Final Report

Identification

Program Name: Collaborative Research and Development Grant

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Project Title: Amino acid utilization and peripheral tissue metabolism in ruminants fed full-fat canola-based diets

File Number: CRDPJ 327170 - 2005

Co-Applicant: John JJ. McKinnon, Animal and Poultry Science, Saskatchewan

Co-Applicant: David DA. Christensen, Animal and Poultry Science, Saskatchewan

Supporting Organization: Canola Council of Canada
Agricore United
Saskatchewan Canola Development Comm.

Public Summary of Outcomes and Benefits to Canada

Lactating dairy cows or growing beef cattle require amino acids for milk production and growth. These amino acids are the "building blocks" of milk protein and skeletal muscle. Animas will meet this amino acid requirement from protein that is provided in the diet and from the digestion in the small intestine of microorganisms that grow in the fore-stomachs. In western Canada, there is considerable interest in feeding full-fat canola seed or oilseed meals because they provide high quality protein and can increase the energy density of diets fed to cattle because of their high residual oil content. In addition, full-fat canola seed has the potential to alter the fatty acid composition of milk and meat to contain less saturated and more unsaturated fatty acids. Unsaturated fatty acids have been reported to have beneficial effects on human cardiovascular health and to reduce the risk of cancer. Before the initiation of this project, relatively little research had been done to evaluate the effects of the protein fraction in full-fat canola seed as a source of protein for beef cattle and dairy cows, so the overall objective of this research was to determine the effects of full-fat canola seed, treated canola meal and canola by-products on milk production and composition, feed digestion and growth of microorganisms in the fore-stomachs and nutrient flow from the fore-stomachs to the small intestine.

Because dietary protein is an expensive component of beef and dairy cattle rations, the results from this study indicate that full-fat canola seed and canola presscake (a by-product from the biodiesel industry) are alternative protein supplements that can be included in cattle rations at a cheaper cost without compromising milk production or animal growth. This information is very useful to the livestock and feed industries as it can be used to fine-tune cattle rations to optimize the efficiency of utilization of nitrogen, thereby reducing the negative environmental consequences of intensive livestock operations like beef feedlots and dairy farms. Also, feeding full-fat canola seed to lactating dairy cows increased the milk fat contents of desirable omega-3 fatty acids, thus enhancing the nutritional properties of milk. The use of full-fat canola seed to enrich milk with unsaturated fatty acids with identified beneficial health consequences for humans represents a most significant opportunity for Saskatchewan canola farmers to add value to their canola crop. In addition, it also represents a significant opportunity for Saskatchewan dairy farmers to add value to milk, which should improve the health of western Canadians and expand global markets for both canola products and enriched milk and meat products. As the results of this project are in the public domain, they are available to the canola, livestock and feed industries for use in various ways (e.g., marketing strategies, specialized feed formulations etc.). The knowledge generated from this work has the potential to enhance the competitiveness of the canola and feed industries, and the dairy and beef sectors in the optimum utilization of canola-based proteins to replace animal proteins in ruminant diets. This will increase consumer acceptance of ruminant edible food products and also increase food safety. The information can also be used for marketing purposes. This will allow the development of alternative markets for full-fat canola seed and canola by-products.

This NSERC-CRD project has contributed to the training of one Post-Doctoral Fellow (PDF), one Ph.D. student and one M.Sc. student. The PDF and graduate students received training in advanced techniques that are used ruminant nutrition research, including isotope-based methods to assess nutrient utilization. Training also included the statistical analysis of data, critical interpretation of research data, and the preparation of scientific manuscripts for publication in peer-reviewed journals. This knowledge that the PDF and graduate students acquired is in great demand in academia, the animal nutrition research community, and in the livestock and feed industry. In addition, the PDF and graduate students also got exposure to the wider scientific community by presenting their research findings at numerous scientific conferences. The PDF is now employed as an Instructor in the Department of Animal and Poultry Science at the University of Manitoba. During his recruitment for that position, it was clear that Dr. Gozho's knowledge and skills that he had acquired during his post-doctoral training, and his involvement in a project that involved the feed and livestock industries, were major reasons why he was successful in the interview process. The Ph.D. student is in the last stages of her Ph.D. program and she intends to join the feed industry once she completes he studies. The M.Sc. student completed her M.Sc. studies in 2009 and is currently employed as a Dairy Cattle Specialist by the Government of Victoria Province (Australia). The contribution to the training of highly-qualified personnel on this project was high.

Progress Towards Objectives/Milestones

To what extent were the objectives of the grant achieved? Rate your answer on a scale from 1 to 7.

Not at all			Somewhat		To a great extent	
○ 1	○ 2	○ 3	○ 4	○ 5	● 6	○ 7

FINAL REPORT: REPORT ON PROGRESS

Project Title: Amino acid utilization and peripheral tissue metabolism in ruminants fed full-fat canola-based diets (CRDPJ 327170)

1. Objectives of the Research Project:

1.1.1. To determine the effects of full-fat canola seed on patterns of ruminal fermentation, ruminal microbial yields, ruminal protein degradability and biohydrogenation of canola lipids, duodenal amino acid (AA) and fatty acid (FA) profiles, peripheral tissue protein metabolism, and animal performance in dairy and beef cattle;

1.1.2. To determine the effects of moist heat treatment of full-fat canola seed on patterns of ruminal fermentation, ruminal microbial yields, ruminal protein degradability and biohydrogenation of canola lipids, duodenal AA and FA profiles, peripheral tissue protein metabolism, and animal performance; and

1.1.3. Using the information obtained under Objectives 1 and 2, determine those EAA that are deficient in duodenal protein for milk production or growth in ruminants fed canola seed-based diets and, subsequently, to determine the effects of strategic supplementation with those deficient EAA, either singly or in judicious combinations, on animal performance and peripheral tissue protein metabolism.

2. Progress Made Towards Each of These Objectives:

2.1. Experiment 1:

The objective was to determine the effects of full-fat canola seed on patterns of ruminal fermentation, ruminal microbial yields, ruminal protein degradability and biohydrogenation of canola lipids, duodenal AA and FA profiles, and performance in dairy cows (Objective 1.1.1.).

