**M285 The effect of allocation frequency in rotational grazing systems on the fatty acid (FA) profile in milk fat of dairy cows.** B. Vlaeminck*[1], P. A. Abrahamse[2], V. Fievez[1], J. Dijkstra[2], and S. Tamminga[2], [1]*Laboratory for Animal Nutrition and Animal Product Quality, Ghent University, Melle, Belgium*, [2]*Animal Nutrition Group, Wageningen University, Wageningen, the Netherlands*.

Eight Holstein cows were blocked in 2 groups according to milk yield, parity and DIM to evaluate the effect of frequency of allocation to new grazing plots on profiles of FA in milk fat. The 2 treatments were daily allocation to 0.125-ha plots (1D) or allocation every 4 d to 0.5-ha plots (4D) of Lolium perenne L. and were tested in a randomized block design (2 rotations with 2 measuring periods of 4 d each). The model included the effect of treatment, day within treatment, rotation, cow, the interaction between treatment and rotation and the covariate value. Significance was declared at $P<0.05$. There were no differences in the FA composition of the offered and residual pasture between 1D and 4D. Within days in the 4D treatment, the proportion of 16:0, 18:0 and 18:2n-6 increased, and that of 18:3n-3 as well as total FA content of grass decreased linearly. Treatment effects on milk FA composition and secretion were small. In contrast, milk FA composition was largely affected by day within the 4D treatment. Secretion of de novo synthesised and C16-FA decreased linearly during the 4 d whereas secretion of odd- and branched and C18-FA were not affected by day in the 4D treatment. Proportions of trans-11-18:1 in milk fat increased on d 2 (4.60 g/100 g FA) and decreased thereafter (3.86 g/100 g FA on d 4). Proportions of cis-9, trans-11-18:2 (2.34 and 1.83 g/100 g fatty FA on day 2 and 4, respectively), trans-11, cis-15-18:2 (0.81 and 0.63 g/100 g FA on day 2 and 4, respectively) and 18:3n-3 (0.87 and 0.83 g/100 g FA on day 2 and 4, respectively) in milk fat followed the same pattern. Results from this study suggests that increasing pasture allocation frequency from once every 4 d to every day has no effect on profiles of FA in milk. In addition, short term variation in pasture quality during the 4 days affected milk FA composition with a greater effect on biohydrogenation intermediates in milk fat compared with its major precursor, 18:3n-3.

**Key Words:** dairy cow, rotational grazing, milk fatty acids

**M286 Economic analysis of alfalfa hay inclusion in wet corn gluten feed based diets for lactating dairy cattle.** C. R. Mullins* and B. J. Bradford, *Kansas State University, Manhattan*.

A breakeven analysis was conducted to determine if including alfalfa in a ration containing 31% wet corn gluten feed increases production enough to justify feeding it to lactating dairy cattle. Data was gathered from a pen study where alfalfa hay (AH) was shown to linearly increase ECM production and DMI in 80 Holstein cows. Changes in milk income, feed consumed, and feed costs were incorporated in a partial budget model to determine the relative difference in AH vs. corn silage (CS) value (DM basis) at different milk:feed cost ratios. For the economic model, the value of AH was fixed at $250/ton DM ($0.27/kg), and milk value was fixed at $0.20/lb ($0.44/kg), whereas the value of CS and TMR cost varied with the AH price differential and the milk:feed cost ratio, respectively. Changes to the fixed values had little effect on the results, although the model was somewhat sensitive to the soybean meal to corn grain price differential. To account for this, the model was evaluated using maximum ($188/ton), mean ($120/ton), and minimum ($80/ton) price differentials between soybean meal and corn from 2003 to 2008. The additional value of AH relative to CS ranged from less than $5/ton DM to as much as $60/ton DM. Alfalfa's relative value increased as milk:feed cost ratio increased and also increased with increased soybean meal - corn grain price differential. The mean breakeven price differential was $30/ton DM, substantially less than typical differences in on-farm prices for these feedstuffs. The model was also evaluated using data from a study with 0% and 25% AH diets including 15% distillers grains (Kleinschmit et al. 2007 JDS 90:5587). The effect of milk:feed cost ratio on breakeven AH - CS price differential was far greater with this data (range -$20 to $100/ton DM) due to more dramatic treatment effects on DMI and production, and the mean breakeven differential was slightly greater ($60/ton DM). Based on data from these studies, adding AH to diets with large proportions of non-forage fiber may not be profitable, especially when milk:feed cost ratios are low.

**Key Words:** partial budget, alfalfa, dairy

**M287 Effect of alfalfa hay particle size and source of neutral detergent soluble carbohydrates on intake, chewing activity, ruminal fermentation and nutrient digestibility of midlactation cows.** A. Asadi*, G. R. Ghorbani, M. Alikhani, and M. Bagheri, *Department of Animal Sciences, Isfahan University of Technology, Isfahan, Iran*.

The effects on cows′ intake, performance, and digestibility of partial replacement of neutral detergent soluble fiber (NDSF) for starch were studied in diets with two sizes of alfalfa hay. Eight multiparous Holsteins averaging 146 ± 6 DIM and 36.7 ± 2.6 kg/d of milk were used in a double 4 × 4 Latin square design with a 2 ×2 factorial arrangement. Treatments included short (SH) or long (LN) alfalfa each combined with high starch concentrate (HiSt); and SH or LN alfalfa each combined with high NDSF concentrate (HiSF). The HiSt and HiSF treatments contained 28.5% and 24% starch and 8.5% and 13% NDSF, respectively and diet NDF content was 33%. Periods were 21 d in length with last 7d for sampling and analysis. The DM, OM, and CP intakes were unchanged by particle size (PS), while it was greater for HiSt vs. HiSF treatments ($P < 0.05$). The NDF intake was higher in SH vs. LN and in HiSt vs. HiSF treatments ($P < 0.05$). Interestingly, apparent digestibility of DM, OM, NDF and CP were significantly higher for cows fed HiSF diet than cows fed HiSt diet ($P < 0.05$). Total chewing, eating and ruminating times were not affected by treatments, however, eating time per kg NDF intake was longer in LN vs. SH and tended to be longer in HiSF vs. HiSt diets. Sorting behavior was apparent in LN diets in this study; cows sorted against particles > 19 mm and in favor of particles < 8 mm with majority of sorting occurred after 8 h postfeeding. Rumen pH, total VFA and proportion of individual VFA were unaffected by treatments ($P > 0.05$), however, butyrate was significantly higher with HiSF diets. No interaction between PS and carbohydrate form was found for nutrient digestibility and ruminal fermentation variables. In conclusion, results suggested that in diets exceeding minimum NDF recommendations and containing no more than 30% starch, alfalfa PS may not interact with form of carbohydrates to affect rumen metabolism and nutrient digestibility.

**Key Words:** dairy cow, neutral detergent soluble carbohydrates, chewing activity

**M288 Differentiating effects of effective fiber sources on performance of lactating dairy cows.** R. A. Starkey*, P. N. Gott, M. L. Eastridge, E. R. Oelker, A. R. Sewell, B. Mathew, and J. L. Firkins, *The Ohio State University, Columbus*.

Because of high feed costs and limited availability of some forage sources, alternative effective fiber sources are being considered. The objective of this study was to compare the feeding of corn silage as the sole forage versus feeding corn silage with alfalfa hay, wheat straw, and corn stover to lactating dairy cows. A 5x5 Latin square design was used with 5 multiparous cannulated Holstein cows (122 ± 62 DIM). Diets were: 1) CS = corn silage as sole forage source, 2) ALF = corn silage and 11.5% alfalfa hay, 3) STW-5 = corn silage and 5% straw, 4) STW-10 = corn silage and 10% straw, and 5) STV = corn silage and 5.5% corn stover (without cobs). Diets were formulated for similar concentrations of CP, NFC, and 18% forage NDF, except for STW-10 with 23% forage NDF. Diets were fed for 3 wk, and samples were collected during the last week of each period. Period 4 was extended 9 d because one cow had a displaced abomasum at the beginning of the period. The DMI was lowest for STW-10 (28.6, 28.9, 30.5, 26.7, and 28.6 kg/d for CS, ALF, STW-5, STW-10 and STV, respectively; $P < 0.05$). The BW (734 kg) and BCS (3.18; 1 to 5 scale) were similar. Milk (35.6 kg/d), milk fat (3.56%), milk protein (2.87%), and MUN (17.4 mg/dl) also were similar. The different forage sources resulted in similar total tract digestibilities of OM (76.9%) and NDF (63.5%). Rumen pH (6.12), acetate:propionate (3.10), and ruminal concentrations of total VFA, acetate, propionate, butyrate, and valerate were similar. Concentrations of isovalerate ($P < 0.01$) and isobutyrate ($P < 0.10$) were highest for STW-10 compared to the other diets, and isovalerate was also lower ($P < 0.10$) for ALF than for STW-5 and STV. Feeding the higher level of straw may have caused more rumen fill and thus reduced DMI, which would likely lower milk yield in a longer study. Feeding similar forage NDF concentrations using corn, alfalfa hay, and wheat straw can result in similar animal performance and ruminal fermentation with adequate formulation of NFC and total NDF.

**Key Words:** corn stover, straw, effective fiber

**M289 Effect of roughage to concentrate ratio on ruminal parameters and protein degradability in dairy cows.** L. J. Erasmus*[1], W. A. van Niekerk[1], H. Nienaber[1], and P. H. Robinson[2], [1]*University of Pretoria, Department of Animal and Wildlife Siences, Pretoria, South Africa*, [2]*University of California, Department of Animal Science, Davis*.

Published research suggest it might be feasible to decrease ruminal protein degradability by lowering rumen pH through dietary manipulation of rumen fermentation. Three ruminally cannulated Holstein cows were used in a 3 x 3 Latin square design experiment and fed three total mixed rations differing in roughage : concentrate ratio (60 : 40; 45 : 55 and 30 : 70). The roughage portion consisted of equal parts of Lucerne hay and *Eragrostis curvula* hay with maize the primary energy component and the three feedstuffs to be evaluated the primary dietary CP components.The CP (%DM) content of the three diets were 14.7%,15.4% and 16.2%,the NDF content 38.6%,32.6% and 26.4%, and the NFC content 36.1%,41.4% and 46.7% respectively. Protein degradability of three feedstuffs differing in potential degradability (sunflower oilcake meal, cottonseed oilcake meal, roasted soya) was estimated using the in situ technique. Incubation times were 0,2,4,8,16,24,48 and 72h.Rumen fluid samples were taken every 6 hours for three consecutive days during each 16 day experimental period.Repeated variables were analysed with the GLM Repeated Measures Procedure (SAS,2006). Mean ruminal pH over 24 h differed and was 6.44, 6.27 and 6.00 respectively for the 3 diets ($P < 0.05$). Time below a rumen pH of 5.8 was 2.5 h, only for the high concentrate (70%) diet. Mean ruminal NH3-N and VFA concentrations did not differ among treatments although the high concentrate diet resulted in a lower acetic : propionic acid ratio ($P < 0.05$). Dietary treatment did not affect protein degradability within feedstuff ( $P > 0.05$) suggesting that roughage : concentrate ratios ranging from 60 : 40 to 30 : 70 and pH ranging from 6.0 to 6.4 are still within the physiological boundaries where rumen pH do not affect ruminal protein degradability. Alternatives such as chemical or heat treatment would be more feasible than dietary manipulation of pH to increase dietary RUP content when formulating practical dairy cattle diets.

**Key Words:** ruminal protein degradability, roughage to concentrate ratio, dairy cows

**M290 Effect of decreasing forage fiber in close-up cows diets on rumination time, DMI and subsequent lactation performance.** A. Nikkhah*[1], V. Keshavarz[2], H. Amanloo[2], M. Dehghan[1], and M. Kazemi Bonchenari[1], [1]*Department of Animal Sciences, University of Tehran, Karaj, Iran*, [2]*Department of Animal Sciences, University of Zanjan, Zanjan, Iran*.

One of the main goals of the close-up diets is to maximize DMI in the subsequent lactation period to support high energy demand for early lactation. Previous studies clearly demonstrated that decreasing fiber concentration in dairy cows diets increased DMI of the cows. From the other hand decreasing the fiber in diets reduced the rumination time. The objective of this study was to improve DMI with decrease forage fiber but still provide required eNDF without effect on rumination time and milk fat content. 18 multiparous close-up dairy cows in 31d (SD=6) before expected calving date fed different levels of forage fiber and also effective fiber (the forage fiber and effective fiber were 30.5-30.4, 26.7-27.38 and 22.9-24.61 for diets 1, 2 and 3 respectively) were assigned in to a completely randomized design to investigate the effect of experimental diets on rumination activity and also on DMI and production traits in the subsequent lactation. The different diets were fed to dairy cows in close-up period and a similar diet was fed to all cows in lactation period and data collection period was lasted until 20d after parturition. The Data were analyzed with SAS (SAS institute 1999). As it is given in table 1, the highest DMI in both close-up and early lactation periods were for diet 2. The results showed that decreased forage fiber in close-up period increase DMI but severe decreasing the forage decreased rumination time (P<0.03). It could be concluded that it is recommendable to decrease forage fiber in close-up diets but this decrease should not as much as to address rumination time.

**Table 1. Effect of different forage fiber in close-up diets on rumination time, DMI and production traits**

| | Treatments | | | | |
|---|---|---|---|---|---|
| Item | 1 | 2 | 3 | SE | P<F |
| Close-up period | | | | | |
| Rumination time (min/d) | 473[b] | 443[ab] | 408[a] | 40.15 | 0.03 |
| DMI (kg/d) | 12.07[b] | 14.29[a] | 12.89[b] | 0.32 | 0.0007 |
| Lactation period | | | | | |
| Milk Yield (kg/d) | 28.40 | 31.1 | 29.83 | 0.85 | 0.09 |
| DMI (kg/d) | 17.41[c] | 19.03[a] | 18.72[b] | 0.43 | 0.001 |
| Milk Fat (kg/d) | 1 | 1.02 | 0.93 | 0.06 | 0.23 |
| Milk Protein (kg/d) | 0.83 | 0.97 | 0.95 | 0.04 | 0.07 |

[a,b,c] Least squares means within the same row without a common superscript differ ($P < 0.05$)

**Key Words:** forage fiber, rumination time

**M291 Feed sorting of dairy cows receiving diets different in dietary fiber level.** O. AlZahal*, M. S. Douglas, S. L. Greenwood, and B. W. McBride, *University of Guelph, Guelph, ON, Canada*.

The objectives of this study were to nutritionally induce subacute ruminal acidosis (SARA) in lactating dairy cows by feeding a low-fiber diet and examine the cows' feed sorting responses in comparison with cows consuming a higher-fiber diet. Six rumen-fistulated Holstein dairy cows (639 ± 51 kg of body weight) were used in the study. Cows were randomly assigned to 1 of 2 dietary treatments, a high-fiber (HF; % of dry matter, 40% corn silage, 27% alfalfa silage, 7% alfalfa hay, 18% protein supplement, 4% ground corn, and 4% wheat bran) or a low-fiber (LF; % of dry matter, 31% corn silage, 20% alfalfa silage, 5% alfalfa hay, 15% protein supplement, 19% ground wheat, and 10% ground barley) total mixed ration. Ruminal pH was continuously recorded 3 d a week and feed sorting was assessed on d 19. The rumen fluid in cows receiving the LF diet on average was below pH 5.6 for a longer duration than in cows receiving the HF diet (357 vs. 103 min/d), indicating that the LF cow were suffering from SARA. Results showed that cows sorted fine particles (< 1.18 mm) differently across treatments ($P < 0.05$), cows receiving the HF diet sorted for fine particles, whereas cows receiving the LF diet sorted against those particles, likely in an attempt to attenuate their rumen condition (105.5 ± 2.7% and 93.9 ± 2.7% of predicted intake of fine particles, respectively). Additionally, sorting of the fine particles was positively correlated with mean pH ($R^2 = 0.77$, $P = 0.02$) and was negatively correlated with durations (min/d) ruminal fluid was below pH 5.8 ($R^2 = 0.73$, $P = 0.03$). In summary, the study demonstrated that cows undergoing SARA sorted their feed to avoid fine particles in attempt to attenuate their condition. The extent of this alteration was related to the magnitude of ruminal pH depression.

**Key Words:** dairy cow, subacute ruminal acidosis, feed sorting

**M292 Corn bran vs. corn grain at two levels of forage: Intake and production responses by lactating dairy cows.** C. Arndt*[1], L. E. Armentano[1], and M. B. Hall[2], [1]*Department of Dairy Science, University of Wisconsin, Madison*, [2]*U.S. Dairy Forage Research Center, University of Wisconsin, Madison*.

