

**GRADUATE STUDENT COMPETITION:
ADSA PRODUCTION ORAL
COMPETITION, PhD**

0348 Antioxidant activity after in vitro gastrointestinal digestion of cheese containing catechins encapsulated within liposomes. A. Rashidinejad^{1,2}, D. Everett^{1,2}, J. Birch¹, and D. Sun-Waterhouse³,
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Two important green tea phenolic compounds, (+)-catechin and (–)-epigallocatechin gallate (EGCG) were first encapsulated in soy lecithin liposomes and then incorporated into a low-fat hard cheese to determine the effect of cheese ripening and simulated digestion on the antioxidant activity and recovery of incorporated catechins. The total antioxidant activity (TAA) was measured after in vitro gastrointestinal digestion of cheese that was ripened over 90 d at 8°C to evaluate the efficacy of added antioxidants. Total phenolic content (TPC) was measured using the Folin-Ciocalteu assay, while TAA by both ferric reducing antioxidant power (FRAP) and oxygen radical absorbance capacity (ORAC) assays. The correlation coefficients among the TPC and TAA assays, and the recovery of the encapsulated phenolics for cheese fortification (measured by HPLC), were calculated. Fortification of low-fat cheese with either catechin or EGCG encapsulated in liposomes, led to a significant increase ($p < 0.01$) in TPC, FRAP, and ORAC values measured within the digesta matrix, with no significant impact on cheese composition, pH, and yield. Catechin and EGCG were not detected in the cheese whey, indicating their complete retention in the cheese curd. The phenolic recovery from the digesta was about half of the initial concentration for catechin, i.e., 51.3, 53.9, and 46.0% for the fresh (Day 0) cheese and cheese ripened for 30 and 90 d (Day 30 and 90 cheese), respectively, and more than one third for EGCG, i.e., 38.8, 33.7, and 33.5% for Day 0, 30, and 90 cheese, respectively. TPC values were highly correlated with both the FRAP values (i.e., correlation coefficient was 0.97, 0.98, and 0.96 for Day 0, 30, and 90 cheese, respectively) and ORAC values (i.e., correlation coefficient was 0.98, 0.98, and 0.97 for Day 0, 30, and 90 cheese, respectively). Moreover, the corresponding FRAP and ORAC values were also highly correlated with the coefficients being 0.98, 0.97, and 0.98 for Day 0, 30, and 90 cheese, respectively, suggesting the suitability of these assays for evaluating the TAA of fortified cheese. Thus, the manufacture of a low-fat hard cheese fortified with encapsulated catechin and epigallocatechin gallate in liposomes is feasible with good retention of phenolics and high antioxidant activity. Further investigations on aspects such as bioavailability of fortified phenolics, dose for phenolic consumption,

and sensory attributes of fortified cheese are still required before product commercialization.

0349 Effects of mineral salts and calcium chelating agents on the functionalities of milk protein concentrate prepared by ultrafiltration. X. Luo*, L. Ramchandran, and T. Vasiljevic, Victoria University, Melbourne, Australia.