Eight Holstein cows (656.3 ± 27.7 kg BW; 79.8 ± 12.3 DIM) were used in a 4 x 4 Latin square design with 28-d periods and a 2 x 2 factorial arrangement of dietary treatments. Four cows in one Latin square were fitted with ruminal and "T-type" duodenal cannulae to allow sampling of ruminal contents and duodenal digesta, respectively. The duodenal cannulae were placed approximately 10 cm caudal to the pylorus. Each experimental period consisted of 14 d of dietary adaptation and 14 d of data collection. Experimental treatments were combinations of two sources of dietary supplemental fat (whole canola seed and whole flaxseed) and barley grain processing (dry-rolling or pelleting) as follows: a) dry-rolled barley + whole canola seed; b) dry-rolled barley + whole flaxseed; c) pelleted barley + whole canola seed; and d) pelleted barley + whole flaxseed. The diets were formulated to be isonitrogenous and isolipidic. Flax was included as a dietary treatment for comparative purposes only because its oil content and fatty acid profile differ markedly from that of canola. Feeding dry-rolled or pelleted barley was aimed at manipulating ruminal biohydrogenation of canola or flax lipids in the diet by altering ruminal pH. Experimental diets were fed twice daily at 0900 and 1600 h as total mixed rations (TMR) for ad libitum intake. The forage:concentrate ratio of the TMR was 50:50. The forage component of the TMR was a mixture of barley silage and chopped alfalfa hay. Experimental diets contained

approximately 5.3% total fat. Milk samples were collected on d 25, 26 and 27 from am and pm milkings with or without preservative, pooled daily based on milk yield. Pooled milk samples with preservative were analyzed for crude protein and fat. Pooled milk samples without preservative were analyzed for fatty acid composition. Ruminal pH was measured continuously using indwelling pH probes between d 25 and d 27. Nitrogen kinetics were measured for 4 d between d 15 and d 20 of each experimental period by infusing 384 mg/d of [^{15}N^{15}N]urea (98% APE) into the jugular vein for 4 d to achieve plateau [^{15}N^{15}N]urea enrichments in blood and urine with simultaneous total collection of urine and faeces. Digesta flow to the duodenum was measured using $YbCl_3$ as a solid phase marker and Cr-EDTA as a liquid phase marker. Microbial protein supply was measured using $^{15}NH_4SO_4$ as a marker. Sampling of whole ruminal contents and whole duodenal digesta began at 0700 h on day 27 and was conducted every 2 h until 0600 h on d 28, to represent the 24 h-feeding cycle. Ruminal fluid samples were used for determination of VFA and ammonia-N concentrations. Ruminal bacteria pellet was isolated from whole ruminal contents freeze-dried, and analyzed for N and ^{15}N-enrichment of total N and bacterial purine N. Duodenal digesta samples were analyzed for N (as NAN), amino acids, fatty acids and ^{15}N-enrichment of total NAN and purine N.

Supplementing diets with either canola seed or flaxseed did not affect DM intake. Dry matter intake and N intake were higher in cows fed dry-rolled barley compared to those fed pelleted barley. Actual and energy-corrected milk (ECM) were not affected by method of barley grain processing or source of supplemental fat. Milk fat content and milk fat yield were higher in cows fed dry-rolled barley compared to those fed pelleted barley. Short and medium chain milk fatty acid concentrations were higher for cows fed flax seed than canola. Although milk fat from cows fed flax seed-based diets contained more C18:3 and CLA (cis-9, trans-11), pelleting barley increased the milk content of CLA (cis-9, trans-11) and C18:3. Nitrogen retention was not affected by diet, but fecal N excretion was higher in cows fed dry-rolled barley than in those fed pelleted barley. Actual and energy corrected milk yield were not affected by diet. Source of supplemental fat did not affect urea-N kinetics. Urea-N production was higher, and urea-N entering the GIT tended to be higher, in cows fed dry-rolled barley compared to those fed pelleted barley. The amount of urea-N entry into the GIT that was returned to the ornithine cycle was higher in cows fed dry-rolled barley than in pelleted barley fed cows. Apparent ruminal digestibilities for DM, OM, starch, NDF, ADF, and OMTDR were not affected by both barley grain processing and oilseed. Apparent ruminal digestibilities for crude fat differed, possibly reflecting the differences in *de novo* synthesis as a result of dietary effects on ruminal pH. The total amount of fatty acids reaching the proximal duodenum was not affected by grain processing or source of oilseed, but cows fed pelleted barley rations had a much larger flow of unsaturated and less flow of saturated fatty acids. There was a larger amount of total fatty acids reaching the duodenum compared to the amount consumed. This difference can be attributed to the ability of rumen microbes to synthesize fatty acids. There was a larger amount of C18:0 flowing to the duodenum from all dietary treatments relative to the intakes recorded. This difference is due to the biohydrogenation of unsaturated fatty acids, especially C18:2n6c and C18:3n3, in the rumen. Cows fed diets containing flax had significantly larger duodenal flows of C18:3n3 and *trans*-10, *cis*-12 CLA compared to cows fed canola; however, *cis*-9, *trans*-11 CLA flows only tended to be higher in cows fed flaxseed compared to cows fed canola. The amounts of dietary DM apparently digested in the rumen tended to be higher in cows fed dry-rolled barley compared to those fed pelleted barley; as a consequence, amounts of OM apparently or truly digested in the rumen tended to be higher in cows fed dry-rolled barley compared to those fed pelleted barley.

When expressed in absolute amounts, apparent ruminal digestibility of NDF and ADF was higher in cows fed dry-rolled barley compared to those fed pelleted barley. Source of supplemental fat had no effect on apparent ruminal digestibility of DM, OM, NDF and ADF. Duodenal flows of DM, OM, NDF and ADF were unaffected by diet. Method of barley grain processing did not affect duodenal flow of total N. Duodenal flow of ammonia N tended to be higher in cows fed pelleted barley compared to those fed dry-rolled barley. Duodenal flow of non-ammonia non-microbial N, expressed as absolute amounts or as a proportion of NAN or N intake, were higher in cows fed flax compared to those fed canola; however, method of barley grain processing did not affect duodenal NANMN flow. Duodenal flow of microbial NAN and microbial efficiency were not affected by method of barley grain processing or source of supplemental fat.