A cow performance trial was conducted to estimate the effect of substituting corn bran (CB) for corn grain and to determine if the impact was affected by the level of forage in the diet. Twelve multiparous and 12 primiparous Holsteins were assigned to a 2x2 factorial (CB vs. corn, and high vs. low forage) using 6 4x4 randomized complete block Latin square design with 3 week periods. Cows were blocked by parity and three levels of production within parity. Feed samples were collected every second day over the last 8 days of each period. Dry matter intake and milk weights were recorded over the last 5 days of each period. Milk samples were collected over the last 5 days of each period at am and pm milking. Intake and milk data were averaged across days for each cow in each period. The forage was 64.2 or 38.4% of TMR DM. The high forage corn diet (HFC, 29.7% NDF, 25.6% starch by analysis) had 20% corn, the low forage corn diet (LFC, 23.1% NDF, 34.0% starch) had 39.3% corn, the high forage CB diet (HFCB, 42.6% NDF, 14.8% starch) had 19.3% CB and the low forage CB diet (LFCB, 48.4% NDF, 12.9% starch) had 37.9% CB (DM basis). The LFC diet supported maximum milk and milk protein yield. Milk fat yield on the LFC diet did not differ from HFC or LFCB but milk fat % was lowest for the LFC diet. Loss in milk yield due to substitution of CB for corn was greater on the lower forage diets, but this also corresponded to a larger substitution of CB for corn. Intake was depressed by forage and CB, but lower intake does not appear large compared to reduction in milk yield. Lower milk protein and higher MUN suggests that lack of starch in the CB diets may have interfered with rumen microbial protein synthesis. The effect of substituting CB for both forage and corn grain can be estimated by comparing LFCB to HFC. This way of using CB significantly reduced milk fat yield, but numerical reductions in milk and milk protein yield were not statistically significant.

**Table 1.**

| Item | HFC | HFCB | LFC | LFCB | SE | Forage | Corn | Forage × Corn |
|---|---|---|---|---|---|---|---|---|
| DMI kg/d | 24.8$^{ab}$ | 24.3$^{a}$ | 26.7$^{c}$ | 25.9$^{bc}$ | 0.44 | ** | * | |
| Milk kg/d | 42.0$^{b}$ | 38.7$^{a}$ | 46.7$^{c}$ | 40.5$^{ab}$ | 0.79 | ** | ** | ** |
| Fat kg/d | 1.70$^{c}$ | 1.51$^{a}$ | 1.61$^{bc}$ | 1.58$^{b}$ | 0.04 | | ** | ** |
| Protein kg/d | 1.25$^{b}$ | 1.12$^{a}$ | 1.47$^{c}$ | 1.19$^{ab}$ | 0.02 | ** | ** | ** |
| Fat % | 4.04$^{b}$ | 3.91$^{b}$ | 3.47$^{a}$ | 3.90$^{b}$ | 0.10 | ** | * | ** |
| Protein % | 3.00$^{b}$ | 2.91$^{a}$ | 3.18$^{c}$ | 2.95$^{ab}$ | 0.04 | ** | ** | ** |
| MUN mg/dl | 13.38$^{b}$ | 17.54$^{c}$ | 10.70$^{a}$ | 18.37$^{c}$ | 0.66 | * | ** | ** |

$^{a,b,c}$ Means in a row not sharing a common superscript differ (P<0.05). **<0.01, *<0.05, blank>0.05.

**Key Words:** corn bran, milk

**M293 Corn bran vs. corn grain at two levels of forage: Apparent digestibilities by lactating dairy cows.** C. Arndt*[1], L. E. Armentano[1], and M. B. Hall[2], [1]*Department of Dairy Science, University of Wisconsin, Madison*, [2]*U.S. Dairy Forage Research Center, University of Wisconsin, Madison*.

A digestibility trial was conducted to estimate the effect of corn bran (CB) for corn grain and to determine if the impact was affected by the level of forage in the diet. Apparent digestibility was determined with Yb as an external marker in conjunction with a lactation performance trial using 24 Holsteins in 6 4x4 Latin squares with 3 week periods. Ort, TMR and fecal samples were taken over the last 5 days of each period. Individual feed samples were taken every second day over the last 8 days of each period. Apparent digestibilities and intakes were averaged across days for each cow in each period. The forage (55% corn silage DM, 45% wilted alfalfa silage DM) was 64.2 or 38.4% of TMR DM. The concentrate consisted of either primarily corn or CB (75.1% NDF, 14.6% starch) with added whole cottonseeds, distillers dried grains with solubles, blood meal, soybean meal and a mineral mix. The high forage corn (HFC, 29.7% NDF, 25.6% starch) diet contained 20% corn, the low forage corn (LFC, 23.1% NDF, 34.0% starch) diet contained 39.3% corn, the high forage CB (HFCB, 42.6% NDF, 14.8% starch) diet contained 19.3% CB and the low forage CB (LFCB, 48.4% NDF, 12.9% starch) diet contained 37.9% CB (DM basis). Observed digestible organic matter intake (DOMI) was not significantly different between the two CB diets, as increasing CB based concentrate in place of forage actually reduced organic matter digestibility (OMD) while increasing intake. Corn based diets had greater OMD, and LFC provided the greatest OMD, OM intake (OMI) and DOMI of all diets. No significant difference was found for starch digestibility (StarchD) among the diets, but it was higher than non-starch NFC digestibility (NSNFCD) and NDF digestibility (NDFD), resulting in the greater OMD for corn diets. The greater DOMI of the corn diets results from greater OMI and starch content resulting in greater digestible starch intakes (DStarchI) and digestible NSNFC intakes (DNSNFCI) which compensate their lower digestible NDF intake (DNFDI).

**Table 1.**

| Item | HFC | HFCB | LFC | LFCB | SE | Forage | Corn | Forage × Corn |
|---|---|---|---|---|---|---|---|---|
| OMI kg/d | 22.9[a] | 22.4[a] | 24.7[b] | 24.1[b] | 0.41 | ** | * | |
| DOMI kg/d | 15.2[b] | 13.7[a] | 16.8[c] | 14.0[a] | 0.31 | ** | ** | ** |
| OMD % | 66.3[c] | 61.2[b] | 67.7[c] | 58.3[a] | 0.68 | | ** | ** |
| NDFD % | 38.9[b] | 42.0[b] | 29.1[a] | 41.2[b] | 1.62 | ** | ** | ** |
| StarchD % | 96.7[a] | 97.0[a] | 96.1[a] | 96.8[a] | 0.31 | | | |
| NSNFCD % | 75.3[ab] | 77.4[b] | 71.7[a] | 76.4[b] | 1.36 | | ** | |
| DNDFI kg/d | 4.5[b] | 6.8[c] | 4.1[a] | 8.2[d] | 0.12 | ** | ** | ** |
| DStarchI kg/d | 6.1[b] | 3.5[a] | 8.6[c] | 3.2[a] | 0.13 | ** | ** | ** |
| DNSNFCI kg/d | 1.1[b] | 0.9[a] | 1.1[b] | 0.8[a] | 0.06 | | ** | |

[a,b,c,d]Means in a row not sharing a common superscript differ (P<0.05). **<0.01, *<0.05, blank>0.05.

**Key Words:** corn bran, digestibility

**M294 Effects of increasing levels of concentrate supplementation on milk production of grazing dairy cows.** G. A. Gagliostro*, L. Antonacci, P. Barbera, D. A. Garciarena, and C. A. Cangiano, *Instituto Nacional de Tecnología Agropecuaria*, *Balcarce, Buenos Aires, Argentina*.

The effect of supplemental energy was evaluated in twelve Holstein cows grazing spring pastures (alfalfa 70% and orchardgrass 30%) with an herbage allowance of 27 kg DM/cow/d. Cows were blocked by DIM and randomly assigned to three treatments in a replicated Latin square. At the start of the experiment the six cows from square 1 (514 kg LW) were producing 28.2 kg milk/d and averaged 35 DIM. In Square 2, LW, milk yield and DIM were 554 kg, 31.3 kg/d and 197 d respectively. Treatment periods were 19 d., the first 14 d as adaptation and the last 5 d as the sampling period. Treatments (C3, C6, and C9) were three levels (3, 6, and 9 kg/d) of concentrate (90.4% DM) containing (g/kg DM) CP (160), starch (351), soluble carbohydrates (149), ether extract (39.7) and NDF (248). Supplements were thoroughly consumed by cows. Pasture (30.78% DM) contained (g/kg DM) CP (183), NDF (368), ADF (217), soluble carbohydrates (94.6), starch (33.5) and digestible DM (709). Data were analyzed with effect of square, period, cow within square; treatment, interaction between period and treatment (NS) and residual error. Pasture intake averaged 11.94 kg DM/cow/d (P<0.69). Substitution rate of pasture was low either in C6 (0.10) or in C9 (0.19). Total DM (kg/d) and ME (Mcal/d) intake was higher (P<0.05) in C9 (19.38 and 51.4) compared to C3 (15.12 and 39.3) but not to C6 (17.62 and 46). Milk yield (27.5 kg/d) and FCM (23 kg/d) resulted higher (P<0.05) in C9 compared to C6 (25 and 21 kg) and to C3 (23.6 and 20.3 kg). No differences were detected between C3 and C6. Milk fat content (29.9 g/kg) or yield (754 g/d) and milk protein content (31.6 g/kg) did not differ. Protein yield (g/d) resulted higher (P<0.05) in C9 (885) compared to C3 (735) but not to C6 (789). Lactose (48.6 g/kg) and milk urea (39.98 mg/dl) content did not differ. BW gain (kg/d) tended (P<0.15) to increase with intake of concentrate (C3 = 0 .246, C6 = 0,364 and C9 = 0,772). In high quality spring pastures increasing the amount of concentrate in the diet up to 9 kg/d increased total DM and ME intakes, milk and protein yields without depressing mil fat content or pasture DMI.

**Key Words:** grazing dairy cows, concentrate levels, milk production

**M295 Effect of dietary concentrate level on rumen fermentation, digestibility, and nitrogen losses in dairy cows.** M. Agle*[1], A. N. Hristov[2], S. Zaman[1], and C. Schneider[1], [1]*University of Idaho*, *Moscow*, [2]*Pennsylvania State University*, *University Park*.

The objective of this experiment was to investigate the effect of level of concentrate in the diet on rumen fermentation, digestibility, and N losses in lactating dairy cows. The experiment was a replicated Latin square design with 2-wk adaptation periods. Ruminal contents were exchanged between cows at the beginning of each adaptation period. The diets contained (DM basis): 52 (LowC; control), 61 (MedC), and 72% (HighC) concentrate. Crude protein content of the diets was 16.5, 14.8, and 16.4%. Dry matter intake was not affected by diet ($P = 0.76$). Milk yield was increased ($P = 0.046$) by HighC compared with LowC and MedC (36.0, 33.2, and 32.3 kg/d, respectively). Concentration of milk true protein was not different ($P = 0.23$) among diets, but milk fat percentage (and yield) was decreased ($P = 0.019$) by MedC and HighC compared with LowC. Urinary N excretion and MUN concentration were decreased ($P = 0.003$ and $< 0.001$) and plasma urea N tended to be decreased ($P = 0.09$) by MedC compared with the other diets. Both MedC and HighC decreased ($P = 0.013$) rumen pH compared with the control (6.00, 6.04, and 6.14, respectively). Rumen ammonia concentration was lowered ($P < 0.001$) by HighC and MedC compared with LowC. Propionate concentration was increased ($P = 0.006$) by HighC and tended to be increased ($P = 0.07$) by MedC compared with LowC. Acetate:propionate ratio was similar for LowC and MedC and lower for HighC ($P = 0.008$). Rumen methane production, polysaccharide-degrading activities, and microbial protein synthesis (estimated based on urinary purine derivatives excretion) were not affected ($P = 0.30$ to 0.86) by diet. Apparent total tract digestibility of DM, OM, N, and starch were not affected ($P = 0.08$ to 0.41) by diet, but NDF digestibility was decreased ($P = 0.007$) by MedC and HighC. Increasing concentrate level of the diet increased milk yield and decreased rumen ammonia concentration, but decreased NDF digestibility. The MedC diet decreased urinary N losses.

**Key Words:** dietary energy, rumen fermentation, dairy cow

**M296 Feeding dairy cows rolled barley grain treated with lactic acid and heat delays *in situ* DM disappearance and prevents development of sub-acute ruminal acidosis.** Q. Zebeli*, A. Mazzolari, S. M. Dunn, and B. N. Ametaj, *University of Alberta*, *Edmonton, AB, Canada*.

Due to the high content in readily degradable carbohydrates, inclusion of barley grain in the diet of dairy cows is associated with high risk of sub-acute ruminal acidosis (SARA). The aim of this study was to evaluate the effects of feeding barley grain treated with lactic acid (LA) and heat on *in situ* DM degradation and diurnal rumen pH patterns. Eight rumen-fistulated Holstein cows (170 DIM) were fed once daily (at 0800) a TMR containing rolled barley grain (32.8%, DM basis) steeped for 48h in equal quantity of water (CTR) or 1% LA solution (v/v) and oven-heated at 55°C (TRT). A crossover design with two 21-d periods (first 11d of adaptation diet) was used. Duplicate samples of rolled barley grain were incubated *in situ* (Dacron-bag technique) for 72-h in the rumen of cows fed the CTR- or TRT-diet, and the DM disappearance rate was measured at 0, 2, 4, 8, 16, 24, 48, and 72h post-incubation. Rumen fluid pH was measured on d10 of the measurements period on samples collected at 0h (pre-feeding) and at 2, 4, 6, 8, 10, and 12h post-feeding. The in situ data showed that cows fed the TRT diet had lower DM disappearance rate of barley grain substrate from 0h (19.4 vs. 36.5%; P<0.001) up to 24h (73.4 vs. 80.7%; P=0.02) post-incubation. However, DM disappearance rates were equal in both treatments starting

at 48h (86.5 and 87.2% for TRT- and CTR-diet, respectively; P=0.59) until 72h (P=0.47) post-incubation. Moreover, cows fed the TRT diet showed greater *in situ* lag time than controls (9.2 vs. 4.2h; P<0.001). Additionally, results showed higher (P=0.01) rumen pH for cows fed the TRT diet. Thus, cows fed the TRT diet maintained higher rumen pH (5.92 vs. 5.67) at the nadir (8h post-feeding), as well as at 10h (6.02 vs. 5.75) and 12h (6.10 vs. 5.78) post-feeding. In conclusion, feeding dairy cows 32.8% rolled barley grain treated with 1% LA and heat delayed *in situ* DM disappearance and prevented the decline of rumen pH at SARA levels post-feeding.

**Key Words:** barley grain, dairy cow, lactic acid processing

**M297 Dietary energy source in primiparous dairy cows during the transition period: Blood metabolites, metabolic hormones and milk production.** M. A. T. Artunduaga*[1], S. G. Coelho[1], B. G. Campos[1], A. M. Borges[1], A. M. Q. Lana[1], R. B. Reis[1], H. M. Saturnino[1], H. N. Da Costa[2], and R. V. Sá Fortes[2], [1]*Federal University of Minas Gerais*, *Belo Horizonte, Minas Gerais, Brazil*, [2]*Human Resources on Agribusiness, ReHAgro*, *Belo Horizonte, Minas Gerais, Brazil*.

Increasing the availability of glucogenic and lipogenic nutrients during the transition period has been hypothesized to improve energy balance and to decrease the incidence and severity of metabolic and reproductive disorders in early lactation. Therefore, the objectives of this study were to compare the effects of a glucogenic or a lipogenic diet on blood metabolites, metabolic hormones and milk production in primiparous dairy cows during the transition period. Cows (n= 40) were fed on a standard diet from wk 3 prepartum to wk 6 postpartum. Cows received either a lipogenic or glucogenic supplement during the transition period and were randomly assigned to 1 of 4 treatments as follows: control, calcium salts of soybean fatty acids - Megalac-E (100g prepartum and 250g postpartum), toasted soybean (400g prepartum and 800g postpartum) and propylene glycol (300 ml pre and postpartum). Diets were isocaloric (net energy basis). Blood samples were taken from each animal 10 and 5 days prior to the expected calving date and at days 7, 14, 21, 28, 35 and 42 postpartum. Milk production was recorded on days 10, 20, 30 and 40 postpartum. Data was analysed using a completely randomized split-plot design. Milk yield did not differ among treatments (P=0.067). Fat corrected milk (FCM 3.5%) was higher for cows supplemented with Megalac-E and propylene glycol (P = 0.047) (24.90±4.85 and 24.58±5.32, respectively). Insulin and glucose concentrations were significantly higher in the Megalac-E treatment (P<0.05). The lowest insulin and glucose concentrations were observed on cows supplemented with propylene glycol. Non esterified fatty acid concentration (NEFA) were lower for all treatments compared to control group (0.24±0.16 mmol/l) (P<0.05). Plasma IGF-I concentration was higher for treatments receiving the lipogenic supplements (P<0.05). Overall, results suggest that calcium salts of soybean fatty acids reduced the dramatic metabolic and endocrine changes characteristic of the transition period, reflected in improved energy balance which could be associated with better reproductive and productive performance.