Functionality of milk protein concentrates can be tailored by modifying state of casein micelles through manipulation of processing conditions including temperature, pH and/or addition of calcium chelators. The objective of this study was to investigate the effect of calcium and calcium chelating agents (EDTA and citrate) on the performance of membrane ultrafiltration (UF) process and the functionalities of resulting milk protein concentrates (MPC). Skim milk adjusted to pH 5.9 was pre-treated with EDTA or citric acid (10, 20 or 30 mmol) and ultrafiltered using a polyethersulfone (PES) membrane at 15°C to five times concentration factor. The membrane performance was measured by the permeate flux during UF process. Used membranes were examined using scanning electron microscopy (SEM). The MPC samples were freeze dried and powders were assessed for physical functionalities including solubility, heat stability and emulsification. Addition of chelators led to a shift in a protein-mineral equilibrium and calcium dissociation from the casein micelle. The total calcium in the final MPC was reduced ($p < 0.05$) from 191 (control) to 131 mM or 135 mM for skim milk pre-treated with 30 mmol of EDTA or citrate, respectively. The casein micelle particle size was subsequently reduced ($p < 0.05$) from 200 nm (control) to 28 nm or 24 nm for the milk pre-treated with EDTA or citrate at concentrations equal to or greater than 20 mmol. Consequently, solubility of the MPC increased ($p < 0.05$) from 92% (control) to 98% (EDTA, ≥ 20 mmol) or 98.9% (citrate, 30 mmol); heat stability was also enhanced ($p < 0.05$) from 78% (control) to 83% (EDTA, 20 mmol) or 87% (citrate, ≥ 20 mmol). The emulsion capacity has increased from 1170 (control) to 1392 or 1459 (g oil/g protein) ($p < 0.05$) when 30 mmol of EDTA or citrate were added, respectively. Addition of EDTA or citrate hindered the membrane performance as observed by reduced permeate flux from 10.5 kg/h.m² (control) to 7.9 kg/h.m² (EDTA, ≥ 20 mmol) and 8.6 kg/h.m² (citrate, ≥ 20 mmol) at the start of UF. Consequently UF processing time increased from 5 h (control) to 7 h (EDTA) or 6 h (citrate). This work has provided new insights into the relationship between calcium, calcium chelators and their influence on the casein micelle size and the physicochemical properties of MPC produced using UF, and also demonstrated the potential of using EDTA and citrate acid to manipulate MPC product functionality using UF.

Key Words: milk protein concentrate (MPC), functionality, ultrafiltration (UF), membrane, calcium chelator, casein micelle

0350 Effects of slow-release urea, rumen-protected methionine, and histidine on performance of dairy cows fed metabolizable protein-deficient diets.

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The main objective of this experiment was to investigate the effects of slow-release urea and rumen-protected (RP) Met and His supplementation of a metabolizable protein (MP)-deficient diet on lactation performance of dairy cows. Sixty Holstein cows (DIM, 87 ± 40 and BW, 640 ± 70 kg) were used in a 10-wk randomized complete block design trial. After a 2-wk covariate period, cows were blocked by parity, DIM, and milk yield, and randomly assigned to 1 of 5 dietary treatments: MP-adequate diet [AMP; 107% of MP requirements (NRC, 2001)]; MP-deficient diet [DMP; 95% of MP requirements]; DMP supplemented with slow-release urea as Optigen (Alltech Inc.; DMPO); DMPO supplemented with RPMet as Mepron (Evonik Industries AG; DMPOM); and DMPOM supplemented with RPHis (Balchem Corp.; DMPOMH). The basal diet consisted of (DM basis): 43% corn silage, 8% grass hay, 4% cottonseed hulls, and 45% concentrate and contained 16.7, 15.8, and 14.8% CP for AMP, DMPO, and DMP, respectively. Total-tract apparent digestibility of nutrients, and urinary N and urea excretions were decreased ($P < 0.01$) by DMP compared with AMP. Relative to AMP, milk N secretion as a proportion of N intake tended to be higher ($P = 0.07$) for DMP. DMI was not affected by MP level but tended to be higher ($P = 0.09$) for the DMPOMH (28.4 kg/d) compared with DMPOM (27.0 kg/d). Yields of milk and milk fat were not affected by treatment, averaging 44.0 kg/d and 1.56 kg/d, respectively; milk fat content tended to be lower ($P = 0.06$) for DMPOMH (3.36%) than DMPOM (3.78%). Milk true protein content was increased (3.26 vs. 3.16%, $P = 0.04$) and milk protein yield was numerically increased (1.49 vs. 1.39 kg/d, $P = 0.14$) by DMPOMH, compared with DMPOM. Cows fed DMP gained 14 g/d BW whereas cows on all other treatments gained on average 267 g/d ($P \leq 0.10$). Supplementation of the DMPO diet with RPAA increased ($P = 0.03$) plasma glucose and numerically increased ($P = 0.12$) plasma insulin. In conclusion, feeding a 5% MP-deficient diet did not decrease DMI and yields of milk and milk components, despite the reduction in nutrient digestibility. Supplementation of the DMPOM diet with RPHis tended to increase DMI and increased milk protein content. These results confirm previous data and suggest that His may have a positive effect on voluntary feed intake in high-yielding dairy cows.

