**M431 Prediction of residual feed intake in beef heifers by infrared thermography.** J. J. Colyn*[1], A. L. Schaefer[1], J. A. Basarab[2], E. K. Okine[3], T. Liu[1], K. L. Robertson[2], and S. L. Scott[4], [1]*Agriculture and Agrifood Canada, Lacombe Research Centre*, *Lacombe, AB, Canada*, [2]*Alberta Agriculture*, *Lacombe, AB, Canada*, [3]*Department AFNS, University of Alberta*, *Edmonton, AB, Canada*, [4]*Agriculture and Agrifood Canada*, *Brandon, MB, Canada*.

The cow herd is estimated to require 65–75% of the total energy required for beef production. Measurements of residual feed intake (RFI) to improve feed efficiency in beef breeding programs have mainly focused on replacement bulls but potential for improvements exist in replacement heifer selection. A limitation to the widespread use of measuring RFI is the cost and complexity of collecting individual daily feed intake. An alternative method to predicting RFI, which has shown utility in mature cows, growing bulls, and steers, is to measure radiated energy losses by infrared thermography (IRT). The purpose of the present study was to investigate the potential of IRT as a predictor of RFI in growing replacement heifers. Sixty-one crossbred beef heifers were fed a balanced conventional barley silage ration. At Day 0, 94, and 113 a sequence of IRT images of the head were collected with a FLIR S60 camera. Average IRT temperature of the cheek region (CHK) was calculated for each date. Actual feed intake (FI), as measured by the Growsafe feeding system was regressed against average daily gain, metabolic mid-point weight and final ultrasound backfat to obtain expected feed intake (EFI). RFI was the difference between FI and EFI, and values ranged from −1.55 to 2.19 kg d-1 as-fed (avg = 0.0; sd = 0.777). Heifers were then classified into low, medium, or high RFI groups based on ± 0.5 of the standard deviation around the RFI mean. Using CHK for each date in a repeated measure analysis, heifers with a low RFI value (n = 17; avg = −0.915 kg d-1; sd = 0.316) or a medium RFI value (n = 27; avg = −0.003 kg d-1; sd = 0.214) displayed a CHK temperature of 19.98°C (se = 0.230) and 20.33°C (se = 0.184) respectively and were significantly different ($P <$ 0.01) from heifers with a high RFI value (n = 17; avg = 0.919 kg d-1; sd = 0.552) which displayed a CHK temperature of 21.31°C (se = 0.230). Data from this study suggest measurements of IRT may have utility as a rapid screening tool to predict growth efficiency in heifers.

**Key Words:** feed efficiency, cattle, infrared

**M432 Predicting ME and metabolizable protein (MP) balances of Santa Gertrudis cows under grazing conditions using a nutrition model.** A. D. Aguiar*[1,4], L. O. Tedeschi[1], K. McCuistion[2], D. S. DeLaney[3], and S. Moore[3], [1]*Texas A&M University*, *College Station,* [2]*Texas A&M University-Kingsville*, *Kingsville,* [3]*King Ranch*, *Kingsville, TX,* [4]*University of Florida*, *Gainesville*.

Predictions of Santa Gertrudis cow requirements of ME and MP (g/d) grazing pastures containing Kleberg bluestem (*Dichanthium annulatum*) and Coastal bermudagrass [*Cynodon dactylon* (L.) Pers] were performed using the Large Ruminant Nutrition System (LRNS). Simulations were performed using measured clime, forage, and animal data; and concentrate DMI. Forage DMI was predicted by the LRNS. Three reproductive cycles were evaluated: P1 (05/06 – 04/07), P2 (05/07 – 04/08), and P3 (05/08 – 04/09). The fractional fermentation rate (kd) of monthly forage NDF was determined using in vitro gas production technique. TDN values were estimated as 0.98 × (100 - NDF - CP - EE - ASH) + Digestible CP + 2.25 × (EE - 1) + (NDF - NDIN) × (kd/(kd + kp) + IDNDF) – 7 in which EE is ether extract, NDIN is N in the NDF, IDNDF is indigestible NDF, and kp is fractional passage rate. When kp was assumed to be 4%/h, predicted values had the greatest accuracy (Cb = 0.82), the least mean bias (−2.19%), but the least precision ($r^2$ = 0.59) compared with a published theoretical equation. The predicted TDN varied from 48.5 to 59 (P1), 46.6 to 61.5 (P2), and 46 to 59.6% (P3). The predicted DMI by the LRNS was 1.75, 1.99, and 1.86% BW for P1, P2, and P3; respectively, contrasting with literature data of 2.6% BW. When the LRNS predicted DMI was used, ME and MP balances were negative for most months of the year. When 2.6% of BW was used, the ME and MP balances were negative only for May and Jun for P1; Apr (ME) and Apr to Jun (MP) for P2; while cows in P3 had predicted positive ME and MP balances for Jul, Aug, and Sep. The results of 2.6% BW to predict DMI were more consistent with the observed BW and BCS changes during these periods (ADG = 0.16 kg/d), suggesting that cows had an overall positive MP and ME balances and likely used the surplus of nutrients for growth and to increase BCS. The LRNS underpredicted forage DMI given the predicted forage TDN. The use of empirical equations to predict DMI of grazing beef cows may not be acceptable in designing supplementation strategies. More integrated and mechanistic nutrition models are needed to predict DMI.