**Key Words:** insulin, glucose, IGF-I

**M298 Corn endosperm type influences nutrient digestibility in lactating dairy cows.** J. C. Lopes*[1], R. D. Shaver[1], P. C. Hoffman[1], M. S. Akins[1], S. J. Bertics[1], H. Gencoglu[2], and J. G. Coors[3], [1]*Department of Dairy Science, University of Wisconsin*, *Madison*, [2]*Department of Animal & Nutritional Sciences, Faculty of Veterinary Medicine, Uludag University*, *Bursa, Turkey*, [3]*Department of Agronomy, University of Wisconsin*, *Madison*.

The objective of this study was to evaluate the effect of corn endosperm type on ruminal fermentation parameters and total-tract nutrient digestibility in lactating dairy cows. Near-isogenic variants of an Oh43xW64A normal dent endosperm hybrid carrying floury-2 or opaque-2 alleles were grown in spatial isolation in field plots and harvested as dry shelled corn. Six ruminally-cannulated multiparous Holstein cows (67 ± 9 DIM and 638 ± 5 kg BW at trial initiation) were randomly assigned to a replicated 3 x 3 Latin Square design with 14-d periods; the first 11 d of each period were for diet adaptation followed by 3 d of sampling and data collection. Treatment diets that contained dry rolled vitreous-, floury-, or opaque-endosperm corn (33% of DM), alfalfa silage (55% of DM) and protein-mineral-vitamin supplement (12% of DM) were fed as TMR. The percentage vitreous endosperm was zero for the modified corn endosperm types and 64 ± 7% for the vitreous corn. The prolamin protein content of the modified corn endosperm types was only 30% of the content found in vitreous corn. Degree of starch access and in vitro ruminal starch digestibility measurements were 32% and 42% greater on average, respectively, for the modified corn endosperm types than vitreous corn. Dry matter intake and milk yield averaged 25.6 kg/d and 45.2 kg/d, respectively, for the trial and were unaffected (P > 0.10) by treatment. Dry matter and starch disappearances after 8-hr ruminal in situ incubations were 24 and 32%-units on average greater (P < 0.001), respectively, for the modified corn endosperm types than vitreous corn. Ruminal pH and acetate molar % were lesser (P < 0.01), propionate molar % was greater (P < 0.01), and acetate:propionate ratio was lesser (P < 0.01) for the modified corn endosperm types. Total-tract starch digestibility was 6.3%-units, on average, greater (P < 0.001) for diets containing modified corn endosperm types than the vitreous corn. Apparent total-tract NDF digestibility was lesser (P < 0.001) for diets containing the modified corn endosperm types. Corn endosperm type influenced starch and fiber digestibility in lactating dairy cows.

**Key Words:** corn, digestibility, starch

**M299 Performance of dairy cows fed extruded or hydrated and ensiled mature corn grain.** L. L. Bitencourt[1], S. Siécola Júnior[1], L. Q. Melo[1], N. M. Lopes[1], V. A. Silveira[1], I. R. Rios[1], J. R. M. Silva[2], R. A. N. Pereira[3], and M. N. Pereira*[1], [1]*Universidade Federal de Lavras*, *Lavras, MG, Brazil*, [2]*Centro Federal de Educação Tecnológica*, *Januária, MG, Brazil*, [3]*Better Nature Research Center*, *Ijaci, MG, Brazil*.

In Brazil, all the corn used in concentrate feedstuffs has low digestibility, flint type endosperm. We evaluated the response to corn extrusion or to proteolysis and fermentation induced by ensiling. A mature flint hybrid (AG 2040) was ground and had its moisture content reconstituted to 43.7% at ensiling, for 358 days. Other treatments were the same hybrid ground in the same feed mill (11.8% moisture) or extruded (9.7% moisture). Fifteen Holsteins (160±84 DIM, three primiparous) received the treatments in five, 3x3 Latin Squares, with 21-day periods. Diet composition averaged (% of DM): Corn silage (41.5); soybean meal (21.5), citrus pulp (17.5), CP (17.3), NDF (30.9), and NFC (39.1). The content of corn was 16.7% of DM for ensiled, 17.4 for ground, and 17.7 for extruded. Two contrasts were evaluated: 1) Ensiled *vs.* Ground. 2) Extruded *vs.* Ground. Extruded corn depressed the secretions of milk energy and fat and DMI. Eating behavior was not as responsive as DMI. The content of MUN, the daily secretion of urinary allantoin, and the total tract apparent digestibility of the non-NDF OM (Non-NDF OM D) were not a supportive reasoning for the tendency of increased milk protein content with extruded corn. There was a tendency for increased OM

digestibility (OM D) with ensiled corn. Extrusion or ensiling of mature flint corn resulted in a slight increase in the conversion of feed into milk. *Funded by Fapemig and D'Vita Rações.*

**Table 1.**

| | Ensiled | Ground | Extruded | Contrast 1 | Contrast 2 |
|---|---|---|---|---|---|
| DMI | 21.6 | 22.1 | 21.1 | .30 | .05 |
| Milk yield | 33.4 | 33.0 | 33.5 | .46 | .20 |
| Fat yield | .767 | .785 | .704 | .53 | .01 |
| Protein yield | .795 | .782 | .783 | .36 | .94 |
| Fat % | 2.99 | 3.11 | 2.82 | .25 | .01 |
| Protein % | 3.15 | 3.11 | 3.17 | .29 | .09 |
| Milk energy (Mcal/d) | 21.05 | 21.02 | 19.92 | .95 | .03 |
| MUN (mg/d) | 15.2 | 16.2 | 17.1 | .06 | .06 |
| Milk yield/DMI | 1.55 | 1.49 | 1.55 | .12 | .15 |
| Ingestion time (min/d) | 287 | 284 | 277 | .81 | .43 |
| First meal time (min) | 29.5 | 33.2 | 30.0 | .92 | .51 |
| Non-NDF OM D (%) | 86.1 | 85.7 | 86.4 | .66 | .48 |
| OM D (%) | 76.6 | 74.4 | 76.1 | .10 | .19 |
| Urinary allantoin (mmoles/d) | 76.1 | 66.6 | 67.6 | .23 | .89 |

**Key Words:** corn processing, feed efficiency, flint corn

**M300 Effect of starch infusion site on glucose rate of appearance (Ra) and digestibility of starch and nitrogen in dairy cows.** F. Hassanat*, H. Lapierre, and D. R. Ouellet, *Dairy and Swine R&D Centre, Agriculture and Agri-Food Canada*, *Sherbrooke, QC, Canada*.

The objective of this study was to determine the impact of changing the site of starch infusion on whole body glucose Ra and digestibility of starch and N in dairy cows. Four mid-lactation cows fitted with ruminal, duodenal, and ileal cannulae were used in a Youden square design with 3 periods of 28 d. Starch was infused at a total continuous rate of 1.80 kg d-1 either in the Rumen, the Omasum, or equally partitioned in both sites (Mix), with dietary starch intake averaging 2.59±0.21 kg $d^{-1}$. On d 20, cows were infused in a jugular vein with D[6,6-$d_2$]glucose (12.0 mmol $h^{-1}$) for 180 min. Blood samples were obtained from the contralateral jugular vein every 20 min from 100 to 180 min of the infusion to determine glucose enrichment. Duodenal, ileal, and fecal samples were collected on days 27 and 28 (n=4 per d) to determine starch and N disappearance from the different digestive tract segments. Milk yield (29.1±5.7 kg $d^{-1}$) and composition were not affected by treatment. Similarly, plasma glucose concentration (3.99±0.23) was unaffected by treatments, although glucose Ra numerically increased with Mix treatment. Total tract starch digestibility was lowest when starch was infused only in the omasum as a result of decreased absolute disappearance in the rumen. This led to an increase in small intestine and hindgut disappearance, resulting in a reduced fecal pH. Infusing starch in the omasum decreased N total tract and hindgut digestibility. Indeed, negative N digestibility indicates N release across the hindgut with Omasum treatment. These results indicate that supplying additional starch in the small intestine did not increase plasma glucose concentration or Ra but that undigested starch was highly fermented in the hindgut. This will impact true N digestibility and endogenous N secretions lost by lactating cows.

**Table 1. Impact of starch infusion on glucose Ra and digestibility of starch and nitrogen**

| | Rumen | Mix | Omasum | SEM | *P* value |
|---|---|---|---|---|---|
| Total tract starch digestibility, % | 99.5$^{a}$ | 99.1$^{a}$ | 94.1$^{b}$ | 1.1 | 0.04 |
| Starch disappearance g $day^{-1}$ | | | | | |
| - rumen | 3735$^{a}$ | 2895$^{b}$ | 2395$^{c}$ | 152 | 0.01 |
| - small intestine | 380$^{b}$ | 984$^{ab}$ | 1213$^{a}$ | 146 | 0.02 |
| - hindgut | 157$^{b}$ | 369$^{ab}$ | 813$^{a}$ | 120 | 0.02 |
| Nitrogen digestibility, % | | | | | |
| - total tract | 73.4$^{a}$ | 69.5$^{ab}$ | 67.4$^{b}$ | 1.4 | 0.05 |
| - hindgut | 15.9$^{a}$ | 0.5$^{b}$ | -7.0$^{b}$ | 2.9 | 0.04 |
| Glucose Ra, mmol $hr^{-1}$ | 666 | 732 | 700 | 35 | 0.43 |
| Fecal pH | 6.72$^{a}$ | 6.72$^{a}$ | 5.32$^{b}$ | 0.11 | 0.01 |

$^{LS}$means with different letters differ significantly

**Key Words:** starch infusion, glucose kinetics, digestibility

**M301 The effects of different sources of nonstructural carbohydrates and addition of full fat roasted canola seed on milk production and composition in lactating cows.** M. Sari, A. A. Naserian*, R. Valizadeh, and S. Salari, *Ferdowsi Universuty of Mashhad*, *Mashhad, Khorasan Razavi, Iran*.

The objective of this study was to examine the effects of modifying the dietary sources of neutral detergent-soluble carbohydrate (NDSC), addition of full fat roasted canola seed (RCS), and possible interactions on milk production and composition in lactating cows. Twelve lactating Holstein cows (BW=596±29 kg, DIM=85±14) were used in a 4×4 Latin squares design. Treatments were in a 2×2 factorial arrangement, and periods were 21 d. The first 15 d were used for diet adaptation. The cows were fed individually four experimental diets as TMR ad libitum. The diets, which contain 15% barley (S), 5% barley and 10% dried citrus pulp (NDSF), with or without addition of 6% ground RCS at the expense of corn and canola meal, were formulated to meet NRC (2001) recommendations. Diets contained 17.5% CP and constant forage to concentrate ratio (45:55). Cows were milked three times a day and samples were collected at each milking over the last three d of each period. Data were analyzed using the GLM procedure of SAS (2001). Daily DMI of cows fed S diets was higher than cows fed NDSF diets (24 vs 22.9 kg/d). Cows receiving diet S produced more milk (33.8 vs 32 kg/d) and animals fed NDSF diet produced milk with greater concentration of milk fat (3.20 vs 2.95%). Consequently, NFC source did not affect 3.5% FCM yield. An interaction (P<0.05) between NDSC sources and RCS was observed for ECM because of higher production with NDSF than S diet when RCS was added (P<0.05). Cows fed the S diet had an increased milk protein content than when fed the NDSF diet (P<0.05), and the inclusion of RCS reduced the protein content (P<0.01). Milk lactose was similar among treatments. Production efficiency was not affected by NDSC sources, but was higher for cows fed RCS diets. Results indicated that citrus pulp and barley differed in the type and quantity of metabolizable nutrients they provided. Adding RCS had positive effects on milk production and production efficiency using citrus pulp.

**Key Words:** dairy cows, nonstructural carbohydrates, roasted canola seed

**M302 Supplemental starch in postpartum dairy cow diets 1. Effect on productivity.** B. L. Dyck*[1], L. Doepel[1], and M. G. Colazo[2], [1]*University of Alberta, Edmonton, AB, Canada*, [2]*Alberta Agriculture and Rural Development, Edmonton, AB, Canada*.

The objective of this study was to determine if increasing the level of dietary starch in postpartum Holstein cows would improve their productivity and metabolic status. One of 3 diets was fed from calving until 70 DIM. The diets contained 45% concentrate and 10% alfalfa hay. In addition, the low starch diet (n=14) contained 45% barley silage, the medium starch diet (n=13) contained 45% alfalfa silage, and the high starch diet (n=13) contained 41% barley silage plus 4% corn starch. Resulting starch levels were 23.5%, 25.3% and 26.9%, respectively. Body weight and BCS were measured biweekly, and DMI and milk yield recorded daily. Milk samples were obtained weekly; blood samples were taken at calving and then biweekly until 70 DIM for analysis of glucose, insulin, IGF-1, BHBA and NEFA. Data were statistically analyzed using the repeated measures option in Proc Mixed of SAS. Treatment had no effect on DMI, milk yield, milk fat or protein content ($P$>0.10) but cows fed the low starch diet had significantly higher levels of milk urea nitrogen compared to the other diets ($P$=0.001). Cows fed the high starch diet had higher BW than the other two treatments ($P$=0.04) but BCS was not different among treatments. Blood metabolite concentrations were unaffected by treatment. These results indicate that starch supplementation to well-balanced diets has little impact on productivity or metabolic status of postpartum cows.

**Table 1. Effect of dietary starch level on production and metabolic parameters**

| | Starch level | | | |
|---|---|---|---|---|
| | Low | Medium | High | SEM |
| DMI, kg/d | 18.0 | 19.3 | 18.5 | 0.8 |
| Milk yield, kg/d | 34.9 | 34.4 | 35.5 | 1.6 |
| Milk fat, % | 3.81 | 3.67 | 3.6 | 0.2 |
| Milk protein, % | 3.01 | 2.94 | 3.06 | 0.1 |
| MUN, mg/dl | 16.25[a] | 13.56[b] | 13.35[b] | 0.7 |
| Glucose, mg/dl | 59.4 | 60.0 | 60.2 | 1.1 |
| Insulin, µIU/ml | 6.33 | 6.61 | 7.77 | 0.7 |
| IGF-1, ng/ml | 55.7 | 52.1 | 56.0 | 3.2 |
| NEFA, mEq/L | 294.2 | 349.8 | 268.0 | 30.4 |
| BHBA, mg/dl | 8.38 | 7.84 | 8.54 | 0.3 |
| BW, kg | 560[b] | 557[b] | 594[a] | 14 |

Means within a row with different superscripts differ significantly ($P$<0.05)

**Key Words:** starch supplementation, postpartum cows, metabolic status

**M303 Effect of dietary protein level on rumen fermentation, digestibility, and nitrogen losses in dairy cows.** M. Agle*[1], A. N. Hristov[2], S. Zaman[1], and C. Schneider[1], [1]*University of Idaho, Moscow*, [2]*Pennsylvania State University, University Park*.

The objective of this experiment was to investigate the effect of dietary CP level on rumen fermentation, digestibility, and performance of lactating dairy cows. The experiment was a replicated Latin square design with 2-wk adaptation periods. Three diets varying in CP concentration were tested (CP, % of DM): 15.3 (HighCP), 13.3 (LowCP), and 12.8% (ExLowCP). RDP and RUP concentrations (estimated based on NRC, 2001) were: 10.2 and 5.1, 8.3 and 5.0, and 7.0 and 5.8% of DM, respectively. Estimated RDP supply exceeded requirements in HighCP and was deficient in LowCP and ExLowCP. In all diets, metabolizable protein intake met or exceeded the requirements of the cows. Concentration of $NE_L$ was similar among diets (1.51 Mcal/kg). Dry matter intake was numerically lower ($P$ = 0.15) for LowCP and ExLowCP, compared with HighCP. Milk (30 to 32 kg/d), 4% FCM, and energy-corrected milk yields and milk fat and protein concentrations and yields were not affected by treatment ($P$ = 0.16 to 0.88). Rumen pH and ammonia concentration were decreased ($P$ = 0.02 and <0.001, respectively) by both LowCP and ExLowCP, compared with HighCP. Ruminal VFA concentrations were not affected ($P$ = 0.53 to 0.89) by diet. Plasma urea and MUN concentrations and absolute and relative (as proportion of N intake) urinary N losses were lower ($P$ < 0.001 to 0.02) for LowCP and ExLowCP, compared with HighCP. Compared with HighCP, total urinary and fecal N losses were reduced ($P$ = 0.05) by ExLowCP, but not by LowCP. As proportion of N intake, urinary and fecal N losses were not different ($P$ = 0.97) among diets. Urinary allantoin excretion and estimated microbial protein outflow from the rumen were numerically reduced ($P$ = 0.15) by LowCP and ExLowCP, compared with HighCP. Total tract apparent digestibility of DM, OM, N, and NDF was not affected ($P$ = 0.07 to 0.35) by diet. Decreasing dietary CP within the range tested in this experiment did not affect cow productivity, but resulted in decreased urinary N losses.