Key Words: metabolizable protein, slow-release urea, rumen-protected methionine, rumen-protected histidine

0351 Effect of dietary phosphorus on intestinal P absorption in growing Holstein steers. X. Feng^{*},

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The effect of dietary phosphorus (P) intake on intestinal P absorption was evaluated in growing steers. Diets varying in P content (0.15%, 0.27%, 0.36% and 0.45%, DM basis) were fed to 8 growing Holstein steers (174 ± 10 kg BW) fitted with permanent duodenal and ileal cannulas in a replicated 4×4 Latin square with 14 d periods. Ytterbium-labeled corn silage and cobalt-EDTA were used as particulate and liquid phase markers, respectively, to measure digesta flow. Duodenal and ileal samples and spot samples of urine were collected every 9 h from d 11 to 14. Total fecal collection was conducted on d 11 to 14 with fecal bags. Blood samples were collected from the coccygeal vessel on d 14. Feed, digesta, and fecal samples were analyzed for total P and Pi using the molybdovanadate yellow method and blue method, respectively. Data were analyzed using PROC GLIMMIX in SAS with a model including treatment, square, period and interaction of treatment and square. Preplanned contrasts were used to evaluate linear and quadratic treatment effects. Results are reported as least square means. Dry matter intake (mean = 4.90 kg/d, 2.8% of BW) and apparent DM digestibility (mean = 78.1%) were unaffected by treatment. Duodenal and ileal flow of total P increased linearly with increasing P intake (13.4, 18.5, 23.0 and 27.4 g/d, $P < 0.01$; 6.80, 7.87, 8.42, and 10.4 g/d, $P < 0.05$). Increasing P intake linearly increased the quantity of P absorbed from the small intestine (6.96, 11.1, 14.6 and 17.2 g/d, $P < 0.01$) but absorption efficiency was unchanged (mean = 59.6%). Phosphorus was absorbed on a net basis from the large intestine, but this was not affected by treatment and was a small percentage of total P absorption. Blood Pi increased linearly with increased dietary P (4.36, 6.31, 7.68, and 8.5 mg/dL, $P < 0.01$) and salivary P secretion was unchanged (mean = 5.79 g/d) suggesting that rumen function was prioritized during short-term P deficiency. The absence of change in absorption efficiency and salivary P secretion in the face of short term P deficiency may be used to improve published models of P digestion, absorption, and metabolism.

Key Words: phosphorus, absorption, growing steers

0352 A survey of calving and colostrum management practices on Irish dairy farms. C. Cummins^{*1,2},

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This study aimed to identify calving and colostrum management practices on Irish dairy farms that may compromise neonatal

calf health. The study population was randomly selected from the Irish Cattle Breeding Federation (ICBF) HerdPlus group ($n = 320$) and balanced for herd size and geographical location. The survey consisted of four sections: cow management, calving management, colostrum management and calf management (calving and colostrum are described here). Questions related to hygiene, type of calving pens used, and colostrum collection, storage, and feeding management. Surveys were mailed between 11 July and 15 August 2013. Responses were entered onto the online package SurveyMonkey (www.surveymonkey.com). Coded responses were downloaded to one file and data were collated using Microsoft Excel. A univariable chi-square analysis (significance $P < 0.05$) was performed using 'PROC FREQ' in SAS (v9.3), with two independent variables: milk production (MP), and enterprise. Milk production category one (MP1) included suppliers with a milk production limit (quota) of $\leq 380,000\text{L}$; MP2 $> 380,000\text{L}$ and $< 600,000\text{L}$; and MP3 $\geq 600,000\text{L}$. Enterprise was divided into specialist dairy farms (SD) and dairy farms with another enterprise (DO). The final response rate was 85%. On univariable analysis, group calving pens were more common among MP3 (60%) than MP1 (37%; $P < 0.05$), who tended to use individual pens (24% MP3; 47% MP1; $P < 0.05$). Cleaning of calving pens was infrequent across all study herds ($42\% \leq 1 \times \text{month}$), while 81% left calves in calving pens for ≥ 2 h after birth. Regarding colostrum, MP3 respondents more commonly collected colostrum at first herd milking post-calving compared to MP1 and MP2, the majority of which collected within 2 h ($P < 0.05$). Most SD herds collected colostrum at the first scheduled milking post-calving compared to DO herds (43% SD; 26% DO; $P < 0.01$). More MP1 herds allowed calves to consume own dam colostrum (79%), compared to MP2 (56%) and MP3 (44%), however they also allowed calves suckle their dam (48% MP1 vs. 40% MP2 and 32% MP3). Consequently, MP1 calves received colostrum earlier compared to MP3 (45% MP1 vs. 35% MP3 within 1 h; $P < 0.05$). Of farms not feeding calves colostrum from their own dam, 32% of MP3 used pooled colostrum for the calf's first feed compared to MP1 (13%) and MP2 (15%). The most common storage method was freezing (46%), mainly for 1–6 mo (44%). This study indicates that calving and colostrum management practices on many Irish dairy farms are suboptimal and may lead to compromised calf health.