2.2. Experiment 2:

The objective of this experiment was to determine the effects of heated canola meal and dietary CP level on patterns of ruminal fermentation, ruminal microbial yields, duodenal nutrient flows, and whole-animal nitrogen kinetics in beef heifers (Objectives 1.1.1 and 1.1.2).

Four beef heifers (437 kg BW) were fitted with ruminal and "T-type" duodenal cannulae to allow sampling of ruminal contents and duodenal digesta, respectively. The duodenal cannulae were placed approximately 10 cm caudal to the pylorus. A 4 x 4 Latin square experimental design was used in this experiment with a factorial arrangement of dietary treatments and with 21-d experimental periods. Each 21-d experimental period consisted of 14 d of dietary adaptation, followed by a 7-d period when measurements were taken. Dietary treatment comparisons were source of canola meal (heated *vs* unheated) and dietary crude protein level (10 *vs* 13% CP). To assess dietary impact on whole-animal N kinetics, the total balance technique described for dairy cows under Experiment 1 was used. Briefly, on d 14, experimental animals had catheters inserted in the right and left jugular veins to allow for simultaneous [$^{15}N^{15}N$]-urea infusion and blood sampling. Indwelling bladder catheters were also inserted to allow urine collection. Beginning on d 15, 220 mg/d of [$^{15}N^{15}N$]-urea (98% APE) was infused into one jugular vein for 4 d to achieve plateau [$^{15}N^{15}N$]-urea enrichments in blood and urine. During this period (d 15 to d 20), total collection of urine and faeces were conducted as already described above. To determine rates of digesta flow to the duodenum, 2.2 g/d of $YbCl_3$ were used intraruminally as digesta markers for the particle phase using the protocol described above under Experiment 1.

Increasing dietary CP increased N intake and urinary N excretion, but had no effect on fecal N excretion (Table 3). Retained N tended to increase as dietary CP level increased. Dietary RDP level had no effect on N balance. Urea-N entry rate increased with dietary CP level, but the amounts of endogenous urea-N production partitioned to the GIT and urea-N utilized for anabolic purposes were unaffected. Urea-N returned to the ornithine cycle tended to increase as dietary CP increased, whereas urea-N transferred to feces tended to decrease as dietary CP increased. Urea-N fluxes were unaffected by dietary RDP level and interactions between dietary levels of CP and RDP did not affect N balance or urea-N kinetics. Fractional transfers of urea-N were largely unaffected by diet; however, the proportion of urea-N that was recycled to the GIT that was voided in feces decreased as dietary CP increased. Microbial N supply was unaffected by diet.

2.3. Experiment 3:

The objective of this experiment was to determine the effects of feeding canola by-products on patterns of ruminal fermentation, ruminal microbial yields, and duodenal amino acid flows in beef heifers (Objectives 1.1.1, 1.1.2 and 1.1.3). The canola by-products used were regular canola meal, canola meal from biodiesel production (which contains 11 – 15% oil), and heated canola meal to investigate the effects of oil content on ruminal fermentation.

Four beef (initial BW = 451 ± 26 kg) that were fitted with 10-cm ruminal and simple T-type duodenal cannulae were used in a 4 x 4 Latin square design with 21-d periods. Duodenal cannulae were inserted proximal to the common bile and pancreatic duct, approximately 10 cm distal to the pylorus. Each experimental period consisted of 14 d of dietary adaptation and 7 d of data collection. The main ingredients of the experimental diets were barley grain, barley silage, and pelleted concentrate. The protein in the concentrate was supplied by regular canola meal (RCM), canola presscake (CPC) from biodiesel oil extraction, or a proprietary high RUP canola meal that is marketed as Alberta Gold (RUCM). According to the manufacturer, Alberta Gold is produced using a patented manufacturing process that results in a meal with 55% of CP as RUP (assuming a ruminal outflow rate of 4%). This would be greater than the RUP content of RCM as indicated by previous work which showed effective ruminal degradabilities of RCM ranging from 62.5 to 74%. In Canada, RCM is produced by the pre-press solvent extraction process. Generally, this process involves seed conditioning at 75 to 78ºC, flaking and cooking at 90ºC for 20 to 30 min, followed by solvent extraction in hexane. The resultant presscake is then desolventized-toasted at 100 to 110ºC for approximately 60 min, dried, and then cooled. Biodiesel oil extraction uses a cold press system without the use of a solvent or heat. Seeds are pressed once, subjected to extrusion under pressure, and then pressed a second time to remove remaining oil, and the presscake is dried in a cooler. The protein sources for the experimental diets (DM basis) were: 1) 8.78% RCM, 2) 9.25% RCM plus 1.80% canola oil (RCMO), 3) 11.10% CPC, or 4) 8.14% RUCM plus 1.32% canola oil (RUCMO). Canola oil was added to the RCMO and RUCMO treatments in order to simulate the higher residual oil content of CPC. Diets were fed as total mixed rations that were offered in 2 equal portions at 0800 and 1600 h. The diets were fed at approximately 1.5 x NE_m requirement with an expected ADG of 0.84 kg/d. The respective DM, CP (DM basis), and ether extract (DM basis) were: 87.8, 39.2, and 3.34% for RCM; 91.7, 33.0, and 15.2% for CPC; and 92.9, 42.8, and 4.68% for RUCM.