**Key Words:** dietary protein, nitrogen losses, dairy cow

**M304 Effect of dietary crude protein concentration on production and nitrogen balance of lactating dairy cows.** T. Sun, Z.-J. Cao*, Y.-X. Dong, H.-T. Zhang, and S.-L. Li, *College of Animal Science and Technology, China Agricultural University, Beijing, China*.

Four ruminally cannulated multiparous cows averaging 73±13 days in milk (DIM) were used in a 4 × 4 Latin square design to determine the effects of different dietary crude protein levels on ruminal fermentation and production performance . Each experimental period was 21-d. The diets contained [dry matter (DM) basis] 20% alfalfa hay, 20% corn silage, 10% Chinese wildrye, and 50% concentrate mainly from corn, bran, rapeseed meal and soybean meal. Four experimental diets were formulated to contain similar concentrations of ADF, NDF, net energy of lactation (NEL), but with different crude protein (CP), rumen-degradable protein (RDP) and rumen-undegradable protein (RUP) levels: 12.8%CP, 7.5% RDP, 5.3% RUP (LD-LU diet); 14.6% CP, 9.2% RDP, 5.4% RUP (HD-LU diet); 14.0% CP, 7.6% RDP, 6.4% RUP (LD-HU diet) and 15.6% CP, 9.2% RDP, 6.4% RUP (HD-HU diet). Diets were fed as a total mixed ration. Results showed that milk yield tended to be increased in the cows given the HD-LU, LD-HU and HD-HU diets compared with that in the cows fed on the LD-LU diet. Higher DM intake (DMI) and milk yield/DMI were also detected in the cows fed the HD-HU diet and HD-LU diet compared with that for the LD-LU diet. Milk fat content was higher for the HD diets than the LD diets. Milk protein percentages and yields were not significantly affected by the level of RUP or RDP. Ruminal NH3 concentration was higher for the HD diets than the LD diets, and isovalerate concentration tended to be increased by increasing dietary CP level. Apparent nutrient digestibility and estimated bacterial CP synthesis were similar for all treatments. Blood and milk urea-N were increased by feeding higher dietary CP; Increasing dietary CP from 12.8% (diet LD-LU) to 15.6% (diet HD-HU) increased urinary N excretion by 37g/d and reduced apparent N efficiency (milk protein N/N intake) by 4.4 percentage units. Under the conditions of this trial, milk production and composition were improved by feeding higher

CP diets. But higher CP diets depressed N efficiency and increased N loss in urine.

**Key Words:** nitrogen metabolism, nitrogen excretion

**M305 Use of milk urea nitrogen (MUN) to improve dairy farm management.** M. Nourozi*[1,2], A. Heravi Moussavi[1], and M. Abazari[2], [1]*Department of Animal Science , Ferdowsi University of Mashhad, Mashhad, Iran*, [2]*Department of Animal Science, Khorasan Razavi Agricultural and Natural Resources Research Center, Torogh, Mashhad, Iran*.

The hypothesis of this study was that providing farmers with information regarding their herd′s milk urea nitrogen (MUN) would result in more accurate feed management, lower feed cost and a change in MUN toward target values. Milk samples of high (38 ±3 kg/d) and low (24 ±3 kg/d) yield dairy cows' and bulk tanks in thirty dairy milk producers were tested for MUN and crude protein each month for six months from the last month of spring until the last month of fall. Farms were divided into two equal groups. The first group of dairy Farms (n=15) was provided with the results of their MUN analysis each month along with interpretive information. Nitrogen intake, urinary and fecal N and efficiency of feed N utilization were estimated from survey data and milk analysis for each herd. Data were analyzed by using a mixed model (PROC MIXED, SAS Inst. Inc., Cary, NC) for a completely randomized design with repeated measures. For milk samples of high yielding dairy cows and bulk tanks, average MUN and efficiency of feed N utilization for the first group of farms (13.29±0.58, 12.31±0.59 mg/dl and 30.37±0.78, 27.10±0.58 percent; respectively) were lower and higher respectively than second group (18.52±0.66, 16.09±0.67 mg/dl and 27.30±0.92, 24.47±0.66 percent; respectively)($p<0.05$). The average MUN across all farms in the study increased in late summer, but the increase was significantly lower for farms receiving MUN results than other farms ($p<0.05$). Conversely, the average feed N utilization was decreased for all farms in late summer. Again, average feed N utilization calculated from high-producing cow or bulk tank samples for that time period was significantly higher for the informed group than the second ($p<0.05$). The farms which received MUN results were able to improve their management and significantly lower feed cost (45$/cow/305DIM, $p<0.05$) compared to farms without that information.

**Key Words:** dairy farms, milk urea nitrogen, fecal N

**M306 Varying ruminally degradable protein concentrations in the lactating dairy cow diets maintains rumen fiber digestion and outflow of nutrients.** J. Cyriac*[1], A. G. Rius[1], J. A. D. R. N. Appuhamy[1], R. E. Pearson[1], J. H. Herbein[1], K. F. Knowlton[1], J. L. Firkins[2], and M. D. Hanigan[1], [1]*Virginia Polytechnic Institute and State University, Blacksburg*, [2]*The Ohio State University, Columbus*.

This study investigated the effects of feeding diets containing varying concentrations of ruminally degradable protein (RDP) on ruminal metabolism and outflow of nutrients in lactating dairy cows. Seven ruminally cannulated, Holstein cows were used in a replicated Latin square design with 4 periods and 4 treatments. Period length was 21 d. Cows were randomly assigned to one of four diets formulated to contain 11.3, 10.1, 8.8 or 7.6% RDP with constant ruminally undegradable protein (7.1% of DM). All diets contained 47.5% forage and 52.5% concentrate on a DM basis. Ruminal outflow of nutrients was determined from omasal samples using a double marker system with Co-EDTA and Yb-labeled forage as markers. Data were analyzed using the Proc Mixed procedure of SAS. The varying levels of RDP in the diet did not significantly alter DM (17.8, 18.7, 17.4 and 17.0 kg/d) or fiber intakes. Protein intake decreased linearly (3.19 to 2.56 kg/d; $P<.05$) with decreasing concentrations of RDP in the diet. Milk yield (32.7, 34.5, 34.1 and 29.8 kg/d) and milk composition were similar across treatments excepting milk urea-N which decreased linearly with decreasing dietary RDP ($P<.05$). Ruminal $NH_3$ concentrations also decreased linearly with decreasing concentrations of RDP in the diet (from 10.8 to 3.96 mg/dL; $P<0.0001$). Ruminal outflow of DM declined from 14.9 to 13.3 kg/d for the high and low RDP diets, respectively indicating a trend for a linear reduction in flow ($P=0.08$). Ruminal outflow of acid detergent and neutral detergent fiber was not significantly affected by treatment. Ruminal digestibility coefficients for acid detergent and neutral detergent fiber were not significantly affected by treatment. However ruminal outflow of N decreased linearly with decreasing concentrations of RDP in the diet ($P<.01$). Feeding dietary RDP below NRC requirements did not appear to compromise fiber digestion, but it did result in a reduction in total N leaving the rumen. Reduced N availability to the animal could compromise milk production if maintained for longer periods of time.

**Key Words:** ruminally degradable protein, fiber digestion, nutrient flow

**M307 Effect of NPN source and dietary fermentable carbohydrate composition on fermentation, digestion, and N flow in rumen-simulating fermenters.** G. A. Harrison*, M. D. Meyer, and K. A. Dawson, *Alltech Biotechnology, Nicholasville, KY*.

Replacing corn with byproducts in dairy diets can improve feeding economics but lower fermentable carbohydrate (fCHO) could negatively impact ruminal function. Effects on ruminal performance of total replacement of corn grain with byproducts while holding fCHO constant were investigated in single-flow rumen-simulating fermenter cultures. Cultures were fed diets formulated in CPM (version 3.08) based on either corn and byproducts with equal fCHO. In addition, three NPN sources (urea-U, Optigen®-O, urea-Optigen) were utilized with each providing equal NPN (equivalent of 114 g NPN at 22.7 kg DM intake). Six cultures were used in a 2 X 3 factorial design with 6 dietary treatments and 4 experimental runs. Cultures were fed 12.5 g as fed of experimental diets twice daily for 6 days. Samples were collected from all cultures prior to morning feeding during the last 3 days of the experiment for fermentation analysis. Effluent composite samples from each fermenter were used for DM and NDF disappearance determination. Nitrogen flow measures were estimated by using purine to N ratios for effluent DM and bacteria. Data were analyzed for effects of dietary treatment using GLM procedure of SAS. Orthogonal contrasts were used to determine effects of NPN and fCHO. NPN source had no effect on culture pH, digestion, or N flow ($P>0.10$). Bacterial N% was greater ($P<0.05$) in cultures fed the U-O combination. Corn-fed cultures had higher molar proportions of butyrate ($P<0.001$) and isoacids ($P<0.0001$) and lower acetate ($P<0.0001$). Bacterial N yield was similar between cultures fed different fCHO formulations ($P>0.10$). Under dietary conditions utilized in this experiment, no effects of source were noted when NPN was provided by urea, Optigen, or a combination of the two. When diets were balanced for equivalent fCHO, total replacement of corn grain with byproducts resulted in differences in fermentation patterns but did not affect digestibility or nitrogen partitioning.

**Key Words:** non-protein nitrogen, Optigen, fermentable carbohydrate

**M308 Effects of different levels of rumen degradable protein on rumen and plasma parameters in midlactation Holstein cows.** H. Rafiee*, *Aboureihan Campus, Tehran University*, *Tehran, Iran*.

To evaluate the effect of different level of rumen degradable protein (RDP) on rumen and blood parameters, nine multiparous mid-lactation Holstein cows averaging 171 ± 17 days in milk and 24.1 ± 3.3 Kg of milk/d were assigned into a triplicate 3 × 3 Latin square design. Each period was 21-d (14 d of adaptation and 7 d for sampling). Diets were iso-energetic and based on a barley concentrate and 45:55 forage:concentrate ratio. Forage part consists of corn silage and alfalfa hay. The RDP % and crude protein level of diets were respectively: 1) 9.8 and 14.3 2) 10.8 and 15.3 3) 11.8 and 16.3. Urea was supplemented to increase of RDP. Rumen fluid was taken from each cow approximately 3 h after morning feeding and Blood samples were collected from the coccygeal vein of cows at approximately 2 h post feeding. Data were analyzed using PROC MIXED of SAS. Cow within square was the term of the RANDOM statement. Values reported as least squares means. Treatments had no effect on ruminal pH. However, increasing RDP from 9.8 to 11.8% resulted in significant linear increases ($P < 0.01$) in rumen concentration of ammonia (9.95, 12.50, and 14.11 mg/dL, respectively; SEM = 0.70). Treatments had no effect on glucose and total protein of plasma. But, increasing protein from 14.3 to 16.3% resulted in significant linear increases ($P < 0.01$) in plasma urea nitrogen (15.20, 16.86, and 17.76 mg/dL, respectively; SEM = 0.41). With increasing dietary RDP both plasma urea nitrogen (PUN) and rumen ammonia concentration increased significantly. The rumen ammonia not incorporated into microbial protein, is absorbed across the rumen wall and converted to urea in the liver and diffused in blood for secretion in milk, excretion in urine or recycling to the rumen through saliva. Because of the rumen fluid collection was difficult and harmful for animal, this result suggested that we can use form PUN for prediction rumen ammonia level.

**Key Words:** rumen degradable protein, rumen ammonia, plasma urea nitrogen

**M309 Partial replacement of soybean meal by protected urea effects on milk yield and composition.** V. L. Souza[1], D. F. F. Silva[1], P. R. B. Piekarski[1], C. P. Jesus[2], M. N. Pereira[3], and R. Almeida*[1], [1]*Universidade Federal do Paraná*, *Curitiba, PR, Brazil*, [2]*Colégio Agrícola Olegário Macedo*, *Castro, PR, Brazil*, [3]*Universidade Federal de Lavras*, *Lavras, MG, Brazil*.

The objective of this trial was to evaluate the effects of partial replacement of soybean meal by protected urea - Optigen® II (Alltech Inc., Nicholasville, USA) on milk yield and composition. Thirty-four lactating Holstein cows with 128 ± 60 days in milk and yield of 41.6 ± 6.7 kg/day at the beginning of the trial, were blocked by milk production, lactation number and days in milk. Variables measured before the application of treatments were used as covariate in the statistical model. Response variables were measured on days 30 and 60 of the comparison period. Data was analyzed with the mixed procedure of SAS with a model containing the continuous effect of the covariate and the fixed effects of block, period, treatment, day and the interaction of day and treatment. The mean square of cow nested within treatment was used as the error term to test the treatment effect. Treatments were isoenergetic and isonitrogenous diets with 1.66 ELL and 18.4% of CP (dry matter basis), described as: T1= 0.0% Optigen + 11.4% soybean meal; T2= 0.4% Optigen + 9.0% soybean meal, respectively. The composition of the control TMR was (% of DM): corn silage (30.2), grass haylage (11.7), concentrate mix (30.3), whole cottonseed (5.4), soybean meal (11.4), soybean hulls (8.4), Megalac-E® (0.8) and mineral mix (2.0). There was no detectable treatment effect on the daily production of milk and solids, MUN and SCC, although milk yield was numerically greater with Optigen® II. The slow release urea diet reduced milk fat and total solids content. The partial replacement of soybean meal by protected urea did not induce lower performance of lactating cows.

**Table 1.**

| | Soybean meal | | Optigen® II | | | |
|---|---|---|---|---|---|---|
| | Day-30 | Day-60 | Day-30 | Day-60 | SEM | *P* Treat |
| Milk yield (kg/d) | 36.9 | 36.4 | 39.4 | 38.1 | 1.11 | 0.16 |
| Fat content (%) | 3.16 | 2.99 | 2.87 | 2.71 | 0.07 | <0.01 |
| Fat yield (kg/d) | 1.14 | 1.13 | 1.12 | 1.04 | 0.04 | 0.24 |
| Protein content (%) | 3.12 | 2.82 | 3.03 | 2.79 | 0.04 | 0.21 |
| Protein yield (kg/d) | 1.14 | 1.07 | 1.19 | 1.08 | 0.03 | 0.51 |
| Lactose content (%) | 4.58 | 4.72 | 4.64 | 4.80 | 0.03 | 0.08 |
| Lactose yield (kg/d) | 1.70 | 1.73 | 1.83 | 1.84 | 0.06 | 0.13 |
| Total solids content (%) | 11.74 | 11.43 | 11.43 | 11.21 | 0.10 | 0.04 |
| Total solids yield (kg/d) | 4.31 | 4.19 | 4.47 | 4.25 | 0.12 | 0.46 |
| Milk energy (Mcal/d) | 23.6 | 22.6 | 24.1 | 22.5 | 0.62 | 0.77 |

**Key Words:** non-protein nitrogen, Optigen® II, slow-release urea

**M310 Effect of different ratios of ammonia nitrogen to peptide nitrogen on microbial nitrogen synthesis in dairy cows.** A. Nikkhah*, M. Kazemi Bonchenari, K. Rezayazdi, M. Dehghan, and H. Kohram, *Department of animal Sciences, Faculty of agronomy and animal sciences, University of Tehran*, *Karaj, Iran*.

Some of the studies showed that peptide nitrogen (PN) improves ruminal fermentation and also some studies reported that the minimum ammonia nitrogen (AN) concentration (5 mg/dl) is necessary for optimal microbial nitrogen synthesis (MNS). There is limited document related to the optimum ratio of AN/PN for microbial nitrogen synthesis (MNS). Three ruminally fistulated dairy cows assigned in a Latin square design (21d periods; 14d for adaptation and 7d for sample collection) and fed a similar basal diet. Sodium Caseinate was infused directly in to rumen before morning diet portion as peptide nitrogen source (PNS) (0, 50 and 100 gr/d as treatments 1, 2 and 3 respectively). Ruminal fluid samples were collected at 0, 1.5, 3, 4.5 and 6 hours after feeding. Separated aliquots were preserved for later analyses of PN and AN. Daily urinary output was measured by urine collection bag and total purine derivatives (uric acid + allantoin) were measured. MNS was estimated from total purine derivatives. Data were analyzed using Proc Mixed in SAS with the following model; $Y_{jklm} = \mu + P_j + C_k + T_l + \varepsilon_{jkl} + Z_m + ZT_{ml} + \alpha_{jklm}$ in which components respectively are; dependent variable, overall mean, effect of period j, effect of cow k, effect of treatments l, whole plot error, effect of time m, interaction between time m and treatment l, subplot error. The results are given in table 1. This study confirmed that rumen microbes need some of their N requirements as PN and the highest amount of MNS was for AN/PN ratio of 0.82. It could be concluded that it is necessary to fractionating and identification of rumen microbial requirements to different nitrogen sources (AN, amino acid N and PN).