Key Words: colostrum, calving, survey

0353 Effects of supplementing lipid-encapsulated echium oil on lactational responses and milk fatty acid composition.

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Echium oil is a terrestrial source of n-3 fatty acids (FA) that is particularly high in the n-3 FA stearidonic acid (SDA; 18:4

6c,9c,12c,15c) which bypasses the rate limiting step of delta-6-desaturase in conversion to eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) in mammalian tissues. The objective of this study was to evaluate the impact of feeding a lipid-encapsulated echium oil (EO) supplement on animal production and milk fatty acid concentrations. Twelve Holstein dairy cattle (229 ± 62 d in milk) were assigned randomly to treatment sequence in a 3×3 Latin Square design. Treatments were a control diet (CON; no added EO), 1.5% EO (1.5% EO), or 3.0% diet dry matter (DM) added EO (3.0% EO). Treatment periods were 14 d with the final 4 d used for sample and data collection. The statistical model included the random effect of cow nested within square and the fixed effects of treatment and period. Compared with CON, EO treatments had no effect on dry matter intake (26.6 kg/day; $P = 0.93$), milk yield (30.5 kg/day; $P = 0.34$), or milk protein yield (1.1 kg/day; $P = 0.84$). Increasing EO supplementation increased milk fat concentration (4.1, 4.2, 4.3%; $P < 0.05$) and fat yield (1.24, 1.27, 1.32 kg/day; $P < 0.05$) but decreased milk protein concentration (3.56, 3.54, 3.47%; $P < 0.01$) for CON, 1.5% EO, and 3.0% EO, respectively. Compared with CON, the concentration of total saturated FA in milk fat decreased with increasing EO supplementation (73.7, 72.4, and 71.1 g/100 g FA, $P < 0.0001$). Increasing EO supplementation increased milk fat concentration of total n-3 FA (0.49, 0.65, 0.81 g/100 g FA, $P < 0.0001$), α -Linolenic acid (18:3 n-3) (0.38, 0.47, 0.58 g/100 g FA, $P < 0.0001$), and SDA (0.02, 0.06, 0.09 g/100 g FA, $P < 0.001$) for CON, 1.5% EO, 3.0% EO, respectively. For 1.5% EO and 3.0% EO milk fat concentration of EPA was 0.05 g/100 g FA vs. 0.03 g/100 g FA for CON ($P < 0.0001$). DHA was not detected in milk fat. Transfer of SDA from the EO supplement into milk fat was 3.4% and 3.2% for the 1.5% and 3% EO treatments, respectively. In conclusion, supplementation with a lipid-encapsulated EO did not negatively impact lactational responses and increased the concentration of n-3 FA in milk fat. The concentration of n-3 FA in milk fat however, was still a minor component of total milk FA.

Key Words: n-3 fatty acid, milk fat, echium oil

0354 Effects of dietary crude protein level on nitrogen use efficiency and urinary nitrogen excretion during a twelve-week period in late lactation dairy cows.

