 1998 to 2009 (12 years). The estimated amounts of N balance and unaccounted N over the 1998 to 2009 period for the 8 treatments [two crop phases (canola and pea) and four crop rotation durations (1-yr, 2-yr, 3-yr and 4-yr)]; **Table 7** was calculated as the difference between total of N applied as fertilizers in 12 years + N fixed by pea in rotations with pea + N added in seed at seeding in 12 years minus N removed in seed in 12 years (for N balance) + nitrate-N recovered in soil in autumn 2009 (for unaccounted N).

The data for each parameter were subjected to analysis of variance (ANOVA) using procedures as outlined in SAS [20]. Significant ($P \leq 0.05$) differences between treatments were determined using LSmeans (Proc GLM, SAS 6.1 for windows). The least significant difference ($LSD_{0.05}$) test was used to compare treatment means for various parameters. Correlations between mass of organic C or N in soil in autumn 2009 and the amount of crop residue C input from 1998 to 2009 growing seasons were calculated using the linear (REG) procedure.

3. RESULTS AND DISCUSSION

3.1. Soil pH

There was no significant effect of crop rotation duration and crop phase on pH in the 0 - 7.5 and 7.5 - 15 cm soil layers (data not shown). The soil pH ranged from 4.9 to 5.5 in the 0 - 7.5 cm layer and from 4.7 to 5.6 in the 7.5 to 15 cm layer among different treatments. Similar to a previous study at this site, there was no consistent effect of crop diversification/rotation on soil pH [21]. Because soil at this site is already fairly acid, acidification of soil from application of N fertilizer at moderate rates or from including pea legume in the rotation is not expected to be a serious problem in this soil site.

3.2. Organic C and N Fraction in Soil

The mass of TOC and TON in soil varied with the crop rotation duration and/or frequency of canola or pea in the rotation, but the crop rotation duration × crop phase interaction effect was significant only for TOC ($P \leq 0.10$) and TON ($P \leq 0.05$) in the 7.5 - 15 cm soil layer (**Table 2**). For example, mass of TOC and TON in the 0 - 15 cm soil tended to be greater in canola phase than pea phase in the 1-yr (monoculture) and 2-yr crop rotations, while the opposite was true in the 3-yr and 4-yr crop rotations. Averaged across crop phases, mass of TOC and TON in soil generally increased with increasing crop rotation duration (but significant only for TOC ($P \leq 0.06$) and TON ($P \leq 0.05$) in the 7.5 - 15 cm soil layer, with the maximum in the 4-yr crop rotation while no difference in the 1-yr and 2-yr rotations. When ANOVA was conducted separately for canola and pea phases, the ANOVA for canola phase did not indicate any significant effect of crop rotation duration and/or frequency of canola in the rotation on TOC and TON. However, the ANOVA for pea phase showed significant effect of crop rotation duration and/or frequency of pea in the rotation on TOC and TON in the 7.5 - 15 ($P \leq 0.05$) soil layer and in the total 0 - 15 ($P \leq 0.14$) cm soil depth. Mass of TOC and TON (averaged across crop rotation duration) tended to be greater (but not significant) in canola phase than pea phase.

Of interest, despite markedly lower C inputs, TOC under the continuous pea treatment was not greatly dissimilar to other treatments. Similar results have been reported by other workers. In a long-term study on a Brown Chernozem, Campbell *et al.* [22] reported lower C inputs but similar SOC status for a wheat-lentil compared to a continuous wheat treatment. In an incubation study using ^{13}C-CO_2 to label C inputs from growing crops, Comeau [23] estimated significantly lower C inputs to soil under pea compared to canola, but similar amounts of ^{13}C "stabilized" in the soil at the end of two growing cycles. These results support the hypothesis that pea residues are more efficiently stabilized as SOC than wheat and particularly canola.