**Table 1. Effect of PNS infusion in to rumen on microbial nitrogen synthesis**

| | Treatments | | | | P<F |
|---|---|---|---|---|---|
| Item | 1 | 2 | 3 | SE | Treatments |
| NH3-N (mM/L) | 8.11$^{c}$ | 10.6$^{bc}$ | 12.96$^{a}$ | 2.11 | <0.01 |
| Pep-N (mM/L) | 13.93$^{c}$ | 15.12$^{ab}$ | 15.74$^{a}$ | 1.23 | <0.01 |
| AN/PN | 0.58$^{c}$ | 0.70$^{ab}$ | 0.82$^{a}$ | 0.04 | <0.05 |
| Total purine derivatives (mM/d) | 334.4$^{c}$ | 351.3$^{b}$ | 378.4$^{a}$ | 39.1 | <0.01 |
| Microbial nitrogen (g/d) | 222.8$^{c}$ | 238.7$^{b}$ | 257.5$^{a}$ | 25.9 | <0.01 |

$^{a,b,c}$ Least squares means within the same row without a common superscript differ (P < 0.05).

**Key Words:** peptide nitrogen, microbial nitrogen synthesis

**Table 1. Effect of Sodium Caseinate on fiber digestibility and nitrogen efficiency**

| | Treatments | | | | |
|---|---|---|---|---|---|
| Item | 1 | 2 | 3 | SE | P |
| $NH_3$-N mM | 8.11$^{c}$ | 10.6$^{bc}$ | 12.96$^{a}$ | 1.16 | <0.01 |
| Pep-N mM | 13.93$^{c}$ | 15.12$^{ab}$ | 15.74$^{a}$ | 1.2 | <0.01 |
| $NH_3$-N/Pep-N | 0.58$^{c}$ | 0.70$^{ab}$ | 0.82$^{a}$ | 0.04 | <0.01 |
| BUN mg/dl | 12.6$^{bc}$ | 12.9$^{b}$ | 14.3$^{a}$ | 2.1 | <0.01 |
| NDF digestibility % | 45.2$^{b}$ | 46.2$^{ab}$ | 47.3$^{a}$ | 5.12 | <0.05 |
| ADF digestibility % | 48.6$^{bc}$ | 50.1$^{b}$ | 52.5$^{a}$ | 5.8 | <0.01 |

$^{a,b,c}$ Least squares means within the same row without a common superscript differ (P < 0.05)

**Key Words:** peptide nitrogen, fiber digestibility

**M311 Optimum ratio of ammonia nitrogen to peptide nitrogen in ruminal fluid for fiber digestibility and nitrogen utilization efficiency in dairy cows.** M. Kazemi Bonchenari[1], K. Rezayazdi[1], M. Dehghan[1], A. Nikkhah*[1], H. Khalilvandi[1], V. Keshavarz[2], and F. Ghaziani[1], [1]*Department of Animal Sciencs, Faculty of Agronomy and Animal Sciences, University of Tehran, Karaj, Iran*, [2]*Department of Animal Sciences, University of Zanjan, Zanjan, Iran*.

Previous studies showed that increasing peptide nitrogen (Pep-N) concentration in ruminal fluid improve fiber digestibility. Some other studies showed that increased peptide nitrogen concentration decreased ammonia nitrogen ($NH_3$-N) concentration (under 5mg/dl that considered minimum requirement for optimum rumen fermentation) in rumen and therefore fiber digestibility negatively affected by high peptide nitrogen concentration. The optimum ratio of ammonia nitrogen to peptide nitrogen ($NH_3$-N/Pep-N) is not well documented. In this study three ruminally dairy cows was assigned to a Latin square design (21d periods; 14d for adaptation period and 7d for sample collection period) to investigate the optimum ratio of $NH_3$-N/Pep-N for fiber digestibility and nitrogen utilization efficiency. The basal diet was similar and Sodium Caseinate was infused in to rumen as peptide nitrogen source before morning diet (0, 50 and 100 gr/d which considered as treatments 1, 2 and 3 respectively). Pep-N and $NH_3$-N concentrations were measured in ruminal fluid in different hours. Blood urinary nitrogen (BUN) was measured and considered as nitrogen utilization efficiency indicator. Data were analyzed using Proc Mixed in SAS (SAS institute 1999). The results showed that NDF and ADF digestibility significantly increased by Sodium Caseinate infusion and also BUN was increased dramatically by Sodium Caseinate infusion (table 1). Both of the highest fiber digestibility and also the lowest nitrogen utilization efficiency was for the ratio of 0.82. It could be concluded that based on the present study the ratio of 0.70 is the optimum ratio of $NH_3$-N/Pep-N for both traits.

**M312 Effect of whole cottonseed levels on ruminal parameters of dairy cows grazing elephant grass.** J. Cesar Martinez*[1], F. Auguto Portela Santos[2], T. Vinhas Voltolini[2], A. Vaz Pires[2], and C. Maris Machado Brittar[2], [1]*São Paulo State University*, *Jaboticabal, São Paulo, Brazil*, [2]*São Paulo University*, *Piracicaba, São Paulo, Brazil*.

The trial evaluated whole cottonseed (WCS) inclusion on concentrate supplements offered to lactating cows grazing Elephant Grass during the rainy season. Trial was conducted at Animal Sciences Department, USP/ESALQ, Piracicaba/SP-Brazil. 12 multiparous Holstein (543kg LW, 142 DIM at trial beginning) were used on a replicated 4x4 Latin Square design. Data were analyzed by MANOVA and GLM procedures of SAS (2002). Animals were kept on a 4.6ha pasture area divided in 25 0.2ha paddocks fertilized with 80 kg N ha/month. All concentrates had 19% crude protein (CP) and were soybean and ground corn based. WCS was included at 7, 14 and 21% total DM, based on NRC (2001) DMI prediction. The microbial N was analyzed by HPLC, and VFA by photometry. Treatments did not affect ruminal VFA (131.17mM) and ammonia (34.86 mg/dl) concentrations and ruminal pH (6.13). Microbial N flux was affected by WCS14 and WCS21% treatments (P<0.05). Results indicate that WCS can be utilized as a replacement for corn and part of soybean meal on lactating cows rations with no effects on rumen fermentation parameters when used up to 14% of estimated DMI.

**Table 1. Ruminal parameters of dairy cows supplied with differents levels of WCS**

| | | Treatments | | | | |
|---|---|---|---|---|---|---|
| | Corn | 7%WCS | 14%WCS | 21%WCS | Mean | Pr(t) |
| C2[1] | 67 | 69 | 67 | 66 | 68 | 0.8 |
| C3[1] | 41.7 | 43.8 | 37.5 | 35.9 | 40.5 | 0.4 |
| C4[1] | 16.3 | 17.1 | 15.4 | 14.9 | 16.1 | 0.34 |
| iso-C4[1] | 1.3 | 1.4 | 1.4 | 1.4 | 1.4 | 0.59 |
| C5[1] | 2.0 | 2.1 | 2.0 | 1.8 | 2.0 | 0.87 |
| Total VFA[1] | 131 | 136 | 127 | 123 | 131 | 0.35 |
| $NH_3$ , mg/dL | 33.0b | 30.4b | 35.4ab | 40.7a | 34.8 | 0.01 |
| pH | 6.1 | 6.1 | 6.1 | 6.3 | 6.1 | 0.32 |
| Microbial N, gN/day | 201.7a | 191.0a | 175.5b | 169.8b | 182.2 | 0.01 |

Within rows, means followed by different superscripts are significantly different (P < 0.05); [1]mM/mL

**Key Words:** volatile fatty acids, ruminal metabolism, tropical Pastures

**M313 Effect of whole cottonseed levels on performance of dairy cows grazing elephant grass.** J. Cesar Martinez*[1], F. Auguto Portela Santos[2], T. Vinhas Voltolini[2], M. Antonio Penati[2], and A. Mendonça Pedroso[2], [1]*São Paulo State University*, *Jaboticabal, São Paulo, Brazil*, [2]*São Paulo University*, *Piracicaba, São Paulo, Brazil*.

The trial evaluated whole cottonseed (WCS) inclusion on concentrate supplements offered to lactating cows grazing Elephant Grass during the rainy season. Trial was conducted at Animal Sciences Department, USP/ESALQ, Piracicaba/SP-Brazil. Twelve multiparous Holstein (543kg LW, 142 DIM at trial beginning) were used on a replicated 4x4 Latin Square design. Data were analyzed by GLM procedures of SAS (2002). Animals were kept on a 4,6ha pasture area divided in 25 0,2ha paddocks fertilized with 80 kg N ha/month. All concentrates had 19% crude protein (CP) and were soybean and ground corn based. WCS was included at 7, 14 and 21% total DM, based on NRC (2001) DMI prediction. Cows received concentrate according to milk production on a 1:3 basis, fixed at trial beginning, twice daily after each milking. Milk and 3,5% fat corrected milk (FCM) production were lower for WCS21% treatment (P<0,05). Treatments did not affect milk fat, protein, lactose, total solids and urea concentrations (P>0,05) (Table 1). As complement, the live weight (LW) (530 kg), body condition score (BCS) (2,3), plasma urea (49,7mg/dL) and non esterified fatty acids (NEFA) (366,0 meq/L) concentrations were not affected by treatments (P>0,05). Results indicate that WCS can be utilized as a replacement for corn and part of soybean meal on lactating cows rations with no effects on milk production and composition, LBW and blood composition when used up to 14% of estimated DMI.

**Table 1. Milk yield and composition of dairy cows feed with different levels of WCS**

| | | Treatments | | | | |
|---|---|---|---|---|---|---|
| | Corn | 7%WCS | 14%WCS | 21%WCS | Mean | Pr>(t) |
| Milk, kg/day | 17.6a | 17.4a | 16.9a | 15.3b | 16.8 | 0.01 |
| FCM, kg/day | 17.8a | 17.8a | 17.8a | 16.2b | 17.4 | 0.01 |
| Fat, % | 3.5 | 3.6 | 3.8 | 3.8 | 3.7 | 0.27 |
| Fat, kg/day | 0.62 | 0.63 | 0.65 | 0.59 | 0.6 | 0.18 |
| Protein, % | 2.9 | 2.8 | 2.8 | 2.8 | 2.8 | 0.06 |
| Protein, kg/day | 0.51a | 0.49a | 0.47a | 0.43b | 0.48 | 0.01 |
| Urea, mg/dL | 13.4b | 15.0a | 15.9a | 15.8a | 15.05 | 0.01 |

Within rows, means followed by different superscripts are significantly different (P < 0.05)

**Key Words:** supplementation, tropical pastures, milk yield and composition

**M314 Effect of whole cottonseed processing on ruminal degradability of dairy cow grazing elephant grass.** J. Cesar Martinez*[1], F. Auguto Portela Santos[2], T. Vinhas Voltolini[2], and A. Dias Pacheco Júnior[2], [1]*São Paulo State University*, *Jaboticabal, São Paulo, Brazil*, [2]*São Paulo University*, *Piracicaba, São Paulo, Brazil*.

The objective of this trial was to evaluate the degradability of different processing of whole cottonseed (WCS) included on concentrate supplements offered to lactating cows grazing Elephant Grass during the rainy season. The trial was conducted at the Department of Animal Sciences, USP/ESALQ, Piracicaba/SP. Four multiparous Holstein cows (513kg LW, 103 DIM at trial beginning) were used on a 4x4 Latin Square design. Data were analyzed by Ørskov and McDonald (1979) model and GLM procedure of SAS (2002). Animals were kept on a pasture of 4.6ha divided in 25 0.2ha paddocks fertilized with 80 kg N ha/month. All concentrates had 19% crude protein (CP) and were soybean and ground corn based. WCS was included at 7, 14 and 21% total DM, based on NRC (2001) DMI prediction. After each milking, cows were fed concentrate twice daily according to milk production on a 1:3 ratio fixed at the beginning of the trial. The incubation time was 0, 2, 4, 6, 8, 12, 18, 24, 36, 48, 62, 84, 96 and 120 hours. When compared with corn grain concentrate, WCS incubated with linter altered significantly (P<0.05) the ruminal degradation kinetic parameters, lowering both the soluble (22.3 vs 2.38%) and the degradable (76.2 vs 28.63%) fractions. On the other hand, undegradable fraction was higher (66.14 vs 1.56 in corn grain). Degradation rate (4.88 vs 1.6%/h) and effective degradation (63.65 vs 11.93%) were lower for WCS. Ruminal and total retention time were only different for WCS7% treatment and byproduct inclusion did not affect forage ruminal passage. WCS ground at 5 mm had a similar degradation to non processed WCS. However, when it was ground at 2 mm the degradation was similar to that related to corn grain. Thereby, the WCS ground at 2 mm is a source of free lipids, as well as the WCS ground at 5 mm is a source of effective fiber.

**Table 1. Ruminal degradability of dry matter of WCS incubated with different processing**

| | | | Treatments | | | |
|---|---|---|---|---|---|---|
| | Corn | WCS | 2mm | 5mm | Mean | Pr(t) |
| a, % | 22.4a | 2.4c | 2.2c | 30.3a | 13.7 | 0.01 |
| b, % | 76.1a | 31.3c | 68.4cb | 43.2b | 47.4 | 0.01 |
| c, % | 1.5d | 66.1a | 59.3b | 25.8c | 38.7 | 0.01 |
| C, %/h | 4.9a | 1.6c | 1.2c | 3.5b | 2.8 | 0.01 |
| Lag Time, h | 1.5d | 48.3a | 44.4b | 22.7c | 29.5 | 0.01 |
| Kp, %/h | 4.1a | 3.6b | 3.5b | 3.6b | 3.7 | 0.05 |
| Effective Deg.., % | 63.6a | 11.9c | 11.2c | 52.7b | 34.1 | 0.01 |
| Potential Deg., % | 98.1a | 28.9c | 29.4c | 73.4b | 56.7 | 0.01 |

Within rows, means followed by different superscripts are significantly different (P < 0.05)

**Key Words:** ruminal metabolism, tropical pastures, linted whole cottonseed

**M315 Effect of dietary protein on urea concentrations and preovulatory follicle characteristics in dairy cattle.** U. Moallem*[1], R. Blank[2], M. Zachut[1,2], and A. Arieli[2], [1]*ARO*, *Bet Dagan, Israel*, [2]*Faculty of Agriculture*, *Rehovot, Israel*.

The objectives of this study were to determine the effects of dietary crude protein (CP) on metabolic parameters and preovulatory follicle characteristics in dairy cattle. Four intact and 4 rumen cannulated Holstein non-lactating cows were assigned to 3 treatments in 4 periods (39 d each). Diets consisted of 66% wheat straw with different mixtures of corn grain and soybean meal resulted with 6% (LP), 13% (MP) and 20% (HP) of CP. Individual intake was recorded daily, blood samples were taken twice a week and rumen fluids from the cannulated cows were taken twice in each period. Ten d after commencement of treatments the estrus cycle was synchronized with two $PGF_{2\alpha}$ (PG) injections 13d apart. Fourteen d after observed behavior estrus cows received PG injection and 40 h afterward follicular fluid (FF) from follicles ≥ 7 mm were aspirated. The feed intake of the LP group was lower than in other groups. Rumen ammonia concentrations were lower in the LP than in other groups. Blood urea concentrations were higher in the HP and lower

in the LP as compared to other groups. Similarly, the urea concentrations in FF were higher in HP and lower in the LP group than in other groups (17.1, 33.3 and 50.5 mg/dL for LP, MP and HP, respectively). The estradiol ($E_2$) concentrations in FF tended to be higher in the HP than in the MP and the progesterone ($P_4$) concentrations were higher in the HP than in both other groups. No differences in the $E_2/P_4$ ratio or in $E_2$ and $P_4$ contents in FF were observed among groups. The correlation between FF urea concentrations and $P_4$ content in follicles was significant (r=0.45). In conclusion, high dietary protein increased the urea concentrations in plasma and in a very similar manner in FF. However, the higher urea concentrations in FF as a consequence of high dietary CP was not detrimental to steroidogenesis as reflected by $E_2$ concentration and content, and $E_2/P_4$ ratio in preovulatory follicles.