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Our objectives were to determine the impact of decreasing levels of dietary CP and whether there was a treatment by week interaction on intake-N, milk-N (DHI true protein), N use efficiency (NUE, milk-N/intake-N), MUN, and responses related to urinary excretion in late lactation cows. One hundred twenty-eight Holstein cows (means \pm SD; 736 ± 18 kg BW; 224 ± 54 DIM) were used in a 16-pen study with 8 cows

per pen, and fed a TMR once per day (at 8:00 am) for 12 wk (pen = experimental unit). Treatments which included diets of 11.8, 13.1, 14.6, or 16.2% CP (DM basis) were randomly allocated to pen for the entirety of the experiment. Rations consisted of approximately 67% forage (half corn silage; half alfalfa silage); soy hulls replaced soybean meal to achieve the desired dietary CP levels for each treatment. Urine volume was estimated using creatinine concentration in spot urine samples collected 6 h before feeding for a group of four randomly selected cows in each pen. This protocol was repeated 6 h after feeding. LS-means of pen-level data presented in the table were obtained on week 2, 8, and 12. Except for urine volume, there was a linear effect for all responses but there were no quadratic effects. There was a treatment by week interaction for most responses. Regardless of treatment, NUE was high and urinary urea-N excretion was low. Under the conditions of this experiment, the 14.6% CP diet allowed for a reduction in urinary urea-N without affecting milk-N.

Key Words: protein nutrition, MUN, urinary urea

Table 0354. Effect of dietary CP on measured responses

Item	Dietary CP (DM basis)				SEM	<i>P</i> -value ¹	
	11.8	13.1	14.6	16.2		L	Trt*wk
Intake-N, g/d	410 ^d	505 ^c	551 ^b	614 ^a	9.0	< 0.01	< 0.01
Milk-N, g/d	128 ^c	149 ^b	163.4 ^{ab}	172.4 ^a	5.04	< 0.01	< 0.01
Milk-N/ Intake-N, %	31.1 ^b	29.4 ^{ab}	29.6 ^{ab}	28.0 ^a	0.006	< 0.01	0.25
MUN, g/dL	6.3 ^d	8.6 ^c	10.9 ^b	13.47 ^a	0.34	< 0.01	0.06
Urine volume, L/d	17.5	18.2	16.8	17.8	0.93	0.95	0.04
Urinary-N, g/L	5.1 ^d	6.4 ^c	7.7 ^b	8.5 ^a	0.24	< 0.01	0.74
Urinary-N, g/d	88 ^c	115 ^b	127 ^{ab}	150 ^a	6.3	< 0.01	< 0.01
Urinary Urea-N, g/L	2.9 ^d	4.6 ^c	5.8 ^b	6.9 ^a	0.29	< 0.01	0.59
Urinary Urea-N, g/d	50 ^d	83 ^c	99 ^b	122 ^a	3.2	< 0.01	< 0.01
Urinary-N/ Intake-N, %	21.6	23.1	23.2	24.4	1.56	0.29	< 0.01

^{a-d} Least squares means within the same row with different superscripts differ (*P* < 0.05).

¹ Linear (L) effect of CP% level in the diet or interaction treatment by week (Trt*wk).

0355 Evaluation of a handheld device for the detection of β-hydroxybutyrate pre-calving in dairy cattle.

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Individual and herd ketone levels are commonly monitored in dairy cattle post-partum to identify individuals at-risk of metabolic disease and to identify potential improvements to management factors. The Precision Xtra handheld meter has been validated for use in β-hydroxybutyrate (BHBA) measurements post-calving as a convenient cow-side test for ketonemia. Recent research has identified BHBA cut points pre-partum associated with increased risk of post-partum disease, but at much lower cut-offs than those indicating hy-