The mass of LFOC and LFON in soil varied greatly with the duration/frequency of canola/pea in the crop rotations, but the crop rotation duration × crop phase interaction effect was significant only for TOC ($P \leq 0.12$) in the 0 - 7.5 cm soil layer (**Table 3**). Like TOC and TON, mass of LFOC and LFON in soil tended to be greater in canola phase than pea phase in the 1-yr and 2-yr rotations, but the opposite was true in the 3-yr and 4-yr rotations. Compared to 1-yr rotation, mass of LFOC and LFON (averaged across crop phases) in soil decreased with 2-yr and 3-yr rotations, and then increased, with greater LFOC and LFON in soil for 4-yr than 1-yr rotations in the 0 - 7.5 cm layer, but again the effect was significant only for LFOC ($P \leq 0.12$) in the 0 - 7.5 cm soil layer. When ANOVA was conducted separately for canola and pea phases, there was no significant effect of crop rotation duration and/or frequency of canola in the rotation on LFOC and LFON for canola phase. However, for pea phase, the ANOVA showed significant effect of crop rotation duration and/or frequency of canola in the rotation on LFOC in the 0 - 7.5 cm soil layer ($P \leq 0.06$)

Table 2. Effect of broad-leaf crop phase and frequency over 12 years from 1998 to 2009 on mass of total organic C (TOC) and total organic N (TON) in soil in autumn 2009 at Scott, Saskatchewan, Canada.

Treatment		TOC mass ($Mg \cdot C \cdot ha^{-1}$) in soil layers (cm)			TON mass ($Mg \cdot N \cdot ha^{-1}$) in soil layers (cm)		
Crop rotation (duration)	Crop phase	0 - 7.5	7.5 - 15	0 - 15	0 - 7.5	7.5 - 15	0 - 15
Monoculture (1-year)	Canola	24.77	21.57	46.34	2.198	1.965	4.163
Monoculture (1-year)	Pea	22.33	21.95	44.28	2.007	1.960	3.967
Canola-wheat (2-year)	Canola	26.08	23.99	50.07	2.253	2.166	4.419
Pea-wheat (2-year)	Pea	21.19	20.00	41.19	1.858	1.787	3.645
Pea-canola-wheat (3-year)	Canola	23.55	24.03	47.58	2.045	2.103	4.148
Pea-canola-wheat (3-year)	Pea	23.14	26.81	49.95	2.086	2.388	4.474
Canola-wheat-pea-wheat (4-year)	Canola	24.75	24.95	49.70	2.152	2.208	4.360
Canola-wheat-pea-wheat (4-year)	Pea	26.00	25.56	51.56	2.222	2.256	4.478
	$LSD_{0.05}$	ns	4.74	ns	ns	0.387	ns
	SEM (significance)	1.589^{ns}	1.612$^{\bullet}$	2.712^{ns}	0.1348^{ns}	0.1316$^{\bullet}$	0.2330^{ns}
Crop rotation duration							
1-year		23.55	21.76	45.31	2.103	1.962	4.065
2-year		23.64	21.99	45.63	2.055	1.977	4.032
3-year		23.35	25.42	48.76	2.066	2.246	4.312
4-year		25.37	25.25	50.63	2.187	2.232	4.419
$LSD_{0.05}$		ns	3.44	ns	ns	0.291	ns
SEM (significance)		1.151^{ns}	1.178$^{\bullet}$	2.015^{ns}	0.0965^{ns}	0.0997$^{\bullet}$	0.1701^{ns}
	Crop phase						
	Canola	24.79	23.63	48.42	2.162	2.111	4.273
	Pea	23.17	23.58	46.75	2.044	2.098	4.141
	$LSD_{0.05}$	ns	ns	ns	ns	ns	ns
	SEM (significance)	0.814^{ns}	0.833^{ns}	1.425^{ns}	0.0682^{ns}	0.0705^{ns}	0.1203^{ns}
Additional statistical comparisons							
Significance of four crop rotation durations for canola phase only							
	$LSD_{0.05}$	ns	ns	ns	ns	ns	ns
	SEM (significance)	1.109^{ns}	1.670^{ns}	2.515^{ns}	0.0829^{ns}	0.1458^{ns}	0.2108^{ns}
Significance of four crop rotation durations for pea phase only							
	$LSD_{0.05}$	ns	5.00	10.18	ns	0.398	0.832
	SEM (significance)	2.135^{ns}	1.564	3.183$^{\bullet}$	0.1856^{ns}	0.1244*	0.2599$^{\bullet}$

$^{\bullet}$, * and ns refer to significant treatment effects in ANOVA at $P \le 0.1$, $P \le 0.05$ and not significant, respectively.