**Key Words:** dietary protein, follicular fluid

**M316 Relationship between milk odd and branched-chain fatty acids and duodenal flow of microbial protein.** L. Wang, J. Q. Wang*, D. P. Bu, Khas-Erdene, and S. Y. Luan, *State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences*, *Beijing , P. R. China*.

Effects of refined docosahexaenoic acid (DHA) and refined free linoleic acid (LA) or their combination on odd and branched-chain fatty acids (OBCFA) profiles of milk was studied. A correlation was developed between OBCFA secretion and duodenal flow of microbial protein in lactating dairy cows. Four multiparous Holstein-Friesian dairy cows with ruminal and duodenal cannulas averaging 139±10 DIM were used in a 4 × 4 Latin square design experiment. There were 4 periods of 3 wk each. Cows were fed 65% forage and 35% concentrate diet. In addition to the basal diet cows received either 0 (Control) or 0.5% DHA, 2.73% LA, and 0.5% DHA + 2.3% LA in combination. Fat was added by replacing grain in the diet. Milk and duodenal digesta samples were collected on 17, 18, 19 d and 16, 17, 18 d, respectively. Data were analyzed statistically by using PROC MIXED of SAS. Feed intake was not affected by fat supplement compared to control. Milk yields were 18.6, 17.2, 18.4, and 17.0 kg/d for control, DHA, LA, and DHA+LA treatments, respectively and were not affected by fat supplementation P > 0.05. Milk fat and protein percentages were the same in fat supplemented and control treatment. Compared with control, the concentrations of linear odd-chain fatty acids (C15:0 and C17:0) in milk were reduced by LA+DHA treatment, however, fat supplements had no significant (P>0.05) effects on the concentrations of *iso*-fatty acids or *anteiso*-fatty acids. No strong linear relationship (R=0.166) between milk secretion OBCFA and duodenal flow of cytosine were observed in this study, suggesting that milk OBCFA could not be used as a marker for duodenal flow of microbial protein when cows offered diets with fat supplements.

**Key Words:** odd and branched-chain fatty acids, microbial protein, dairy cows

**M317 Comparison of optimal lysine and methionine concentrations in metabolizable protein estimated by the NRC (2001), CPM-Dairy (v.3.0.10) and AMTS.Cattle (v.2.1.1) models.** N. Whitehouse*[1], C. Schwab[1], T. Tylutki[2], D. Luchini[3], and B. Sloan[3], [1]*University of New Hampshire, Durham*, [2]*Integrated Solutions for Sustainable Agriculture, Cortland, NY*, [3]*Adisseo, Atlanta, GA*.

Optimizing concentrations of lysine (Lys) and methionine (Met) in metabolizable protein (MP) is considered to be the first step to maximizing milk and milk component production and efficiency of use of MP by the dairy cow (NRC, 2001). Three commonly used programs to balance for amino acids (AA) are NRC (2001), CPM (v.3.0.10) and AMTS (v.2.1.1). Each program differs in the approach and assumptions taken to estimate AA supply. However, the indirect dose-response approach proposed by Rulquin et al. (1993) for identifying the optimal concentrations of Lys and Met in MP, and subsequently adopted by NRC (2001), can be extended to all models that predict passage of AA to the small intestine. In NRC (2001), this approach showed that maximal content of protein in milk was attained with 7.24% Lys and 2.38% Met in total MP; corresponding values for maximal yield of milk protein were 7.08 and 2.35%. However, a reevaluation of the requirement values for Lys and Met in MP using the NRC (2001) model (see companion abstract) yielded the requirements shown in the table. The objective of this work was to use the same trial dataset used to elaborate the NRC (2001) Lys and Met recommendations to estimate the comparable requirement values for Lys and Met in MP for both CPM (v.3.0.10) and AMTS (v.2.1.1). The dose-response plots were generated as described in NRC (2001). The resulting breakpoint estimates for the required concentrations of Lys and Met in MP for maximal content and yield of milk protein for the two models are shown in the table.

**Table 1. Breakpoint estimates for required concentrations of Lys and Met in MP for maximal content and yield of milk protein**

| | | Requirement for maximal milk protein content | Lys:Met ratio | Requirement for maximal milk protein yield | Lys:Met ratio |
|---|---|---|---|---|---|
| NRC | Lys | 6.80 | 2.97:1 | 7.10 | 2.82:1 |
| | Met | 2.29 | | 2.52 | |
| CPM | Lys | 7.46 | 2.90:1 | 7.51 | 3.00:1 |
| | Met | 2.57 | | 2.50 | |
| AMTS | Lys | 6.68 | 2.78:1 | 6.74 | 2.92:1 |
| | Met | 2.40 | | 2.31 | |

**Key Words:** lactating cows, lysine, methionine

**M318 Reevaluation of the breakpoint estimates for the NRC (2001) required concentrations of lysine and methionine in metabolizable protein for maximal content and yield of milk protein.** C. Schwab*[1], N. Whitehouse[1], D. Luchini[2], and B. Sloan[2], [1]*University of New Hampshire, Durham*, [2]*Adisseo, Atlanta, GA*.

The 7th Revised Edition of the Nutrient Requirements of Dairy Cattle (NRC, 2001) presents dose-response plots that relate changes in milk protein content (Figure 5-12) and yield (Figure 5-13) to predicted concentrations of Met and Lys in metabolizable protein (MP). A rectilinear model was used to describe the dose-response relationships. The breakpoint estimates for the required concentrations of Lys and Met in MP for maximal content of milk protein were 7.24 and 2.38%, respectively; corresponding values for maximal yield of milk protein were 7.08 and 2.35%. Because the amino acid submodel had to be developed before the final version of the NRC model was available, a beta version of the model was used to predict the concentrations of Lys and Met in MP in the studies that were used in developing the dose-response plots. The objective of this study was to repeat all of the steps, as cited in NRC (2001) (p. 82-85), to generate new dose-response plots, using the final version of the model to predict concentrations of Lys and Met in MP. The resulting breakpoint estimates for the required concentrations of Lys and Met in MP for maximal content of milk protein were 6.80 and 2.29%, respectively. Both values are lower than the corresponding values

of 7.24 and 2.38% reported in NRC (2001). The breakpoint estimates for the required concentrations of Lys and Met in MP for maximal yield of milk protein were 7.10 and 2.52%, respectively. The breakpoint estimate for optimal Lys concentration in MP for maximal protein yield (7.10%) is similar to that reported in NRC (2001) (7.08%), whereas the estimate for optimal Met concentration in MP for maximal protein yield (2.52%) is higher than that reported in NRC (2001) (2.35%). The reasons for these differences in required concentrations of Lys and Met in MP for maximal milk protein production are not clear. However, it is suggested that these new values be used as the reference values when using the NRC (2001) model to optimize Lys and Met concentrations in MP for lactating cows.

**Key Words:** lactating cows, lysine, methionine

**M319 Rumen microbial population shifts in dairy cattle experimentally induced with subacute ruminal acidosis (SARA).** E. Khafipour*, S. Li, J. C. Plaizier, and D. O. Krause, *Department of Animal Science, University of Manitoba, Winnipeg, MB, Canada*.

We analyzed rumen microbial composition using terminal restriction fragment length polymorphism (T-RFLP) and real-time PCR in dairy cows experimentally induced with SARA. Our objective was to examine the relationships between microbial population shifts and free lipopolysaccharide (LPS) in the rumen, and the inflammatory response that occur during SARA. SARA was induced by addition of grain pellets or alfalfa pellets into diets in two separate experiments. Cows were clustered based on the severity of SARA with a discriminant multivariate analysis using animals as independent variable and duration of rumen pH < 5.6, LPS, and serum haptoglobin as the dependent variables. SARA cows were separated into mild and severe grain-induced, and alfalfa pellet-induced groups. Severe grain-induced SARA was separated from the mild group based on the greater content of LPS (179,762 vs. 100,175 EU/mL, $P < 0.05$) and inflammatory markers (608 vs. 343.3 μg/mL, $P < 0.05$). The alfalfa pellet-induced SARA was clustered separately ($P < 0.05$) for its high content of LPS (169,266 EU/mL) but no inflammatory response (21 μg/mL). Differences in microbial populations were detected using LSD multiple comparison test ($P < 0.05$). T-RFLP analysis indicated that the predominant shift during SARA was the decline in gram-negative *Bacteroidetes*. However, the proportion of *Bacteroidetes* was greater in alfalfa pellet-induced SARA compared to mild or severe grain-induced SARA (35.4% vs. 26.0% and 16.6% respectively). This shift was also evident from the real-time data for *Prevotella albensis*, *Prevotella brevis*, and *Prevotella ruminicola*, which are members of the *Bacteroidetes*. The real-time data also indicated that severe grain-induced SARA was dominated by *Streptococcus bovis* and *Escherichia coli*, whereas mild grain-induced SARA was dominated by *Megasphaera elsdenii*, and alfalfa pellet-induced SARA was dominated by *P. albensis*. Using discriminant analysis, the severity of SARA and inflammation was correlated with the abundance of *E. coli* and not with LPS in the rumen. We thus suspect that a subset of *E. coli* might be the putative pathogenic trigger for SARA.

**Key Words:** SARA, T-RFLP, real-time PCR

**M320 Molecular population analysis of *Escherichia coli* associated with subacute ruminal acidosis (SARA) in dairy cattle.** E. Khafipour*, J. C. Plaizier, and D. O. Krause, *Department of Animal Science, University of Manitoba, Winnipeg, MB, Canada*.

Grain-induced SARA in dairy cattle is associated with increase in inflammatory markers in peripheral blood, such as haptoglobin and serum amyloid-A. However, induction of SARA with alfalfa pellets does not activate an inflammatory response. We hypothesize that diet induced changes to gut *Escherichia coli* populations may be partially responsible for the difference in inflammatory response. To test this hypothesis, low pH conditions were induced by addition of grain pellets or alfalfa pellets into diets in two separate experiments. Rumen fluid samples were collected before and 6 h after feeding and were cultured on chromogenic medium with or without acid shock for *E. coli* and total coliform numeration. Acid shock was performed by reducing the pH of rumen fluid samples to 5.2 with acetate for 2 h, or 2 with HCl for 1 h. From each non-shocked sample, five *E. coli* strains were isolated, typed with repetitive sequence-based PCR DNA fingerprinting, and genotyped using the A, B1, B2, and D phylotypes. Fingerprints were assessed for presence or absence of 23 bands ranging from 420 to 3,300 bp and were subjected to discriminant and cluster analysis using JMP and MEGA software, respectively. The numbers of *E. coli* in alfalfa pellet-induced SARA were extremely lower than in grain-induced SARA so that acid shock could not perform on these samples. The quantity of acid resistant populations remained stable during grain-induced SARA, while a higher degree of variation occurred in the control animals. Cluster analysis indicated that isolates from controls, and alfalfa pellet-induced SARA tended to cluster together. In contrast, grain-induced SARA *E. coli* were members of distinctly separate *E. coli* populations. There was also a shift from A to B1 type *E. coli* during grain-induced SARA. However, no significant difference was observed among *E. coli* when alfalfa-pellet induced SARA was compared to controls. These results suggest that only grain-induced SARA alters *E. coli* population in the rumen and suggests that *E. coli* from high grain diets may possess virulence factors that could be involved in the inflammatory responses observed with SARA.

**Key Words:** SARA, fingerprinting, *E. coli*

**M321 Factors affecting lipopolysaccharide (LPS) in the feces of dairy cows and its relationship to sub-acute ruminal acidosis (SARA).** J. C. Plaizier*, D. O. Krause, and S. Li, *University of Manitoba, Winnipeg, MB, Canada*.

The concentration of LPS in feces may be an indictor of SARA in dairy cows. We conducted a survey of 10 dairy farms in Manitoba to investigate factors that could affect this concentration. Each farm was visited once to collect blood samples, milk samples and fecal grab samples of 15 early and peak lactation Holstein cows with an average of 62 ± 38 days in milk (DIM, mean ± SD) and 15 late lactation Holstein cows with an average of 272 ± 67 DIM. Daily milk yield, days in milk, and parity were recorded. Blood was analyzed for the acute phase proteins serum amyloid A (SAA) and haptoglobin (Hp). Milk was analyzed for fat, protein, and somatic cell counts (SCC). Feces were analyzed for free LPS. Diets were analyzed for NDF. Farms were divided into 5 farms with low dietary NDF (< 34% DM) and 5 farms high dietary NDF (> 34% of DM) Data were analyzed in SAS PROC Mixed with a model consisting of the fecal LPS concentration as a dependent variable, level of dietary NDF as a fixed factor, farm within NDF level as a random factor, and parity, days in milk, SCC, and the concentrations on milk fat, milk protein, HP and SAA as independent variables. Farm within level of NDF affected fecal LPS. Days in milk, milk protein, milk fat, SAA and Hp did not affect fecal LPS. An increase in parity was associated with a reduction in fecal LPS ($P = 0.04$), whereas as an increase in daily milk yield was associated with an increase fecal LPS ($P = 0.05$).

Farms with low dietary NDF had higher fecal LPS than farms with a high dietary NDF (27,087 vs. 14,672 EU/ml). Fecal LPS tended to increase with increasing SCC (P=0.09). Results suggest that low dietary NDF, which is a risk factor for subacute ruminal acidosis (SARA), is associated with an increase in LPS in the feces. This could be due to increased fermentation and lysis of gram negative bacteria in the large intestine that accompany SARA. Hence, the measurement of LPS in feces could aid in the diagnosis of SARA. The relationship between SCC and fecal LPS suggests a relationship between SARA and mastitis, the mechanism of which warrants future investigation.

**Key Words:** LPS endotoxin, acute phase proteins, ruminal acidosis

**M322 Estimation of herd level risk of subacute ruminal acidosis on four commercial dairies on the Priority P-One Program.** K. Schneider*[1], D. Mertz[2], K. Mertz[2], and R. Breunig[1], [1]*Prority IAC, Manitowoc, WI*, [2]*Agtech Products, Inc., Waukesha, WI*.

Subacute ruminal acidosis (SARA) can often be found in well-managed dairy herds. To estimate herd risk for SARA on dairy farms, Oetzel (2004) recommended testing rumen fluid from 12 cows per group for incidence of ruminal acidosis (pH≤5.5). Priority IAC has developed a program (the P-One Program) which combines ration balancing with feeding Direct-fed microbials containing a lactate and a glucose utilizer to optimize ruminal function and manage risk of SARA. The objective of this study was to estimate risk of SARA in herds on the P-One Program. Four independent dairy herds with 1000 to 4800 lactating cows on the P-One Program were selected for this study. On average, the rations contained 29.4% starch (SD=3.8), 43.5% NFC (SD=2.6), and 16.0% CP (SD=0.4) on a DM basis, and fed to achieve 25.5 kg (SD=3.11) DMI. Rumen fluid samples were collected at 4, 7 and 10 hours post-feeding via oro-ruminal probe from two groups of cows in each herd, 12 cows in early lactation (ELAC, 27 DIM) and 12 cows in peak lactation (PLAC, 92 DIM). Rumen fluid pH was measured on farm using a portable pH probe. Group risk was calculated as the proportion of the sample population with a threshold pH≤5.5. Groups with ≤ 8.3% of the sample population at or below the threshold were not considered at risk. Groups with 16.6% to 33.3% of population at or below the threshold were considered borderline risk, and groups with sample populations >33.3% were positive for SARA. Analysis of variance was performed comparing sample times to determine significant rumen pH fluctuations. Mean pH across all groups for all herds at times 4, 7, and 10 hours was 6.7 (SD=0.3) and not found to be significantly different (P=0.9371). The proportion of cows declining to the threshold limit in both the ELAC and PLAC sample populations in all herds was 0 of 12 cows. The herds on the P-One Program in this study showed no clinical signs of ruminal acidosis and based upon the sample populations, not at risk for SARA.

**Key Words:** acidosis, direct-fed microbials, P-one program

**M323 Use of magnesium exchanged natural zeolite as a source of ruminal buffer additive for lactating dairy cows.** C. M. Dschaak*[1], J.-S. Eun[1], A. J. Young[1], and S. Peterson[2], [1]*Utah State University, Logan*, [2]*Zeotech Corporation, Fort Worth, TX*.