**Key Words:** cattle, requirements, supply

**M433 Estimating rumen microbial crude protein in vitro using purine analysis or real-time PCR.** E. Castillo-Lopez*, P. J. Kononoff, and J. L. Miner, *University of Nebraska-Lincoln*, *Lincoln*.

In ruminants, microbial crude protein (MCP) contributes to metabolizable protein. Estimation of MCP has traditionally been based on purine analysis, but it may be confounded by purines not originating from microbial DNA, therefore, a more direct approach is needed. The objectives of this experiment were to compare real-time PCR and purine analysis on estimates of MCP and to evaluate the impact of fermentation time, and lastly to evaluate the impact of dried distillers grains and solubles (DDGS) when replacing hay and corn. For each treatment, 1 g of substrate was incubated in 100 mL of rumen inoculum and replicated 3 times. A 3X2X2 factorial design experiment was used to determine the impact of 3 fermentation substrates at 2 times estimated with 2 methods on MCP. Treatments were as follows, CONTROL (50% grass hay and 50% rolled-corn), LOW DDGS (33% grass hay, 33% rolled-corn and 33% DDGS) and HIGH DDGS (100% DDGS.). At each time point a pellet was isolated and bacterial, protozoal and yeast crude protein was estimated using real-time PCR and purine analysis. To do so, microbial markers (primers and probe) were designed from the 16S rRNA, 18S rRNA and the second chromosome; for bacteria, protozoa and yeast (*Saccharomyces cerevisiae*), respectively. Estimates of MCP were different ($P < 0.05$) and were 221 and 246 (SEM = 6.55) mg/g DM according to the real-time PCR assay and that based on purine analysis. Fermentation substrate affected ($P < 0.05$) yield of MCP which was 263, 243 and 195 (SEM = 7.77) mg/g DM for CONTROL, LOW DDGS, and HIGH DDGS respectively. Fermentation time did not ($P = 0.25$) affect yield of MCP which was 229 and 239 (SEM = 6.34) mg/g of DM at 0 and 48h fermentation respectively. There was no method by diet interaction ($P$ = 0.44). Inclusion, the real-time PCR estimates of MCP differed from those obtained with the purine analysis and it may be a feasible approach for the estimation of MCP. In addition, increasing level of DDGS may affect the in vitro synthesis of MCP.

**Key Words:** DDGS, microbial crude protein, purine analysis, real-time PCR

**M434 An in vitro gas production technique to evaluate the effect of microwave irradiation on fermentation potential of cottonseed hulls using medium of ruminal fungal isolation.** A. Faramarzi Garmroodi, M. Danesh Mesgaran*, H. Jahani-Azizabadi, A. R. Vakili, A. Tahmasbi, and A. R. Heravi Moussavi, *Dept. of Animal Science (Excellence Center for Animal Science), Ferdowsi University of Mashhad*, *Mashhad, Iran*.

Gas production parameters of microwave irradiated cottonseed hulls (CH) were assessed using gas production procedure of medium containing ruminal fungal isolation. Cottonseed hulls were hydrated by distilled water as 25 g/kg DM, then, irradiated (microwave, 900 W) for 4, 6 and 8 min (mic4, mic6 and mic8, respectively). Approximately, 0.3 g of each sample was placed in a 100 mL glass syringes (n = 4), then, incubated into 40 mL of buffered protozoa and bacteria free rumen fluid (ratio of buffer to rumen fluid was 2:1) for 2, 4, 6, 8, 12, 24, 48, 72 and 96 h. Rumen fluid was obtained from 3 sheep (body weight, 49.5 ± 2.5 kg) fitted by rumen fistulae, before the morning feeding, and strained through 4 layers of cheesecloth. The animals were fed 1 kg/d DM of alfalfa hay and 0.3 kg/d DM of concentrate (165 g CP/kg DM). Rumen fluid was centrifuged (10 min, 3000 rpm) and a solution of penicillin and streptomycin was added to protozoa free supernatant to remove the bacterial population. Data of gas produced over the incubations were applied to an exponential equation of $P = b(1\text{-}e)^{-ct}$ where (b) is the amount of produced gas from the fermentable fraction, (c) is the fractional constant rate and (t) is time. Data were analyzed using the GLM procedure of SAS 9.1 and the means were compared by the Tukey test ($P < 0.05$). The amount of gas produced from the fermentable fraction (b ± SEM) was 3.7 ± 2.82, 3.9 ± 2.17 and 4.4 ± 2.98 for mic4, mic6 and mic8, respectively. The fractional constant rate parameters (c ± SEM) were 0.01 ± 0.011, 0.01 ± 0.011 and 0.01 ± 0.018 for mic4, mic6 and mic8, respectively. Present data indicate that the physical procedure used has not any benefit on fermentation potential of CH while a medium of isolated ruminal fungal was used.