Digesta flow and nutrient digestibility were determined using $YbCl_3$ and Cr-EDTA as markers for the solid and liquid fractions as described above (Experiment 2). Microbial protein synthesis was measured using ^{15}N as the ruminal microbial marker. Whole ruminal contents were taken from each animal to determine background ^{15}N natural abundance (^{15}NB) prior to marker infusion. Marker solutions were then continuously infused into the rumen starting on d 15. On the first day of infusion, half the daily dose of each marker was placed into the rumen via ruminal cannula as a priming dose. From d 16 to d 19, spot urine samples were collected from each heifer before morning and afternoon feeding and analyzed for total N. Diluted samples from spot urine samples (2 mL urine in 8 mL of distilled water) were also composited for each cow and period and analyzed for urea-N and purine derivatives. Starting on d 20, a 1-L composite sample of whole ruminal digesta (taken from the cranial ventral, caudal ventral, central, and cranial dorsal rumen), 300 mL of duodenal digesta, and grab fecal samples were taken at 0800, 1100, 1400, 1700, 2000, and 2300 h and at 0200 and 0500 h on d 21, to represent a 24-h feeding cycle. Ruminal fluid samples were analyzed for ruminal pH, and for VFA and NH_3-N concentrations.

Ruminal bacteria were also obtained via differential centrifugation and bacteria pellets were pooled by period for each cow, freeze-dried, and analyzed for N and ^{15}N-enrichment of total N and bacterial purine N. Duodenal digesta samples were analyzed for N (as NAN), amino acids, fatty acids and ^{15}N-enrichment of total NAN and purine N.

There was no dietary effect on ruminal pH and this was expected because diets were formulated to provide rumen fermentable carbohydrates from the same sources with the only different feed ingredient among the 4 treatments being the source of protein. Compared with feeding the RCM diet, ruminal NH_3-N concentration were lower by 24%, 20%, and 33% in heifers fed the RCMO, CPC, and RUCMO diets, respectively. Despite the observed treatment effect on ruminal NH_3-N concentrations, we did not detect treatment effects on the supply of ruminally degraded protein or blood and urinary urea-N. Compared with the RCM diet, heifers fed the RCMO diet had higher ruminal concentrations of acetate, propionate, valerate, and total VFA. Similarly, heifers fed the CPC diet had higher concentrations of propionate compared with the RCM diet. On the other hand, feeding the RUCMO diet resulted in lower ruminal concentrations of acetate compared with the RCM diet. Because DMI was restricted, intakes of OM, NDF, and ADF were not affected by dietary treatment. Differences among treatments in the ruminal digestibility of OM, NDF, and ADF were not detected**.** Digestibility of OM, NDF, and ADF in the total tract was affected by dietary treatment. Compared with the RCM diet, feeding the RCMO diet decreased the digestibility of ADF and NDF in the total tract. Additionally, feeding the RUCMO diet decreased the digestibility of OM, NDF, and ADF compared with the RCM diet. These data suggest that adding canola oil to the RCMO and RUCMO diets affected fiber digestion in the hindgut. As observed for other nutrients, N intake and flow of N fractions to the duodenum were similar across diets. Averaged across diets, duodenal flow of total N, NAN, NANMN, and microbial N was 135, 133, 25, and 108 g/d, respectively. Similarly, the flow of these N fractions as a percentage of N intake was unaffected by dietary treatment. Microbial efficiency also was not affected by dietary treatment and averaged 34.6 g of microbial N/kg of OM truly digested in the rumen. Pre-experiment diet evaluation using the NRC (1996, Level 2) model revealed that, on average, these diets were expected to provide 636 and 170 g/d of MP from bacteria and RUP, respectively. These values were similar to 673 and 158 g/d calculated from microbial N and NANMN in this study. Surprisingly, the RDP fractions were similar among the 4 treatments. Lower rumen degradable fractions were expected for heifers fed the RUCMO and RCM diets compared with the CPC diet. This is because RCM and RUCM protein supplements are exposed to heat in the desolventizer-toaster step during oil extraction. Also, according to the manufacturer, RUCM contains 55% of CP as RUP. The data from in vivo measurements of RDP in the current experiment revealed no dietary treatment effect and averaged across treatments was 891 g/d or 10.3% of DMI. Flows of individual AA were not affected by diet. These results were expected considering that the contribution of RUP to AA reaching the duodenum could have been minimal because less than 20% of dietary protein escaped rumen degradation.

2.4. Experiment 4:

The objectives of this study were to determine the interactive effects of level of dietary CP and rumen-degradable protein (RDP) level on microbial protein synthesis, ruminal fermentation characteristics, nitrogen balance and milk production response in lactating dairy cows fed regular canola meal or heated canola meal (Objectives 1.1.1 and 1.1.2).

Eight multiparous (711 ± 21 kg BW; 91 ± 17 DIM), lactating Holstein cows were used in a replicated 4 × 4 Latin square design with a 2 × 2 factorial arrangement of dietary treatments and 30-d experimental periods. Four cows in one square were fitted with rumen cannulas. Dietary treatments were 2 levels of CP (14.1 vs. 16.5%) and 2 levels of RDP (63 vs. 69% of CP). Rumen-degradable protein levels were manipulated by varying inclusion levels of canola meal and heated canola meal. After 21 d of diet adaptation, digesta flow and nutrient digestibility were determined using $YbCl_3$ and Cr-EDTA as markers for the solid and liquid fractions as described above (Experiment 2). Microbial protein synthesis was measured using ^{15}N as the ruminal microbial marker. Briefly, whole ruminal contents were taken from each animal to determine background ^{15}N natural abundance (^{15}NB) prior to marker infusion. Marker solutions were then continuously infused into the rumen. On the first day of infusion, half the daily dose of each marker was placed into the rumen via ruminal cannula as a priming dose. A 1-L composite sample of whole ruminal digesta (taken from the cranial ventral, caudal ventral, central, and cranial dorsal rumen), 300 mL of duodenal digesta, and grab fecal samples were taken every 6 hr over 2 d to represent a 24-h feeding cycle. Ruminal fluid samples were analyzed for ruminal pH, and for VFA and NH_3-N concentrations. Ruminal bacteria were also obtained via differential centrifugation and bacteria pellets were pooled by period for each cow, freeze-dried, and analyzed for N and ^{15}N-enrichment of total N and bacterial purine N. Duodenal digesta samples were analyzed for N (as NAN), amino acids, fatty acids and ^{15}N-enrichment of total NAN and purine N. Milk samples were collected on 3 consecutive days (d 27 to 29) and analyzed for CP and fat.