A lactating dairy trial was conducted in a completely randomized design to determine the influence of a developmental ruminal buffer product containing magnesium exchanged zeolite (Zeotech Corporation, Fort Worth, TX) on ruminal fermentation and lactational performance in dairy cows over 12 wk. Experimental TMR diet consisted of 37% alfalfa hay, 19% corn silage, 14% of corn grain, and 30% concentrate mix, and it was fed ad libitum. Thirty primiparous and multiparous lactating Holstein cows (52 ± 23 DIM) were assigned to one of 3 dietary treatments with 10 cows in each treatment: control (TMR diet without ruminal buffer), TMR diet with 1.4% sodium bicarbonate (SBD), and TMR diet with 1.4% zeolite product (ZLD). Ruminal fluid was collected from all animals using an oro-ruminal (Geishauser) probe. Intake of DM, milk yield, and milk composition values were reduced to weekly means before data analysis. Data were analyzed using the MIXED procedure of SAS with repeated measures. Intake of DM (average 26.5 kg/d) did not differ across treatments, and milk yield was similar among the 3 treatments (average 40.7 kg/d). Dairy efficiency (4% FCM/DMI) was not affected by dietary treatments. Milk fat concentration did not differ among treatments, whereas milk protein concentration tended to be higher for the ZLD than the control and the SBD (P = 0.15). Although feeding the ZLD resulted in the tendency of increased milk protein concentration, feed N efficiency for milk N did not differ among the 3 treatments. In addition, milk urea N concentration was not influenced by feeding the ZLD. Ruminal pH increased (P = 0.04) by feeding the ZLD compared to the control (6.42 vs. 6.61), but it resulted in a similar pH from cows fed the SBD. Concentration of ammonia-N did not differ among treatments. Feeding ZLD tended to decrease (P = 0.14) total VFA production compared to the control and the SBD, whereas molar proportions of acetate and propionate were not affected by the treatments. The zeolite product used in this study would replace sodium bicarbonate as a ruminal buffer additive cost-effectively in lactating dairy diet.

**Key Words:** zeolite, sodium bicarbonate, lactating dairy cows

**M324 Dietary cation-anion difference with calcium supplementation: Effects on metabolites and health of Holstein periparturient cows.** W. X. Wu*[1] and J.-X. Liu[2], [1]*College of Animal Science, Guizhou University, Guiyang, China*, [2]*Institue of Dairy Science, Zhejiang University, Hangzhou, China*.

Dietary cation-anion difference (DCAD) have been drawing much attention in dairy cow nutrition. In this study the effects of varying DCAD with calcium (Ca) supplementation on fluid acid-base balance, blood mineral profiles, health status, and subsequent lactational performance were evaluated in 3 blocks of 12 Holstein cows d 20 prepartum. Animals were randomly allocated to 1 of 3 treatments with DCAD levels at +150 (HD), -100 (LD), and -100 plus Ca (30g/d, LDCA). Urine was sampled 3 d prior ot calving, and blood at d-14, -7 and -3. Feeding of the two LD diets induced lower (P < 0.05) urinary and blood pH compared with the diet HD, with no significant difference (P > 0.05) for the two LD diets. There was a significant correlation between DCAD and urinary pH (P < 0.0001) and blood pH (P < 0.01). Serum Ca level was higher (P < 0.05) in cows fed diet LD than those on diets HD and LDCA, with little difference between diets HD and LDCA (P > 0.05). Cows fed diet LDCA had greater (P < 0.05) serum Mg concentration relative to those on diet LD. Diet LD resulted in higher (P < 0.05) serum P value relative to diet HD. The lowest serum Ca levels were observed for all cows near calving. No milk fever and displaced abomasum occurred within all cows. Hypocalcemia cases were higher (P < 0.05) for cows fed diet HD (33%) compared with those on diet LD (0%). Diet LDCA reduced (P < 0.05) retained placenta incidence (8%) over diet HD (50%). The DMI, milk yield and compositions were unaffected (P > 0.05) by dietary treatments. It may be suggested from the above-mentioned results that dietary inclusion of Ca (30 g/d) into DCAD -100 induced metabolic

acidosis. However, it did not improve blood calcium homeostasis as expected.

**Key Words:** dietary cation-anion difference, calcium supplementation, dairy cows

**M325 Influence of subclinical hypocalcemia on post-partum disease incidence in dairy cows.** W. G. Chamberlin*, J. R. Middleton, and J. N. Spain, *University of Missouri*, *Columbia*.

The purpose of this study was to evaluate the effects of calcium status at calving on incidence of common post-partum diseases in Holstein cows. It was hypothesized that cows with subclinical hypocalcemia at calving would have greater frequency of several common post-partum diseases. Forty-four multiparous Holstein cows were assigned to one of two groups 1) hypocalcemic (n=37; ionized calcium [iCa] ï‚£ 1.0 mmol/L) or 2) normocalcemic (n=7; iCa > 1.0 mmol/L) based on whole blood iCa concentrations on the day of calving. Milk samples were collected from all cattle for measurement of somatic cell count (SCC), solid-not-fat, protein, and fat concentration on the third day after calving, and again on 7d, 14d, 21d, 28d, and 35d postpartum. Occurrence of common post-partum diseases such as retained fetal membranes, clinical mastitis, post-partum metritis, clinical ketosis, and displacement of the abomasum were recorded by farm personnel in consultation with herd health veterinarians. On day 21, LSMean (± SE) linear SCC differed between groups with hypocalcemic cows having a linear SCC score (3.52 ± 0.24) that was higher than normocalcemic cows (2.12 ± 0.6; P=0.03). Milk protein concentration also differed between groups on day 7. Hypocalcemic cows had a lower LSMeans (± SE) concentration of milk protein (3.46 ± 0.03) compared to normocalcemic cows (3.65 ± 0.09; P=0.04). Between day 0 and day 14, there was a significant difference in body condition score (BCS) loss between groups. Hypocalcemic cows had a greater loss of BCS (mean ± SD BCS loss of 0.47 ± 0.16) than normocalcemic cows (mean ± SD BCS loss of 0.25 ± 0.0; Pï€¼0.001). Similarly, there was a trend toward greater incidence of clinical ketosis for hypocalcemic cows versus normocalcemic cows. Hypocalcemic cows had an incidence of 49% clinical ketosis, whereas normocalcemic cows had an occurrence of 14% (P=0.08). The results show a relationship between hypocalcemia at calving and increased linear SCC, BCS loss, and increased incidence of clinical ketosis.

**Key Words:** hypocalcemia, periparturient cows, diseases

**M326 Effect of β-carotene supply during close-up dry period on ovulation at the first follicular wave postpartum in dairy cows.** C. Kawashima*[1], S. Nagashima[1], Y. Fujihara[1], F. J. Schweigert[2], K. Sawada[3], A. Miyamoto[1], and K. Kida[1], [1]*Obihiro University of Agriculture and Veterinary Medicine*, *Obihiro, Hokkaido, Japan*, [2]*University of Potsdam*, *Potsdam-Rehbrüke, Germany*, [3]*DSM Nutrition Japan K.K.*, *Tokyo, Japan*.

β-Carotene functions in reproductive performance of dairy cows. Our previous study showed that lower plasma β-carotene level during close-up dry period related to anovulation at the first follicular wave postpartum (pp). The aim of our study was to examine the effect of β-carotene supply during close-up dry period on the ovulation at the first follicular wave pp in dairy cows. Two experiments of β-carotene supply at different levels (Exp.1; recommendation, Exp.2; pasture equivalent) were carried out using 10 and 22 cows, respectively. In both experiment, cows were fed a TMR consisting of grass, corn silage and concentrate. During close-up dry period, 5 cows (24.6±2.6 d) in Exp.1 and 12 cows (24.3±1.5 d) in Exp.2 were supplied daily 500mg and 2000mg of β-carotene, respectively. Blood samples were obtained once a week from 4 wk prepartum to 3 wk pp. Data was analyzed by repeated measures ANOVA. In Exp.1, plasma β-carotene levels in control group decreased and reached nadir at 1 d pp (1.5±0.2 mg/l), whereas it did not change in treated group (2.5±0.2 mg/l). Number of ovular cows at the first follicular wave was 4/5 in treated group and 1/5 in control group. In Exp.2, plasma β-carotene levels during close-up dry period was higher in treated group than control group ($p<0.05$, 5.9±0.3 vs. 5.1±0.0 mg/l), and the level of control group in Exp.2 was higher than treated group in Exp.1 ($p<0.001$, 5.1±0.0 vs. 2.8±0.4 mg/l). Number of ovular cows at the first follicular wave pp was 9/12 in treated group and 5/10 in control group. In Exp.1 and 2, plasma retinol levels and metabolic status from glucose, NEFA and metabolic hormones levels in plasma did not differ between both groups. In conclusion, β-carotene supply during close-up dry period may have an effect on the ovulation at the first follicular wave pp in dairy cows. Both doses of β-carotene supply used in the present study appear to stimulate an effect on the occurrence of the first ovulation pp.

**Key Words:** β-carotene, dairy cow, first ovulation

**M327 Effect of prepartum diet on rumen bacterial adaptation to a lactation diet fed to dairy cattle.** S. E. Stebulis*[1], D. M. Stevenson[2], G. J. M Rosa[1], P. J. Weimer[2,1], and R. R. Grummer[1], [1]*University of Wisconsin*, *Madison*, [2]*USDA-ARS US Dairy Forage Research Center*, *Madison, WI*.

The objective was to analyze changes in rumen bacterial populations during the transition period using automated ribosomal intergenic spacer analysis (ARISA) and rumen fermentation parameters. Fourteen ruminally cannulated, pregnant, nonlactating Holstein cows (n=8) and 3/4 Holstein x 1/4 Jersey cows (n=6) were used in a completely randomized design. Each cow was assigned to one of two dietary treatments, moderate grain (MG; NDF=37.6%) or high forage (HF; NDF=45.5%). Starting 45 d prior to expected calving date, all cows (n=14) were fed HF. At 21 d prior to expected calving date, 7 cows were switched to MG and remained on this diet until calving. The remaining cows continued to receive HF until calving. At calving, all cows were fed the same lactation diet. Rumen pH was not different ($P > 0.05$) prepartum (6.72 vs. 6.81 ± 0.06) or postpartum (6.21 vs. 6.15 ± 0.63) between the two groups. Rumen ammonia-nitrogen concentration was significantly increased ($P < 0.05$; 6.5 vs. 5.2 ± 0.28 mg/dL) in cows fed MG compared to cows fed HF prepartum, but was not affected ($P > 0.05$) postpartum. Total ruminal VFA, acetate, propionate, and butyrate concentrations were not different ($P > 0.05$) during prepartum or postpartum periods. Within Domain Bacteria, there were 293 total operational taxonomic units (OTU) detected using ARISA across the three time periods considered: prepartum (wk -4 relative to expected calving), transition (wk -1 and wk 0 relative to expected calving), and postpartum (wk 2 and wk 3 postpartum). Prepartum, 6.8% of OTU were significantly ($P < 0.05$) affected by diet (MG or HF). During the transition period, 9.9% of OTU were significantly ($P < 0.05$) affected by diet. Postpartum, 8.5% of OTU were significantly ($P < 0.05$) affected by prepartum diet. Principal component analysis showed clear separation between prepartum and postpartum ruminal bacterial communities across treatments, but no distinction between MG and HF diets. Overall, the prepartum diet did not affect the postpartum rumen bacterial population based on rumen fermentation patterns and microbial communities characterized by ARISA.

**Key Words:** rumen bacteria, periparturient

**M328 Effect of feeding level on the sorting behavior of lactating dairy cows.** E. K. Miller-Cushon and T. J. DeVries*, *Department of Animal and Poultry Science, University of Guelph, Kemptville Campus, Kemptville, Ontario, Canada*.

It has been suggested that dairy cows sort their TMR more extensively when overfed. The objective of this study was to examine the effects of feeding level on the feed sorting behavior of lactating dairy cows fed a high-production TMR. Six lactating Holstein cows, individually-fed a TMR once daily, were exposed to 2 treatments in a crossover design with 7-d periods. The treatments were: 1) lower feeding level (LFL; target 10% orts), and 2) higher feeding level (HFL; target 20% orts). Dry matter intake (DMI) was monitored daily for each animal. For the final 4 d of each treatment period, fresh feed and orts were sampled for chemical (NDF and starch) and particle size analysis. The particle size separator had three screens (19, 8, 1.18 mm) and a bottom pan, resulting in 4 fractions (long, medium, short, fine). Sorting was calculated as the actual intake of each particle size fraction expressed as a percentage of the predicted intake of that fraction based on TMR particle size distribution and DMI. Sorting values > 100% indicate selection for, while values < 100% indicate sorting against. Analysis revealed no effect of day within treatment period; therefore, data were averaged across each treatment period. Treatment was tested using a general linear mixed model. Actual orts % averaged 11.5% for the LFL and 18.0% for the HFL treatments (SE=0.8; $P<0.001$). When on the HFL, cows sorted for the medium particles to a greater extent than on the LFL (103.0 vs. 101.1%; SE=0.6; $P=0.05$). Further, when on the HFL cows sorted against short particles to a greater extent than on the LFL (95.2 vs. 98.6%; SE=0.9; $P=0.03$). Despite greater sorting on the HFL, the concentration of NDF (29.6%) and starch (27.1%) in the feed consumed was similar between treatments. Given this, and that DMI was greater on the HFL compared to the LFL treatment (29.7 vs. 26.5 kg/d; SE=1.6; $P=0.02$), greater intakes of NDF (8.7 vs. 7.8 kg/d; SE=0.5, $P=0.02$) and starch (8.0 vs. 7.2 kg/d; SE=0.4, $P=0.02$) were also observed on the HFL treatment. The results suggest that, despite increased sorting, increasing the feeding level of lactating dairy cows promotes higher DMI and a balanced intake of nutrients.

**Key Words:** dairy cow, sorting, feeding level

**M329 Relationship of dairy cattle chewing behavior with forage fragility and fiber digestibility.** K. W. Cotanch*[1], H. M. Dann[1], C. S. Ballard[1], C. S. Mooney[1], R. J. Grant[1], T. Eguchi[2], and K. Yagi[2], [1]*William H. Miner Agricultural Research Institute, Chazy, NY*, [2]*Zen-Noh National Federation of Agricultural Cooperative Associations, Tokyo, Japan*.

The physically effective neutral detergent fiber (peNDF) system predicts chewing response of dairy cattle based on feed particle size, but does not consider forage fragility or NDF digestibility. Fragility is defined as change in physical effectiveness factor (pef) following 15 min of ball milling. Four chopped grass hays that differed in 24-h in vitro NDF digestibility and fragility were fed for ad libitum consumption to 16 Holstein dairy heifers (18.5 mo of age; average body weight of 570 kg) in replicated 4×4 Latin squares with 1-wk periods to measure intake and chewing responses. The 4 grasses were: 1) low NDF digestibility (31.4% of NDF), low fragility (46.2%; LL), 2) medium NDF digestibility (43.7%), low fragility (30.0%; ML), 3) high NDF digestibility (54.8%), high fragility (80.7%; HH), and 4) medium NDF digestibility (47.3%), high fragility (63.9%; MH). Forages were processed to similar particle size (average pef of 0.44). Data for intake (measured d 4 to 7) and chewing (measured d 6 to 7) were analyzed with model effects for diet, period, and replicate using MIXED procedure of SAS with heifer within replicate as random effect. The dry matter intake (% of body weight/d) for heifers fed LL, LM, HH, and HM, respectively, was 1.73, 1.81, 2.04, and 1.96 and differed for each grass (SEM = 0.04; $P < 0.05$). The NDF intake (% of body weight/d) was lower ($P < 0.01$) for heifers fed LL compared with those fed ML, HH, and MH (0.98, 1.08, 1.05, 1.05, respectively, SEM = 0.03). When heifers were fed ML versus MH, rumination and eating were similar (84 and 48 min/kg NDF intake, respectively; SEM = 2.0; $P > 0.05$) indicating a greater influence of NDF digestibility than fragility on chewing. However, rumination and eating were greater ($P < 0.01$) for heifers fed LL versus HH (99 and 85; 50 and 45 min/kg NDF intake, respectively; SEM = 2.0). For grasses within the range of NDF digestibility and fragility that we evaluated, fragility was less important than digestibility in eliciting chewing response to similarly sized grass particles.

**Key Words:** physically effective fiber, chewing, dairy cattle

**M330 Parity and its effect on conjugated linoleic acid (CLA) content in confined Holstein cows milk, in the northwest of México.** A. Martínez-Borraz, H. González-Rios, S. Y. Moya-Camarena, J. Hernández, and A. Pinelli-Saavedra*, *Centro de Investigación en Alimentación y Desarrollo, A.C., Hermosillo, Sonora, México*.