Table 3. Effect of broad-leaf crop phase and frequency over 12 years from 1998 to 2009 on mass of light fraction organic C (LFOC) and light fraction organic N (LFON) in soil in autumn 2009 at Scott, Saskatchewan, Canada.

Treatment		LFOC mass ($kg·C·ha^{−1}$) in soil layers (cm)			LFON mass ($kg·N·ha^{−1}$) in soil layers (cm)		
Crop rotation (duration)	Crop phase	0 - 7.5	7.5 - 15	0 - 15	0 - 7.5	7.5 - 15	0 - 15
Monoculture (1-year)	Canola	3283	889	4172	212	48	260
Monoculture (1-year)	Pea	2520	694	3214	168	42	210
Canola-wheat (2-year)	Canola	2733	831	3564	171	46	217
Pea-wheat (2-year)	Pea	2317	814	3131	154	49	203
Pea-canola-wheat (3-year)	Canola	2193	816	3009	150	47	197
Pea-canola-wheat (3-year)	Pea	2723	962	3685	176	54	230
Canola-wheat-pea-wheat (4-year)	Canola	3081	923	4004	186	51	237
Canola-wheat-pea-wheat (4-year)	Pea	3348	855	4203	201	48	249
	$LSD_{0.05}$	918	ns	ns	ns	ns	ns
	SEM (significance)	$312.0^{•}$	110.5^{ns}	365.9^{ns}	18.5^{ns}	6.3^{ns}	22.7^{ns}
Crop rotation duration							
1-year		2901	792	3693	190	45	235
2-year		2525	822	3347	162	47	209
3-year		2458	889	3347	163	50	213
4-year		3215	889	4014	193	49	242
$LSD_{0.05}$		677	ns	ns	ns	ns	ns
SEM (significance)		$231.8^{•}$	77.2^{ns}	273.1^{ns}	13.4^{ns}	4.3^{ns}	16.2^{ns}
	Crop phase						
	Canola	2823	865	3688	180	48	228
	Pea	2727	831	3558	175	48	223
	$LSD_{0.05}$	ns	ns	ns	ns	ns	ns
	SEM (significance)	163.9^{ns}	54.6^{ns}	193.1^{ns}	9.5^{ns}	3.0^{ns}	11.5^{ns}
Additional statistical comparisons							
Significance of four crop rotation durations for canola phase only							
	$LSD_{0.05}$	ns	ns	ns	ns	ns	ns
	SEM (significance)	334.5^{ns}	125.2^{ns}	430.8^{ns}	21.9^{ns}	7.2^{ns}	27.1^{ns}
Significance of four crop rotation durations for pea phase only							
	$LSD_{0.05}$	769	ns	818	ns	ns	ns
	SEM (significance)	$240.3^{•}$	112.4^{ns}	255.8^{*}	13.2^{ns}	6.3^{ns}	17.6^{ns}

•, * and ns refer to significant treatment effects in ANOVA at P ≤ 0.1, P ≤ 0.05 and not significant, respectively.

and in the total 0 - 15 cm soil depth ($P \leq 0.05$), but for LFON it was significant at $P \leq 0.16$ in the 0 - 7.5 soil layer. Mass of LFOC and LFON (averaged across crop rotation duration) in soil tended to be greater in canola phase than pea phase in most cases.