Nitrogen intake, and both urinary N and urinary urea-N output were greater for the high CP diets as compared to cows fed low CP diets. Ruminal ammonia-N concentration tended to be greater in cows fed high CP than those fed low CP diets (20.3 vs. 17.4 mg/dL), and feeding heated canola meal to reduce dietary RDP resulted in a lower ruminal ammonia-N concentration compared to feeding canola meal (21.5 vs. 16.2 mg/dL). However, nitrogen balance, milk yield, milk composition and microbial N supply were unaffected by dietary treatment. Lowering the level of dietary CP had no effect on production parameters and reduced the urinary excretion of nitrogen, therefore increasing the efficiency of dietary nitrogen utilization and reducing the impact of environmental nitrogen pollution.

3. Deviations from the Original Objectives:

None, except that due to technical problems that were encountered in the use of duodenally-cannulated animals to obtain samples of duodenal digesta, Experiment 4 was conducted with ruminally-cannulated animals, and the omasal sampling technique was used to obtain samples of digesta exiting the rumen. In initial experiments, we used duodenal cannulas to sample digesta arriving at the proximal duodenum; however, problems were encountered due to the loss of duodenal cannulas from some experimental animals, and intestinal blockage and reduced intestinal motility with other experimental animals. For these reasons, the care of animals with duodenal cannulas was deemed too difficult and subsequent experiments were conducted with ruminally-cannulated animals, and the omasal sampling technique was used to obtain samples of digesta exiting the rumen. Care of ruminally-cannulated animals was far less demanding, and the

use of the omasal sampling technique did not compromise the stated experimental objectives in any way.

4. Scientific Significance of the Results Achieved:

The results from these experiments indicate that, even though feeding differently produced canola protein supplements to ruminants had no effect on ruminal digestion or nutrient flows to the duodenum per se, changes in ruminal fermentation patterns consistent with fat supplementation were observed. Compared with feeding regular canola meal, feeding the high RUP canola meal (heat-treated) or the high RUP canola meal with added oil reduced the digestibility of ADF, NDF, and OM in the total tract, suggesting that the high oil content might have impaired ruminal function to some degree. There were decreases in ruminal NH_3-N and changes in the concentrations of ruminal VFA associated with adding canola oil or having higher residual oil in the protein supplement. Notwithstanding these changes, lipid content in the meal did not have an impact on microbial protein synthesis or N fractions reaching the duodenum. These results suggest that high RUP canola meal can be fed to ruminants, with or without added oil, without any adverse effects on ruminal microbial protein synthesis, indicating that post-ruminal metabolizable protein supply will not be compromised. Also, these results show that rumen degradability of protein from canola meal types is very high, which would ensure an adequate supply of ruminally-degradable protein for microbial growth, thus potentially maximizing microbial protein supply at the small intestine. Overall, these results suggest that, apart from minor changes in ruminal fermentation as a result of the higher residual oil compared with regular canola meal, canola presscake is a good protein source that can substitute for other canola protein supplements. Because of the high residual oil in canola presscake that is high in unsaturated fatty acids, an additional advantage of feeding canola presscake will be the enhancement of ruminant products with desirable fatty acids. Results also indicated that milk fat contents of desirable omega-3 fatty acids (C18:3) was enhanced in cows fed canola compared to those fed flax, and that milk fat contents of C18:3 and *cis*-9, *trans*-11 C18:2 (CLA) were higher in cows fed pelleted barley compared to those fed dry-rolled barley with flax as the source of oilseed, but not with canola. These important interactions need to be taken into account when feeding full-fat canola seed to alter milk fatty acid composition in dairy cows as the method of barley grain processing appears to have important effects on biohydrogenation pathways of unsaturated fatty acids in the rumen.

Problems Encountered

Identify the problems encountered during the research project. (Select all that apply.)

- [x] Technical or scientific problems
- [ ] Problems with direction of research or findings
- [ ] Equipment and facilities
- [ ] Staffing issues (e.g., availability of students, staff leaving project)
- [ ] Funding problems
- [ ] Partners withdrew from project
- [ ] Partners interaction issues
- [ ] No problems were encountered
- [ ] Other (specify)

Problems Encountered

If problems were identified, briefly describe them and the steps taken to resolve each one.

Due to technical problems that were encountered in the use of duodenally-cannulated animals to obtain samples of duodenal digesta, Experiment 4 was conducted with ruminally-cannulated animals, and the omasal sampling technique was used to obtain samples of digesta exiting the rumen. In initial experiments, we used duodenally-cannulated animals to sample digesta arriving at the proximal duodenum; however, problems were encountered due to the loss of duodenal cannulas from some experimental animals, and intestinal blockage and reduced intestinal motility with others. For these reasons, the care of animals with duodenal cannulas was deemed too difficult and subsequent experiments were conducted with ruminally-cannulated animals, and the omasal sampling technique was used to obtain samples of digesta exiting the rumen. Care of ruminally-cannulated animals was far less demanding, and the use of the omasal sampling technique did not compromise the stated experimental objectives in any way.

Research Team

Entry 1 of 7

Consent obtained: ● Yes ○ No

Name: Dr. T. Mutsvangwa

Role: Applicant

If role is "Other", specify:

Contribution

Dr. Mutsvangwa was the Principal Investigator and he had been responsible for the overall day-to-day management of the research project at the University of Saskatchewan. Dr. Mutsvangwa was the Graduate Supervisor for Ms. M. Hobin (M.Sc. student) and, together with Dr. J. J. McKinnon, he also co-supervised Ms. Kate Baker (nee Davies; a Ph.D. graduate student) and Dr. Gozho (a Post-Doctoral Fellow; PDF). In conjuction with Ms. Hobin and Ms. Baker, Dr. Gozho was responsible for the day-to-day managemnet of all animal experiments related to this project.

Entry 2 of 7

Consent obtained: ● Yes ○ No

Name: Dr. John J. McKinnon

Role: Co-Applicant

If role is "Other", specify:

Contribution

Dr. McKinnon contributed heavily to the development of most experimental protocols of experiments that were conducted under this project. Together with Dr. Mutsvangwa, he co-supervised Dr. Gozho and Ms. K. Baker (nee Davies).