Studies in experimental animals with conjugated linoleic acid (CLA) have demonstrated beneficial properties such as anticarcenogenic, antiobesity and immune stimulatory. Milk and dairy products are a significant source of CLA, predominantly *cis*-9, *trans*-11-CLA. Most of the efforts to increase CLA content in milk fat have been focused in manipulation of diet; however, there are other less studied factors that may affect the CLA milk content such as parity. To date, the studies related to this factor are scarce. The aim of this study was to evaluate the effect of parity on the CLA content in confined Holstein cow´s milk in a commercial dairy farm in Northwest México. A total of 120 Holstein dairy cows were divided into groups of parity, including: primiparous (P, 1 parity, n = 42); earlier multiparous (EM, from 2 to 3 parities, n = 48); and late multiparous (LM, from 4 to 6 parities; n = 30). All cows were fed ration (as DM) based on 30.2% alfalfa, 27.6% maize silage and 42.2% concentrate. This ration met the requirements of dairy cows of 505 kg, 120 DIM and milk yield 30 L/d (CP: 15.7% DM; ENI: 1.55 Mcal /kg DM). The final forage-to-concentrate ratio was 58:42. Milk samples were collected from December to February. Pooled milk samples were analyzed for chemical composition and *cis*-9, *trans*-11-CLA content. Quantification of *cis*-9, *trans*-11-CLA was achieved by gas chromatography. All data were analyzed using the PROC MIXED of SAS. The model included Parity as fixed effect, and animal and DIM as covariates. The average CLA content in milk fat was 6.65 mg/g fatty acid. Milk from P cows had higher CLA content (6.43 ± 0.3 mg/g fatty acid, $P < 0.05$) compared to milk from LM cows (5.54 ± 0.3 mg/g fatty acid), but not significantly higher than milk from EM cows (6.17 ± 0.2 mg/g fatty acid, $P > 0.05$). These data shows that parity has an effect on *cis*-9, *trans*-11-CLA content in milk.

**Key Words:** conjugated linoleic acid, milk, parity

**M331 Concentration of mammalian lignan enterolactone in milk of dairy cows fed different levels of flaxseed hulls.** N. Gagnon*, C. Côrtes, C. Benchaar, and H. V. Petit, *Agriculture and Agri-Food Canada, Sherbrooke, QC, Canada*.

Flax lignans are concentrated in the outer fibre-containing layers, resulting in higher concentration of secoisolariciresinol diglucoside (precursor

of enterolactone: EL) in hulls than seeds. The objective of this study was to determine the effect of feeding different levels of flaxseed hulls (FH), a rich source of flax lignans, on milk concentration of the mammalian lignan EL, which has been linked to have potential human health benefits. Twenty lactating Holstein cows were assigned randomly to five different levels of FH in the diet: 0, 5, 10, 15, and 20% (DM basis). The experiment was carried out from wk 18 to 24 of lactation. Feed consumption was recorded daily. Milk samples were obtained weekly from wk 20 to 24 from each cow for two consecutive milkings and pooled separately (AM and PM) on a 5-wk basis for EL analysis using enzyme immunoassay. Concentrations of EL in AM and PM milkings increased (P=0.03) linearly with greater concentrations of FH, but there was no difference (P=0.84) between consecutive milkings. The CV of milk EL concentration for cows fed 0, 5, 10, 15, and 20% of FH was 20.4, 23.5, 36.2, 43.9, and 50.4%, respectively. The inter-variation of cow within the same diet increased with feeding concentration of flax lignan while there was no difference for the level of EL among consecutive milkings. Increasing dietary intake of flax hulls contributes to improving dairy milk composition through increased concentration of the mammalian lignan EL.

**Key Words:** enterolactone, flax hulls, milk

**M332 Weekly excretion of the mammalian lignan enterolactone in milk of dairy cows fed flaxseed meal.** N. Gagnon*, C. Côrtes, and H. V. Petit, *Agriculture and Agri-Food Canada*, *Sherbrooke, QC, Canada*.

Flaxseed meal (FM) is rich in the plant lignan secoisolariciresinol diglucoside (SDG), which is converted to the mammalian lignans enterodiol (ED) and enterolactone (EL) by ruminal microbiota. Feeding FM to dairy cows increases EL concentration linearly in milk but ED is not detected. The objectives of the study were to determine the time required to reach optimal concentrations of EL in the milk of dairy cows fed FM and the time required to return to the baseline level of EL when cows were switched from high to low intake of SDG. A total of 12 multiparous lactating Holstein cows used in a completely randomized design were assigned to one of two dietary treatments: control (CO) diet with no FM and a diet with 20% FM (DM basis). The control group was fed the CO diet for 6 weeks while the other group was fed the FM diet from wk 0 to 3 and was switched to the CO diet from wk 3 to 6. The concentration of SDG was determined in the total mixed diets. Milk samples were taken weekly for EL analysis. Concentrations of SDG and EL were determined by HPLC and enzyme immunoassay, respectively. The percentages of SDG averaged 0.048% and 0.357% (DM basis) in the CO and FM diets, respectively. Milk EL concentration from cows on the CO diet was similar over time and ranged from 116.27 to 179.30 nmol/l. Milk EL concentration increased significantly from wk 0 to 1 for cows on the FM diet (225.05 to 2312.47 nmol/l, P=0.03 according to a Wilcoxon signed rank test). Concentrations of milk EL remained at high levels from wk 1 to 3 (P=0.002 according to Wilcoxon rank-sum test comparing CO to FM cows for each week) before decreasing when the CO diet was reintroduced (from wk 3 to 4; 2167.37 to 264.47 nmol/l, P=0.03 and no statistical difference between the two groups on wk 4 to 6). This study shows that the metabolism of SDG to EL and the transfer of EL to the mammary gland of dairy cows were well established after one week of FM intake and that the baseline level of EL is recovered after one week of FM privation.

**Key Words:** milk, excretion, enterolactone

**M333 Effect of dietary plant antioxidant on milk fatty acids oxidation.** D. Tedesco*, L. Garavaglia, and L. Chiesa, *University of Milan, VSA Dep.*, *Milan, Italy*.

Milk fat with high level of UFA and PUFA has been shown to have several potential health benefits. The aim of the present study was to evaluate the effect of a plant post-processing derivative waste product (SAFEWASTES, EU project n. 513949), administered to dairy cows in preventing/minimizing lipid oxidation in milk. Thirty Holstein cows selected according to parity, milk production (32,57±1.98 kg/d), DIM (115±25) and SCC (<100,000), were divided into two groups. The treated group received 300 g/d of SAFEWASTE products and the control group 300g/d of placebo, mixed with 1 kg of total mixed diet, for 14 d. Milk production was daily recorded and samples collected on 0 and 14 d. Composition (protein, fat, lactose and urea) was assessed on milk samples. Milk samples collected on day 14 were stored at 4°C for four days and analyzed for hexanal accumulated by Head-Space solid-phase microextraction (HS-SPME) in combination with gas chromatography flame ionization detection using a Trace GC Ultra (Thermo-Fisher Scientific; Waltham, MA) Gas Chromatograph coupled to a quadruple Mass Spectrometer Trace DSQ (Thermo-Fisher Scientific; Waltham, MA) and equipped with a RT-wax column. Concentration of hexanal was determined in duplicate as relative area (analyte/I.S.ratio) using the calibration curve. Peak identification was based upon MS spectra in comparison with spectra and retention time of standards. Statistical analysis was performed using PROC MIXED of SAS. Treatment did not affect milk production and composition (P>0.05). Milk was assessed for the antioxidant quality by milk hexanal content, a secondary lipid peroxidation product from the oxidation of milk fatty acids. A significative lower level was observed in milk stored at 4°C for 4 days after milking, in the treated group. The exanal concentration was 14.27 ng/mL in the control group and 7.92 ng/mL in the treated group (SEM 1.097; P < 0.01). This result suggests an antioxidant effect on milk fatty acids oxidation and an extension of milk shelf life.

**Key Words:** milk, fatty acids oxidation, exanal

**M334 Performance and ruminal fermentation parameters of lactating dairy cows during hot environment.** J. P. Wang[1,2], J. Q. Wang*[2], D. P. Bu[2], F. D. Li[1], X. K. Huo[2], T. J. Guo[2], H. Y. Wei[2], and L. Y. Zhou[2], [1]*Gansu Agricultural University*, *Lanzhou, Gansu, China*, [2]*Chinese Academy of Agricultural Sciences*, *Beijing, China*.

Influence of temperature and heat index (THI) on milk yield and rumen parameters was studied in high and medium producing dairy cows during mid- and late-lactation. Forty eight cows were assigned to high production (40.4 ± 4.7 kg/d), mid production (30.5 ± 1.7 kg/d), primiparous, multiparous, mid-stage (136 ± 17 DIM) or late-stage (205 ± 3.4 DIM) groups with a factorial design. There were 6 cows in each group. The experiment was conducted for 11 weeks from June 15 to September 2, 2008. All cows were offered 45% forage and 55% grain diet three times a day. All data was analyzed using the PROC MIXED model of SAS (2004). Cow was the random effect, and milk yield, parity, and stage were the fixed effects. Cows were housed in a close-side barn with tie stalls and cooled with a combination of sprinklers and fans. Cows were milked three times a day. Milk samples were collected weekly from 3 consecutive milkings and analyzed for composition. At the end of the experiment, rumen fluid sample was collected from each cow at 5h post-feeding using stomach-tube. During the experiment, average temperature and THI were 27.9°C, 30.7°C and 27.6°C, and 79.5, 82.1 and 79.2 at 7:00h, 14:00h, and 22:00h, respectively. Milk yield reduced faster for high than medium milk producing cows (0.96 vs. 0.61 kg/

wk, P<0.01), primiparous than multiparous cows (0.95 vs. 0.62 kg/wk, P<0.01) and late than mid-lactation dairy cows (0.82 vs. 0.74 kg/wk, P=0.26). Milk protein contents were higher in high producing cows than mid producing cows (3.1% vs. 2.9%, P<0.01). Milk fat content did not differ among treatments. The ruminal fermentation parameters were the same among treatments except the ratio of acetate to propionate (3.26 vs. 2.54, P<0.01) was higher for mid than high milk producing dairy cows. Results from this study suggests that production of high producing and younger dairy cows is affected more than medium producing or older cows when exposed to 79-82 THI and fed similar diets.

**Key Words:** cows, hot weather, performance

## Ruminant Nutrition: Forages

**M335 Efficiency of different chemicals in deactivation of phenolic compounds in Sainfoin (*Onobrychis viciifolia Scop.*).** H. Khalilvandi-Behroozyar, M. Dehghan-Banadaki*, and K. RezaYazdi, *Research Center of Excellence for Improving Sheep Carcass Quality and Quantity, Animal Science Department, University of Tehran, Karaj, Tehran, I.R. Iran*.

Sainfoin (*Onobrychis viciifolia*) is a member of the Fabaceae family (Leguminosae), tannin rich legume. Reports about condensed tannin content of sainfoin have very wide range from 25 to 100g/kg DM. No reports about effects of tannin destructive or binding matter on tannin deactivation of sainfoin were found. This study was conducted to determine (TP), total tannins (TT) and condensed tannins (CT) content of sainfoin. Samples of forage (from effects of different chemicals on total phenolics different regions of 30 bale) were chopped 3-5 cm length, and then treated with solutions of $KMnO_4$ (0.03 M), NaOH (0.05 M), sodium bicarbonate (0.1 M), wood ash (180 g/L) and water with forage to reagent volume ratio of 1:4 (W/V). 5% solution of PEG (6000 MW) was sprayed to forage with 1:1 ratio. Treatment with urea (20g/ 100 ml/1 kg of DM) was done using adhesive rubber to create anaerobic conditions for 1 week. All of above treatments were carried out in triplicates, in 25°C temperature, for 20 min, with hand shaking. Treated forages were then exposed to 40°C temperature in a forced air oven, for 48 hour. All forage samples were ground to pass 0.5 mm screen size (Wiley mill). Determinations of TP, TT and CT content of treated and control forages were done tenth time, using Folin Ciocalteu reagent, polyvinyl polypyrrolidone (PVPP) and Butanol-HCl reagent, respectively. For preparation of plant extracts 200 mg of dried (0.5 mm ground) plant material is taken in a glass beaker of approximately 25 ml capacity. Ten ml of aqueous acetone (70%) is added and the beaker is suspended in an ultrasonic water bath (Branson 3210) and subjected to ultrasonic treatment for 30 min at room temperature. Data were analyzed using by SAS 9.1, using GLM procedure with complete random design ($P \leq 0.05$). TP, TT and CTs content of sainfoin was 39.4±0.6, 38.5±1 and 21.3±0.4 g/kg of DM. Results (table 1) showed that PEG and water were more effective in CT deactivation. Although all of the chemicals reduced CT levels far from 90%, but TP and TT deactivation ranged from 54.12 to 75.40 and from 55.06 to 76.57 percent, respectively. In both cases, water was the best.

**Tabble 1. Reduction of TP, TT and CT of treated sainfoin.**

| | Reduction of TP (%) | Reduction of TT (%) | Reduction of CT (%) | S.E.M |
|---|---|---|---|---|
| Water | 75.40$^{a}$ | 76.57$^{a}$ | 92.06$^{cd}$ | 0.12 |
| Urea | 60.47$^{d}$ | 59.46$^{d}$ | 92.53$^{c}$ | 0.28 |
| KMnO4 | 71.82$^{b}$ | 70.44$^{b}$ | 92.41$^{c}$ | 0.08 |
| PEG | 66.39$^{c}$ | 67.09$^{c}$ | 98.57$^{a}$ | 0.19 |
| Wood ash | 57.41$^{e}$ | 59.57$^{d}$ | 91.61$^{d}$ | 0.14 |
| NaOH | 54.12$^{f}$ | 55.11$^{e}$ | 93.79$^{b}$ | 0.19 |
| Sodium bicarbonate | 54.89$^{f}$ | 55.06$^{e}$ | 91.84$^{d}$ | 0.22 |

**Key Words:** sainfoin, condenced tannin, PEG

**M336 The effect of high sugar grass on nitrogen and methane output in cattle: A modeling approach.** J. L. Ellis*[1], A. Bannink[2], J. Dijkstra[3], A. J. Parsons[4], S. Rasmussen[4], G. R. Edwards[5], E. Kebreab[6], and J. France[1], [1]*Centre for Nutrition Modelling, Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada*, [2]*Animal Sciences Group, Division Animal Production, Wageningen University and Research Centre, Lelystad, The Netherlands*, [3]*Animal Nutrition Group, Wageningen Institute of Animal Sciences, Wageningen University, Wageningen, Then Netherlands*, [4]*AgResearch, Palmerston North, New Zealand*, [5]*Lincoln University, Lincoln, New Zealand*, [6]*Department of Animal Science, University of Manitoba, Winnipeg, MB, Canada*.

The potential of high sugar grass varieties to reduce nitrogen (N) excretion of pasture fed cattle has received considerable attention. It is the purpose of this work to (1) evaluate the prediction of N excretion within a dynamic mechanistic dairy cattle model, (2) evaluate the effect of high sugar grasses on N excretion across multiple studies, and (3) evaluate the effect of high sugar grasses on enteric methane production. The database consisted of 4 published studies for which high sugar grasses were being evaluated for their effect on N excretion. Root mean square prediction error (RMSPE, % observed mean) for urine N, fecal N and milk N (all g/d) were 23.7%, 22.3% and 11.4%, respectively, with the majority of error coming from regression slope deviation and random sources. Proc GLM in SAS was used to analyse the results with study as a fixed effect, and showed that total N excretion was negatively related to water soluble carbohydrate (WSC) content of the diet ($P = 0.006$) and the WSC:CP ratio ($P < 0.001$). Predicted methane production (% GE intake) was significantly affected by treatment, and was positively related to WSC content of the diet ($P < 0.001$), negatively related to N content of the diet ($P < 0.001$) and positively related to the WSC:CP ratio ($P = 0.001$). Results show that the model predicts N excretion adequately, that total N excretion, particularly urine N, is reduced with high sugar grasses, but that this mitigation strategy may also increase methane production (% GE intake).

**Key Words:** sugar grass, nitrogen, methane

**M337 Lipolysis and biohydrogenation of forage species at vegetative and reproductive stages of growth.** A. Cabiddu[1], M. R. F. Lee*[2], L. Salis[1], N. D. Scollan[1], and M. L. Sullivan[3], [1]*AGRIS, Sardinia, Italy*, [2]*Aberystwyth University, Wales, UK*, [3]*USDA-DFRC, Madison, WI*.

Fresh forage is known to increase polyunsaturated fatty acids (PUFA) in ruminant products. Besides forage′s proportionally high levels of PUFA, other factors such as plant secondary metabolites may play a role. In particular polyphenol oxidase (PPO) production of phenol bound protein (PBP) has been shown to reduce ruminal lipolysis. This study investigated the effect of forage species on ruminal lipolysis and C18:3 biohydrogenation (C18:3-Bio). Common vetch (CV); Crimson clover (CC), red clover (RC+) and PPO1 gene silenced red clover (RC-) were harvested, at reproductive (R) and vegetative (V) growth stages, freeze-