There were strong and highly significant positive correlation coefficients between TOC and TON, and between LFOC and LFON fractions in soil (**Table 4**). The correlations between TOC and LFOC (significant at $P \leq 0.12$), and between TON and LFON (significant at $P \leq 0.10$) were moderate. The crop residue C or N input over 12 growing seasons (**Table 1**) had significant correlation coefficients with TOC (significant at $P \leq 0.05$) and TON (significant at $P \leq 0.06$). For linear regressions between crop residue C input and TOC, TON, LFOC or LFON, the R^2 values were significant (significant at $P \leq 0.05$) for TOC and TON (**Table 5**).

In an 8-yr study with 4-yr rotations on a Black Chernozem soil in northeastern Saskatchewan, previous research has shown no effect of broad-leaf frequency (1, 2 or 3 broad-leaf crops in 4-yr rotations) on organic C and N in soil [10]. In another study on a Brown Chernozem soil in southern Saskatchewan, annually cropped rotations stored more organic C in soil than crop rotations with bare summer fallow, but there was no influence of crop phase on soil organic C under continuously cropped rotations [9]. However in our present 12-yr study, TOC, TON, LFOC and LFON all increased (although significant only in a few cases) with increasing duration of crop rotation, with the maximum soil organic C and N in the 4-yr rotations and also greater organic C and N in soil in canola phase than pea phase. It is possible that inclusion of other crops, such as wheat with high lignin content in straw which is relatively slow to decompose, may have resulted in this slow build-up of organic C and N in soil in the longer duration rotations, especially 4-year rotation with two wheat crops.

Because of relatively much smaller amounts of annual inputs of crop residue C or N compared to the amounts of organic C or N stocks in soil, there is generally a slow build-up of organic C or N in soil. This makes it very difficult to detect significant increase in storage of organic C or N in soil due to management practices, especially from crop rotations where crop residues (roots and

Table 4. Relationships among organic C or N fractions (TOC, TON, LFOC, LFON) in the 0 - 15 cm soil, or between crop residue input from 1998 to 2009 growing seasons and organic C or N stored in the 0 - 15 cm soil sampled in autumn 2009 at Scott, Saskatchewan, Canada.

Parameter	Correlation coefficients (r)			
	TOC	**TON**	**LFOC**	**LFON**
		Relationships among soil organic C or N fractions		
TOC		0.986^{***}	$0.598^{\bullet}$	0.463^{ns}
TON			$0.626^{\bullet}$	0.516^{ns}
LFOC				0.973***
LFON				
		Relationships between crop residue C input and soil organic C or N fractions		
Crop residue C input	0.717^{*}	$0.678^{\bullet}$	0.511^{ns}	0.359^{ns}

$^{\bullet}$, *, *** and ns refer to significant treatment effects in ANOVA at $P \leq 0.1$, $P \leq 0.05$, $P \leq 0.001$ and not significant, respectively.

Table 5. Linear regressions for relationships between crop residue C input from 1998 to 2009 growing seasons and organic C or N (TOC, TON, LFOC, LFON) stored in the 0 - 15 cm soil sampled in autumn 2009 at Scott, Saskatchewan, Canada.

Crop parameter (X)	Soil C or N parameter (Y)	^{z}Linear regression (Y = a + bX)	R^2
Crop residue C input	TOC	Y = 37.42 + 0.0005X	0.514^{*}
	TON	Y = 3.408 + 0.00004X	0.458^{*}
	LFOC	Y = 2640 + 0.047X	0.261^{ns}
	LFON	Y = 192.4 + 0.002X	0.131^{ns}

^{z}Y = Soil organic C or N fraction (TOC and TON as Mg C or N·ha^{-1}; and LFOC, LFON as kg C or N·ha^{-1}; a = Intercept on Y, origin of the line; b = Regression coefficient of Y on X, slope of line; X = Crop residue and/or swine manure C input (Mg·ha^{-1}); * and ns refer to significant treatment effects in ANOVA at $P \leq 0.05$ and not significant, respectively.

straw) are returned to soil in all crop phases. Similarly, in our study there was an increase in storage of TOC or TON in soil with increasing length of crop rotation from monoculture pea (1-year) to canola-wheat-pea-wheat rotation (4-year) but the differences were significant only in a few cases even after 12 years.