Entry 3 of 7

Consent obtained: ● Yes ○ No

Name: Mr. Vern J. Racz

Role: Co-Applicant

If role is "Other", specify:

Contribution

Mr. Racz retired from his position as Executive Director of the Prairie Feed Resource Centre in June 2006. Prior to his retirement, he was involved in the development of experimental protocols.

Research Team

Entry 4 of 7

Consent obtained: ● Yes ○ No

Name: Dr. David A. Christensen

Role: Co-Applicant

If role is "Other", specify:

Contribution

Dr. Christensen contributed to the development of most experimental protocols for experiments that were conducted under this project.

Entry 5 of 7

Consent obtained: ● Yes ○ No

Name: Ms. M. Hobin

Role: Graduate Student

If role is "Other", specify:

Contribution

Ms. Hobin was a M.Sc. graduate student. Her main focus was to determine the impact of canola-containing diets on ruminal dynamics, microbial protein production, whole-body nitrogen dynamics, and milk production and composition in lactating dairy cows (Experiment 1).

Entry 6 of 7

Consent obtained: ● Yes ○ No

Name: Ms. Kate Baker (nee Davies)

Role: Graduate Student

If role is "Other", specify:

Contribution

Ms. Kate Baker (nee Davies) is a Ph.D. graduate student. Her main focus was to determine the impact of canola-containing diets on ruminal dynamics, microbial protein production, whole-body nitrogen dynamics, and milk production and composition in lactating dairy cows and beef cattle (Experiments 2 and 4).

Research Team

Entry 7 of 7

Consent obtained: ● Yes ○ No

Name: Dr. George G. Gozho

Role: Postdoctoral Fellow

If role is "Other", specify:

Contribution

In conjunction with the graduate students, Dr. Gozho was responsible for the day-to-day management of the experiments, including protocol preparation, diet formulations, management and care of experimental animals, organization of sample collection, laboratory analyses of samples, data entry and analysis, and preparation of reports and scientific manuscripts. He provided valuable technical expertise in bladder and jugular catheterization of experimental animals, and protocols for jugular infusions of isotopic tracers. He worked on the project from February 2006 to August 2009.

Training of Highly Qualified Personnel (HQP)

What types of interactions did the HQP have with the partners during the project? (Select all that apply.)

- [x] HQP presented research results to the partners
- [ ] HQP discussed the project directly with partners to obtain input
- [ ] Partners jointly supervised thesis projects of HQP
- [ ] HQP worked regularly in the partner's facilities
- [ ] HQP did not interact with the partners
- [ ] Other (specify)

Entry 1 of 3

Name:	Dr. George N. Gozho
Type:	Postdoctoral Fellows
If type is "Other", specify:	
Start Date yyyy/mm:	2006/02
End Date yyyy/mm:	2009/08
Percentage (%) of time this individual spent on this project:	100
Percentage (%) of salary from this grant (NSERC and industry contribution):	100
Total person-months:	42
To the best of your knowledge trainee is:	Employed in Academia / Faculty
If "Employed by Other", specify:	

Entry 2 of 3

Name:	Ms. Morgan R. Hobin
Type:	Master's Student
If type is "Other", specify:	
Start Date yyyy/mm:	2005/09
End Date yyyy/mm:	2009/07
Percentage (%) of time this individual spent on this project:	80
Percentage (%) of salary from this grant (NSERC and industry contribution):	0
Total person-months:	37
To the best of your knowledge trainee is:	Employed by Government
If "Employed by Other", specify:	

Training of Highly Qualified Personnel (HQP)

Entry 3 of 3

Name:	Ms. Kate Baker (nee Davies)
Type:	Doctoral Student
If type is "Other", specify:	
Start Date yyyy/mm:	2005/09
End Date yyyy/mm:	2010/12
Percentage (%) of time this individual spent on this project:	50
Percentage (%) of salary from this grant (NSERC and industry contribution):	0
Total person-months:	32
To the best of your knowledge trainee is:	Continuing Academic Training
If "Employed by Other", specify:	

Dissemination of Research Results

Refereed Journal Articles Submitted :	0
Refereed Journal Articles Accepted or Published:	2
Conference Presentations/ Posters:	8
Other (Technical Reports, Non-Refereed Articles, etc.):	0
How many of the publications, conference presentations, etc. identified above were co-authored with a non-academic partner?	0

FINAL REPORT: DISSEMINATION OF RESEARCH RESULTS

Project Title: Amino acid utilization and peripheral tissue metabolism in ruminants fed full-fat canola-based diets (CRDPJ 327170)

1. Refereed Journal Articles, Submitted:

None.

2. Referred Journal Articles, Accepted or Published:

a) G. N. Gozho, J. J. Mckinnon, D. A. Christensen, V. Racz, and T. Mutsvangwa, 2009. Effects of Type of Canola Protein Supplement on Ruminal Fermentation and Nutrient Flow to the Duodenum in Beef Heifers. Journal of Animal Science 87: 3363-3371.

b) G. N. Gozho, M. R. Hobin, and T. Mutsvangwa, 2008. Interactions between Barley Grain Processing and Source of Supplemental Dietary Fat on Nitrogen Metabolism and Urea-Nitrogen Recycling in Dairy Cows. Journal of Dairy Science 91: 247-259.