perketonia after calving: 0.6 to 0.8 mmol/L in comparison to 1.0 to 1.4 mmol/L. The objective of the current research is to validate the handheld device, Precision Xtra, in the measurement of BHBA in whole blood against the gold standard method, laboratory evaluation of serum, to assess its diagnostic accuracy in detecting BHBA pre-calving in the range of 0.6 to 0.8 mmol/L. As part of a larger study, 212 cows in 6 herds across southern Ontario were sampled between 3 and 9 d before the expected calving date. Blood was collected and tested on-site with the Precision Xtra device. The serum portion of the sample was separated and sent to a laboratory for measurement of BHBA and non-esterified fatty acid (NEFA) concentrations. The results of the two BHBA measurement methods were compared and evaluated with concordance coefficients. The sensitivity and specificity of the Precision Xtra were determined with receiver operator characteristic curves at cut points of 0.6, 0.7 and 0.8 mmol/L. The two tests had a moderate concordance correlation of 0.77 ± 0.03 (CI_{95} : 0.72 – 0.83) and the area under the curve for each cut point was high with values between 0.90 and 0.93. The Precision Xtra had sensitivities of 85 to 93% and specificities of 76 to 87% depending on the cut point tested. The level of agreement between Precision Xtra cut points and at-risk pre-calving NEFA concentrations of 0.4 and 0.5 mEq/L was calculated. The level of agreement between Precision Xtra BHBA concentrations ≥ 0.8 mmol/L and NEFA concentrations ≥ 0.5 mEq/L was substantial, with a kappa of 0.64. Based on the moderate level of correlation and the good level of sensitivity and specificity, the Precision Xtra is a valid tool in the detection of elevated BHBA pre-calving and may be helpful in identifying individuals at risk of metabolic disease.

Key Words: ketosis, β-hydroxybutyrate, diagnostic test evaluation

0356 Effects of dietary nitrate supplementation on enteric methane and nitrous oxide emissions from beef cattle.

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Feeding nitrate has been proposed as a means to reduce enteric greenhouse gas emissions from ruminants. Nitrate can compete with methanogens for hydrogen in the rumen and therefore reduce methane from eructation. However, increasing the nitrate concentration in the rumen could induce enteric nitrous oxide emissions, potentially nullifying the greenhouse gas reduction achieved from lowering methane emissions. The present study investigated the effects 2% nitrate (on DM basis) versus an isonitrogenous concentration of urea supplemented to finishing steers on enteric methane and nitrous oxide emissions. Sixteen steers were allocated to nitrate and urea treatments in a randomized complete block design (*n* = 8). Eructated emis-

sions were collected using head chambers for 12 h following the morning feeding. Methane was measured using the TEI 55C direct methane analyzer and nitrous oxide using the 46i nitrous oxide analyzer (both were Thermo Environmental Instruments, Franklin, MA). All data were analyzed using the Proc Mixed Model in SAS. The nitrate versus urea treatment lowered methane production at measurement h 1 and 2 ($P < 0.01$), but did not lower overall methane production during the 12 h measurement period. The nitrate versus urea treatment increased nitrous oxide production at h 1, 2, and 3 ($P < 0.05$) of measurement and the overall 12 h measurement period ($P < 0.0001$). Nitrous oxide was detected in both treatments at each time point, with a sixfold increase in production in the nitrate (~600 mg/12 h) versus urea treatment (~100 mg/12 h). Overall, combined greenhouse gas production expressed as carbon dioxide equivalents was similar between treatments. This study indicates that nitrate supplementation in finishing beef cattle is effective at reducing eructated methane in the time immediately following feeding, and might need to be supplemented at a higher concentration and/or more frequently to achieve more optimal methane reduction. Furthermore, this study suggests that cattle could be a source of the potent greenhouse gas nitrous oxide, which is further stimulated by nitrate supplementation. Additional research is necessary to evaluate more effective means of reducing methane with nitrate in finishing beef cattle and the production of nitrous oxide with and without supplementation of nitrate.

Key Words: greenhouse gas, hydrogen sink, ruminant

0357 Early pair housing influences the feeding behavior and development of dairy calves. J. H. C. Costa*, R. K. Meagher, M. A. von Keyserlingk, and D. M. Weary, *Animal Welfare Program—University of British Columbia, Vancouver, BC, Canada.*