Previous research has shown that dynamic organic C or N fractions are more responsive to management practices than total organic C or N under annual crops [24-27]. Similarly, in our study the changes in LFOC and LFON were more pronounced than TOC and TON, although the increases in LFOC and LFON in soil due to crop rotations were not significant.

3.3. Residual Nitrate-N and Ammonium-N in Soil

The crop rotation duration × crop phase interaction effect was significant in most soil layers (**Table 6**). This was due to usually greater amounts of nitrate-N in pea phase than canola phase, particularly in the 1-yr rotation. The mean effect of crop rotation length/duration on soil nitrate-N was significant (at $P \leq 0.1$) only in the total 0 - 60 cm depth, but the amounts of nitrate-N were usually highest in all soil layers for monocultures, with the lowest nitrate-N in the 4-yr rotation. When ANOVA was conducted separately for canola and pea phases, there was no significant effect of crop rotation duration and/or frequency of canola in the rotation on soil nitrate-N. However, for pea phase there was a significant (at $P \leq 0.1$) effect of crop rotation duration and/or frequency of pea in the rotation on nitrate-N in the 15 - 30 and 30 - 60 cm soil layers. The amount of nitrate-N (averaged across crop rotation duration) was significantly greater in pea phase than canola phase in all soil layers. The generally higher soil nitrate-N in most layers with monoculture was probably due to relatively lower cumulative crop yield and total N uptake in seed in 1-yr rotation compared to longer rotations [11]. In another study in Saskatchewan, nitrate-N in soil was also higher in 6-yr continuous rotations with low crop diversity compared to rotations with high diversity of annual grain crops [21]. Earlier research has also suggested that residual N in soil can be decreased with efficient cropping systems [28]. Similarly, in our study soil nitrate-N in most soil layers was usually lowest in the 3-yr and 4-yr rotations with pea and 4-yr rotation with canola, suggesting the importance of 3- or 4-year rotations in reducing residual nitrate-N in the soil profile.

The mean effect of crop phase on ammonium-N in soil was not significant in any soil layer, but the crop rotation duration × crop phase interaction effect was significant in the 0 - 60 cm depth most likely due to greater amounts of ammonium-N in soil in canola phase than pea phase (**Table 6**). When ANOVA was conducted separately for canola and pea phases, there was no significant effect of crop rotation duration and/or frequency of pea in the rotation on soil ammonium-N. But, there was a significant effect of crop rotation duration and/or frequency of canola in the rotation on ammonium-N in the 15 - 30 and 30 - 60 cm soil layers. The tendency of greater amounts of ammonium-N (averaged across crop phases) in 1-yr or 2-yr rotation than 3-yr or 4-yr rotations in the 30 - 60 cm soil layer or 0 - 60 cm depth were most likely due to contribution through ammonification of N of recently added residue from crops with relatively longer taproots at deeper depth, particularly canola.

3.4. Amounts of N Uptake in Seed, Residual Nitrate-N in Soil and N Balance

The N balance over the 1998 to 2009 period for the 8 treatments (4 rotation durations × 2 crop phases) included amount of inorganic N applied as fertilizers, N added in seed at seeding time, estimated biologically fixed N (BFN) by pea when it was grown, amount of N recovered in seed over 12 years and mineral N (nitrate-N + ammonium-N) recovered in the 0 - 60 cm soil in autumn 2009 after 12 growing seasons, and the estimated amount of unaccounted N (**Table 7**). The estimated amount of N recovered in seed (which was removed from the land/field) plus the amount of mineral-N recovered in soil in various treatments ranged from 658 to 1103 $kg \cdot N \cdot ha^{-1}$. The corresponding values of N applied as inorganic fertilizers, plus N added in seed and BFN during the 12-year experimental period ranged between 556 and 1522 $kg \cdot N \cdot ha^{-1}$. The amounts of N that could not be accounted for ranged from 441 to −296 $kg \cdot N \cdot ha^{-1}$.