3. Conference Presentations and Posters:

a) K. L. Davies, J. J. McKinnon, D. A. Christensen, and T. Mutsvangwa. 2010. Effects of dietary crude protein (CP) and rumen-degradable protein (RDP) level on microbial protein synthesis, ruminal fermentation characteristics, nitrogen balance, performance in lactating Holstein dairy cows. Greenhouse Gases and Animal Agriculture Conference held at Banff, Canada, October 3-8, 2010.

b) G. N. Gozho, J. J. McKinnon, D. A. Christensen, V. Racz, and T. Mutsvangwa, 2009. Effects of Source of Canola Protein on Ruminal Fermentation and Nutrient Flow in Beef Heifers. Canadian Journal of Animal Science 89: 144.

c) G. N. Gozho, M. R. Hobin, and T. Mutsvangwa, 2008. Effects of Barley Grain Processing and Source of Supplemental Dietary Fat on Nutrient Digestion and Microbial Protein Synthesis in Dairy Cows. Journal of Dairy Science 91(E-Suppl. 1): 69.

d) T. Mutsvangwa, G. N. Gozho, and D. Kiran, 2008. Effects of Degree of Unsaturation of Supplemental Dietary Fat on Ruminal Fermentation, Nitrogen Metabolism, and Urea Nitrogen Recycling in Dairy Cows. Journal of Dairy Science 91(E-Suppl. 1): 78.

e) K. L. Baker, T. Mutsvangwa, J. J. McKinnon, G. Gozho, and T. A. McAllister, 2008. Effects of Dietary Crude Protein and Ruminally-Degradable Protein on Urea Recycling and Microbial Protein Production in Beef Heifers. Canadian Journal of Animal Science 88:179.

f) M. R. Hobin, G. N. Gozho, and T. Mutsvangwa, 2007. Effects of Barley Grain Processing and Source of oilseed on Milk Fatty Acid Composition in Dairy Cows. Canadian Journal of Animal Science 88:149.

g) K. L. Baker, T. Mutsvangwa, J. J. McKinnon, G. Gozho, and T. A. McAllister. 2007. Effects of Dietary Crude Protein and Ruminally-Degradable Protein on Urea Recycling and Microbial Protein Production in Beef Heifers. Proceedings of the 2nd International Symposium on Energy and Protein Metabolism. EAAP Publication No. 124: 433-434.

h) G. N. Gozho, M. Hobin, and T. Mutsvangwa, 2007. Interactions between Oilseed Supplementation and Barley Grain Processing on Urea-Nitrogen Recycling and Nitrogen Metabolism in Dairy Cows. Journal of Dairy Science 90(Suppl. 1): 101.

4. Other (Technical Reports, Non-Refereed Articles etc.)

None.

Intellectual Property Protection

Filing of patent applications:	Not applicable
Registration of copyright for computer software or databases:	Not applicable
Registration of copyright for educational materials:	Not applicable
Registration of industrial designs:	Not applicable
Filing for protection of trademarks:	Not applicable
Registration of integrated circuit topographies:	Not applicable
Filing of applications for plant breeders' rights:	Not applicable
Execution of non-disclosure or confidentiality agreements:	Not applicable
Other (specify):	Not applicable

Collaboration with the Partners

How was this research project initiated?

- [x] The university researcher approached the partners
- [x] The partners approached the university researcher
- [ ] The government partner approached the university
- [ ] There was a previous collaboration with the partners
- [ ] This is a new collaboration
- [ ] Other (specify)

Did this project arise from a grant funded by the NSERC Strategic Workshops Program? ○ Yes ● No

Did this project arise from a grant funded by the Interaction and/or Engage Program? ○ Yes ● No

Collaboration with the Partners

Briefly describe the process.

The Department of Animal and Poultry Science (Drs. D. A. Christensen and J. J. McKinnon) at the University of Saskatchewan had been involved in various feed evaluation projects, including some projects investigating canola by-products. Based on this prior research, Dr. Dave Hickling initiated discussions with this group to conduct further research focused on the impact of full-fat canola seed and canola by-products on ruminal fermentation and post-ruminal flow of nutrients, particularly amino acids and fatty acids, in lactating dairy cows and beef cattle. Dr. T. Mutsvangwa had recently joined the University of Saskatchewan and was involved in the discussions as he had technical expertise in the metabolic approaches that would be implemented in the project.

Collaboration with the Partners

To what extent were the partners involved in the project? Rate your answer on a scale from 1 to 7.

Not at all			Somewhat			To a great extent
○ 1	○ 2	○ 3	○ 4	● 5	○ 6	○ 7

In what way were the partners directly involved in the project? (Select all that apply.)

- [x] Partners were available for consultation
- [ ] Partners provided facilities
- [ ] Partners provided training
- [ ] Partners co-supervised students' theses
- [ ] Partners received training from university personnel
- [ ] Personnel from the partner organization received training from the university
- [x] Partners discussed the project regularly with the university team

  Average number of meetings per year: 1
- [ ] Partners were involved in the research
- [ ] Other (specify)

Collaboration with the Partners

Describe the partners' involvement and comment on the collaboration.

The involvement of the industry partners in the day-to-day implementation of this project was minimal. Dr. D. Hickling was actively involved in the development of the NSERC-CRD research proposal. Mr. J. R. Dean (formerly Manager of Market Development, Agricore United) and Mr. R. Button (formerly Executive Director, Saskatchewan Canola Development Commission) were involved in the initail planning stages and in initial development of research protocols, but both retired during the first year of the project and there was little involvement thereafter from Agricore United and the Saskatchewan Canola Development Commission.

Future Plans

What links are you maintaining with the partners? (Select all that apply.)

- ☐ Collaborating with the partners on the same research
- ☑ Collaborating with the partners on other research
- ☐ Collaborating with other partners on the same research
- ☐ Continuing the research without partners
- ☐ No contact with the partners currently and none planned
- ☐ No contact with the partners currently but future collaboration planned

Future Plans

Describe any follow-up or related work that will be undertaken as a result of this project, who will be involved in this work (including partners) and how it will be funded.

Dr. T. Mutsvangwa is now working with Dr. Dave Hickling of the Canola Council of Canada on a Canola Meal Cluster Project that involves scientists from the University of Saskatchewan, Agri-Food and Agriculture Canada and several Universities in the U.S.A. The overall objective of this collaborative research project is to elucidate the mechanism(s) by which canola meal enhances milk production in dairy cattle and to determine and measure the factors that influence that response. Dr. Mutsvangwa's involvement was made possible because of his interaction with Dr. Hickling on the NSERC-CRD project.