Calves are social and gregarious animals. Pre-weaned calves are typically kept in individual pens but little is known about how individual versus social rearing affects development of feeding behavior. The aim of this study was to assess the effects of early and late pairing feeding behavior and weight gain before and after weaning. Holstein bull calves were reared individually ($n = 8$ calves) or paired with another calf at 3 d of age ($n = 8$ pairs) or 42d of age ($n = 8$ pairs). All calves were fed 8 L of milk/d for 4 wk, 6 L/d from 4 to 6 wk and weaned at 8 wk of age. Calves were provided ad libitum access to calf starter and a total mixed ration (TMR). Body weight and feed consumption were followed weekly from 6 wk until 10 wk of age. At 6 wk, intake of TMR averaged (\pm SEM) 0.25 ± 0.05 Kg/d, 0.41 ± 0.12 Kg/d, 0.32 ± 0.09 Kg/d, for individual, and early paired late paired housed calves, respectively. Starter intake was similar for the individually reared and late-paired calves (0.09 ± 0.03 Kg/d and 0.04 ± 0.01 Kg/d) but higher for the early-paired calves (0.23 ± 0.07 Kg/d). Consumption increased after weaning in all treatments, but this increase was

greatest for the early-paired calves. At 10 wk of age, TMR intake averaged 2.89 ± 0.54 Kg/d, 3.27 ± 0.72 Kg/d and 3.08 ± 0.46 Kg/d for individual, early paired and late paired housed calves, respectively. Starter intake averaged 1.26 ± 0.33 Kg/d, 2.20 ± 0.22 Kg/d and 1.09 ± 0.25 Kg/d for the same three treatments. Calves in the early pair treatment showed higher average daily gains (1.13 ± 0.05 Kg/d versus 0.92 ± 0.04 Kg/d and 0.84 ± 0.05 Kg/d for the individual and late-paired calves). Pair housing soon after birth increased calf feed intake and weight gains in comparison with late pairing and individual housing.

Key Words: feeding behavior, group housing, dietary transition

0358 Epigenetic differences of cows classified with biased antibody and cell mediated immune response traits. M. A. Paibomesai*¹ and B. Mallard², ¹*University of Guelph, Guelph, ON, Canada,* ²*Dept Pathobiology, University of Guelph, Guelph, ON, Canada.*

The identification of cattle that are better able to respond immunologically to pathogens would be useful to reduce the incidence of disease on commercial dairy farms. High Immune Response (HIR) technology provides a unique tool to rank animals based on their ability to respond to test antigens, and high ranking dairy cows have been shown to have a lower incidence of disease. HIR evaluates two branches of the adaptive immune response: the antibody mediated immune response (AMIR, prominent IL-4 production) which responds primarily to extracellular pathogens, and the cell mediated immune response (CMIR, prominent IFN-g production), which responds primarily to intracellular pathogens. Genetic control of the immune response has been well studied in past years, but epigenetic influences on phenotype remain to be defined. Epigenetics is defined as modifications to DNA that control gene expression without changing the DNA sequence. Specifically, DNA methylation, an epigenetic modification, is associated with a decrease in gene transcription, while the lack of DNA methylation is associated with increased gene expression. CD4+ helper T cells are mediators of AMIR and CMIR, producing cytokines, such as IL-4 and IFN- γ , for the direction of an appropriate response. Biased immune responder cattle respond strongly with either high AMIR (H-AMIR) or high CMIR (H-CMIR) to test antigens. Cows with H-AMIR/L-CMIR (H-AMIR; $n = 10$) and H-CMIR/L-AMIR (H-CMIR; $n = 11$) phenotypes were used to investigate mechanisms of immune response variation and the role of epigenetics in cattle immune response traits. Isolated CD4+ helper T cells from H-CMIR and H-AMIR cows were stimulated with a T cell mitogen (ConA) and cell culture supernatants were harvested at 24 h to quantify IL-4 and IFN γ by ELISA. DNA was extracted from unstimulated and stimulated cells and bisulfite pyrosequencing was used to quantify DNA methylation for both IL-4 and IFN γ promoters. CD4+ T cells from H-CMIR cows pro-

duced more IFN- γ ($P = 0.059$), and significantly more IL-4 ($P = 0.02$) than T cells from H-AMIR cows, when sampled 21 d into lactation. In H-CMIR cows higher secretion of IFN- γ was associated with decreased methylation in the promoter region of the IFN γ gene compared to H-AMIR cows ($P = 0.01$). In contrast, there was no difference in DNA methylation at the

IL-4 promoter observed between the two immune response phenotypes. This study is the first to show an association between DNA methylation and specific phenotypes.

Key Words: immune response, epigenetics, dairy cows