The amounts of unaccounted N were positive with monoculture pea, and the unaccounted N was much greater with pea than canola in the monoculture and 2-yr rotations, while the differences were small between the canola and pea phases in the 3-yr and 4-yr rotations. The positive unaccounted N reflects an excess of N that was applied to and/or fixed by pea compared to the N recovered in crop seed yield plus nitrate-N recovered in soil in the 1-yr rotation. The results of our study do not suggest any over-application of N, because little N was applied to pea and annual rates of applied N to other crops were moderate, and in canola phase the N balance was negative. It is possible that a portion of the fixed/applied N in 1-yr rotation in pea phase may have leached down below the 60 cm soil depth, as evidenced by greater amounts of nitrate-N recovered in the 30 - 60 cm layer in this study (**Table 6**), and also in the 60 - 90 cm layer in some treatments in the same experiment in autumn 2005 in our previous report [11]. Other researchers have reported an increase in the concentration of residual soil nitrate-N at

Table 6. Effect of broad-leaf crop phase and frequency over 12 years from 1998 to 2009 on nitrate-N and ammonium-N in soil in autumn 2009 at Scott, Saskatchewan, Canada.

Treatment		Nitrate-N ($kg \cdot N \cdot ha^{-1}$) in soil layers (cm)				Ammonium-N ($kg \cdot N \cdot ha^{-1}$) in soil layers (cm)			
Crop rotation (duration)	Crop phase	0 - 15	15 - 30	30 - 60	0 - 60	0 - 15	15 - 30	30 - 60	0 - 60
Monoculture (1-year)	Canola	12.7	0.0	0.0	12.7	6.9	6.8	14.0	27.7
Monoculture (1-year)	Pea	23.7	5.6	11.5	40.8	6.1	5.0	7.6	18.7
Canola-wheat (2-year)	Canola	12.7	1.0	0.5	14.2	4.8	4.2	9.1	18.1
Pea-wheat (2-year)	Pea	13.5	2.9	6.8	23.2	7.3	6.2	11.8	25.3
Pea-canola-wheat (3-year)	Canola	12.2	1.3	2.2	15.7	4.5	2.8	7.5	14.8
Pea-canola-wheat (3-year)	Pea	14.0	2.0	1.8	17.8	4.2	5.8	8.7	18.7
Canola-wheat-pea-wheat (4-year)	Canola	8.1	0.5	0.2	8.8	5.7	4.8	9.0	19.5
Canola-wheat-pea-wheat (4-year)	Pea	15.5	2.2	1.5	19.2	3.6	3.1	6.2	12.9
	$LSD_{0.05}$	ns	2.1	6.7	14.8	ns	ns	ns	10.4
	SEM (significance)	3.39^{ns}	0.72^{***}	2.26^{*}	5.04^{**}	1.20^{ns}	1.13^{ns}	2.00^{ns}	$3.54^{\bullet}$
Crop rotation duration									
1-year		18.2	2.8	5.6	26.6	6.5	5.9	10.8	23.2
2-year		13.1	2.0	3.7	18.8	6.1	5.2	10.5	21.8
3-year		13.1	1.7	2.0	16.8	4.4	4.3	8.1	16.8
4-year		11.8	1.4	0.9	14.1	4.7	4.0	7.6	16.3
$LSD_{0.05}$		ns	ns	ns	11.2	ns	ns	ns	ns
SEM (significance)		2.39^{ns}	0.61^{ns}	1.77^{ns}	$3.86^{\bullet}$	0.86^{ns}	0.87^{ns}	1.51^{ns}	2.72^{ns}
	Crop phase								
	Canola	11.4	0.7	0.7	12.8	5.5	4.7	9.9	20.1
	Pea	16.7	3.2	5.4	25.3	5.3	5.0	8.6	18.9
	$LSD_{0.05}$	4.9	1.3	3.7	8.0	ns	ns	ns	ns
	SEM (significance)	1.69^{*}	0.43^{***}	1.25^{*}	2.73^{**}	0.61^{ns}	0.61^{ns}	1.06^{ns}	1.93^{ns}
Additional statistical comparisons									
Significance of four crop rotation durations for canola phase only									
	$LSD_{0.05}$	ns	ns	ns	ns	ns	3.5	4.6	9.6
	SEM (significance)	3.50^{ns}	0.47^{ns}	1.00^{ns}	3.89^{ns}	1.09^{ns}	$1.09^{\bullet}$	1.45^{*}	$3.00^{\bullet}$
Significance of four crop rotation durations for pea phase only									
	$LSD_{0.05}$	ns	2.8	9.3	ns	ns	ns	ns	ns
	SEM (significance)	3.71^{ns}	$0.88^{\bullet}$	$2.92^{\bullet}$	6.33^{ns}	1.36^{ns}	1.14^{ns}	1.90^{ns}	3.39^{ns}