Knowledge and Technology Transfer

Research results transferred to the partners

- [x] Through informal discussions
- [x] Through reports provided to the partners
- [ ] As a result of the partners participating in the research
- [x] Through formal publications
- [ ] Through patents
- [ ] Through licencing arrangements
- [ ] The research results have not been transferred to the partner
- [ ] Other (specify)

Research results being used and/or will be used by the partners

As a stimulus for future R&D:	Potential to be used
To enhance the skills and knowledge of personnel in the partner's organization:	Potential to be used
To improve an existing product:	Potential to be used
To improve an existing process:	Potential to be used
To improve an existing service:	No potential to be used
To develop a new product:	Potential to be used
To develop a new process:	Potential to be used
To develop a new service:	No potential to be used
To contribute to a policy, regulation or standard:	Potential to be used

Other (specify):

Knowledge and Technology Transfer

Briefly describe these outcomes

Knowledge and Technology Transfer

Describe any environmental or social benefit that resulted or could result in the future from this research

Impact on Researcher

Impact the project had on your teaching

- [ ] Creation of new courses
- [x] New content for existing courses
- [x] Use of real world examples in courses
- [ ] Guest lectures from partners
- [ ] New equipment/material
- [ ] Project has had no impact on my teaching
- [ ] Other (specify)

Impact the project had on your research

- [x] Influenced the direction to more industrially relevant topics
- [x] Opened up new opportunities for research beyond the original objectives
- [ ] The project has had no impact on my research
- [ ] Other (specify)

Contributions from Other Sources

Partners	Total Cash		Total In-Kind	
Company Name	**Committed**	**Received**	**Committed**	**Received**
Canola Council of Canada	60,000	60,000	0	0
	Have you had previous research collaborations with this partner? No			
Saskatchewan Canola Development Commission	80,000	80,000	0	0
	Have you had previous research collaborations with this partner? No			
Agricore United	20,000	10,000	0	0
	Have you had previous research collaborations with this partner? No			
	0	0	0	0
	Have you had previous research collaborations with this partner?			
	0	0	0	0
	Have you had previous research collaborations with this partner?			
	0	0	0	0
	Have you had previous research collaborations with this partner?			
	0	0	0	0
	Have you had previous research collaborations with this partner?			

Contributions from Other Sources

Other Sources	Total Cash		Total In-Kind	
		Received		Received
		0		0
		0		0
		0		0
		0		0
		0		0
		0		0
		0		0
Total (partners and other sources)	**$160,000**	**$150,000**	**$0**	**$0**

Financial Information

Consolidated balance remaining at the end of the project:

Budget Items	Total Budget	Total Actual Expenditure	Percent Variation
1) Salaries and benefits			
PhD students	81,360	0	-999
Master's students	37,290	6,120	-84
Undergraduate students	0	14,526	999
Postdoctoral fellows	0	124,333	999
Technical/professional assistants	60,000	0	-999
	0	0	0
2) Equipment or facility			
Purchase or rental	0	0	0
Operation and maintenance costs	0	0	0
User fees	0	0	0
	0	0	0
3) Materials and supplies			
	90,474	103,777	15
Professional/tec services	5,300	25,143	374
	0	0	0
4) Travel			
Conferences	16,420	15,277	-7
Field work	0	0	0
Project related travel	0	0	0
	0	0	0
5) Dissemination			
Publication costs	10,850	1,973	-82
	0	0	0
6) Technology transfer activities			
Field trials	0	0	0
Prototypes	0	0	0
	0	0	0
7) Others (specify)			
Indirect costs + overhead	9,152	10,593	16
	0	0	0
Total	310,846	301,742	-3

Financial Information

Explanation for the variation of each budget item

SALARIES and BENEFITS: At the beginning of the project, there was a delay in identifying a suitably-qualified graduate student so the project team decided to hire Dr. G. Gozho as a post-doctoral fellow (PDF) in order to avoid undue delays in initiation of the project. This was brought to the attention of NSERC in the first progress report that was submitted to NSERC in November 2006. The experimental protocols in this project (cannulation of the digestive tract, jugular catherization, isotopic infusion into jugular veins etc.) were technically-demanding, and Dr. Gozho's technical expertise in these areas was invaluable in the implementation of these experimental protocols and in the training of graduate students (Ms. M. Hobin and Ms. K. Baker nee Davies). In the original proposal, $118,650 had been budgeted for graduate student stipends, and these funds covered Dr. Gozho's PDF salary. Two graduate students were still trained on this NSERC-CRD project: 1. Ms. Hobin, whose thesis research project involved Experiment 1; and 2. Ms. Baker, whose Ph.D. thesis research involved Experiments 2 and 4. These graduate students were supported largely from scholarships awarded by the Department of Animal and Poultry Science.

TECHNICAL/PROFESSIONAL ASSISTANTS: $60,000 had been budgeted for a part-time research technician (RT) to assist graduate students with experimental work, animal care etc. However, when Dr. Gozho was hired as a PDF he assumed some of the duties and responsibilities that a RT would have performed, so part of this budget was used to cover Dr. Gozho's salary. Also, undergraduate students were hired when needed primarily to assist with animal care duties (feeding, cleaning etc.) that would have been performed by a RT. A major portion for this budget ($25,143 under professional/tec services) was used to cover the purchase of experimental animals and professional/technical services (veterinary surgeons) relating to surgical procedures on experimental animals (intestinal and rumen cannulation etc.). Because of technical problems that were encountered (see the section on "Problems Encountered" in this report) at the beginning of the project, animal care costs escalated way beyond what had been budgeted for initially, and additional costs were encountered in replacing lost animals and the use of alternative experimental protocols to achieve project objectives. For these reasons, most of the $60,000 that had been budgeted for a RT was diverted to paying for these unforeseen costs. Nonetheless, all experimental objectives were still achieved within the allocated budget.

PUBLICATION COSTS: $10.850 had originally been budgeted for publications costs, but only $1973 was spent on this budget item. The original budget had been calculated assuming 1 peer-reviewed paper in Year 1 and 2 in each of Years 2 to 4. However, only 2 peer-reviwed papers have been published by the termination of the project. Numerous scientific abstracts were published on research conducted under this project and it is envisaged that additional peer-reviewed manuscripts will be published from Ms. Baker's thesis research. The major reason for the delay in publishing these results is that Ms. Baker's research progress was impeded when she needed time away to deal with personal issues.