$^{\bullet}$, *and ns refer to significant treatment effects in ANOVA at $P \leq 0.1$, $P \leq 0.05$ and not significant, respectively.

Table 7. Balance sheets of broad-leaf crop phase and frequency over 12 years from 1998 to 2009 at Scott, Saskatchewan, Canada.

Parameters	Crop rotation duration							
	1-yr (monoculture)		2-yr		3-yr		4-yr	
	Canola	Pea	Canola	Pea	Canola	Pea	Canola	Pea
Mineral-N recovered in soil (0 - 60 cm) after 12 years in autumn 2009 (kg·N·ha^{-1})	40	60	32	49	31	37	28	32
N recovered in seed in 12 years (kg·N·ha^{-1})	618	1021	820	1054	954	954	972	972
N recovered in soil after 12 years + N recovered in seed in 12 years (kg·N·ha^{-1})	658	1081	852	1103	985	991	1000	1004
Inorganic N applied in fertilizers to non-legume crops in 12 years (kg·N·ha^{-1})	552	84	534	300	384	384	477	477
Organic N added in seed in 12 years (kg·N·ha^{-1})	4	78	22	59	41	41	41	41
Organic N fixed by pea in 12 years (kg·N·ha^{-1})	0	1460	0	730	487	487	365	365
Total N added in fertilizers + in seed + fixed by pea in 12 years (kg·N·ha^{-1})	556	1522	556	1089	912	912	883	883
N balance (N applied in fertilizers/seed/fixed – N recovered in seed) (kg·N·ha^{-1})	−62	501	−264	35	−42	−42	−89	−89
Unaccounted N (N applied in fertilizers/seed/fixed – N recovered in soil + seed) (kg·N·ha^{-1})	−102	441	−296	−14	−73	−79	−117	−121

high N fertilizer rates [28-30] and nitrate leaching in the 90 - 240 cm soil profile [21], suggesting the need for deep soil sampling below the 60 or 90 cm depth in future in this long-term study. Soil nitrate-N below the effective root zone of crops is susceptible to leaching, and the loss of nitrate-N through leaching can result in N contamination of groundwater, and thus represents a potential risk to groundwater quality and soil health [31-32]. Furthermore, a portion of the applied N in these treatments may have been immobilized in soil organic N, as evidenced by large amounts of soil N in LFON in this study (**Tables 3** and **4**). In addition, a small portion of the applied N may have been lost from the soil-plant system through denitrification (e.g., nitrous oxide and other N gases) due to wet soil conditions which temporarily exist in the present study in some years after occasional heavy rainfall during summer and/or autumn [33,34]. The negative amounts of N balance and unaccounted N, especially in canola phase, suggest that N became available to the crop through mineralization of organic matter in the growing season, and possibly soil may be gaining N from wet deposition and/or non-symbiotic N fixation but this needs further research to verify any contribution of N from precipitation and non-symbiotic N fixation.

4. CONCLUSION

The findings suggest that the quantity of organic C and N can be maximized by increasing duration of crop rotation and by including hybrid canola in the rotation.

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