**Key Words:** cottonseed hulls, gas production, microwave irradiation

**M435 The effect of microwave irradiation on gas production parameters of cottonseed hulls using medium containing ruminal bacterial isolation.** A. Faramarzi Garmroodi, M. Danesh Mesgaran*, H. Jahani-Azizabadi, A. R. Vakili, A. Tahmasbi, and A. R. Heravi Moussavi, *Dept. of Animal Science (Excellence Center for Animal Science), Ferdowsi University of Mashhad*, *Mashhad, Iran.*

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**Key Words:** cottonseed hulls, gas production, microwave irradiation

**M436 The influence of extrusion of low-glucosinolate full-fat rapeseed and whole pea on site and extent of protein digestion in dairy cows.** C. Bayourthe* and F. Enjalbert, *UMR 1289 INRA/INPT/ENVT TANDEM*, *31326 Castanet-Tolosan, France*.

The objectives of the study were to develop suitable treated blends for protection of low-glucosinolate rapeseed (canola) proteins from rumen degradation and to determine the protein quality after rumen exposure. Full-fat canola seeds (CS) were mixed either with canola meal (CM) or with canola meal plus whole pea seeds (PS). The effect of extrusion at 130 and 150°C on in situ crude protein (CP) degradability of raw and treated blends was measured by the nylon bag technique using 3 fistulated non-lactating Holstein cows. Ruminal degradation rate of CP was estimated as percent nitrogen degradation (DgN) from polyester bags incubated in rumen for 2, 4, 8, 16, 24 and 48 h. Data were fitted to the nonlinear regression equation: $DgN(t) = a + b(1 - e^{-ct})$ where DgN is percentage disappearance of N at time t, a the soluble fraction and b the less rapidly degradable fraction which disappears at the constant fractional rate c per time t. Heating the blends at 130 and 150°C decreased the effective degradability of crude protein (EDCP) when compared with the raw blend: 37.3 and 33.6 respectively vs 57.2% for CM/CS blend; 53.8 and 42.4 respectively vs 62.5% for the CM/CS/PS blend. Total CP disappearing in the digestive tract was estimated by incubating bags in the rumen for 16h, followed by a pepsin bath for 2h and then introduced into the duodenum for subsequently recovery in feces. For the CM/CS blend, amounts of rumen undegraded dietary CP digested in the intestine were increased from 32.7 (raw blend) to 49.3% at 130°C and 53.6% at 150°C. Similarly, for the CM/CS/PS blend, corresponding values were 27.9 for the raw blend to 38.2 and 53.3% respectively for 130 and 150°C heat treatments. The results showed that PS was the most effective carrier of rapeseed during extrusion. The tested blends and the treatments applied appeared to be a viable and consistent method of increasing the ruminally undegradable protein fraction

**Key Words:** extrusion, canola, ruminal degradation

**M437 In situ ruminal degradability of dry matter and crude protein of soybean meal treated with formaldehyde and extrusion.** A. A. Naserian* and H. Gholizadeh, *Ferdowsi University of Mashhad*, *Mashhad, Iran*.

The objective of this study was to determine effect of formaldehyde and extrusion on in situ ruminal degradability of DM and CP of soybeans. Treatments include; untreated soybean meal (USM) treated with formaldehyde (TF) and extruded soybean (ES). The treated soybean meal was sprayed with 2.5 (w/v) formaldehyde at the rate of 0.2 l/kg. Extrusion was conducted on ground soybeans (EXP 160 KW). Extrusion temperature and residence time averaged 1550 C and 25 s respectively. Four ruminally fistulated steers (400 ± 20 kg, body weight) were used. Steers were fed 5.2 kg of alfalfa hay, 1.3 kg of corn silage and 2.6 kg of concentrate. Bags (12 × 19 cm, pore size of 48 µm) containing 5 g DM of each sample was incubated in the rumen (4 replicates per each animal) for 0.0, 2, 4, 8, 16, 24, 48, and 72 h. After removal of the bags

from the rumen, they were washed using cold water and dried in a forced air oven (60 0C, 48 h), weighed to determination DM disappearance, and CP of the samples determined. Data was fitted to exponential model to calculate degradation parameters of CP and DM (Orskove et al.,1980). TF had the lowest in situ quickly degradable (a) fraction (27%, $P < 0.05$). No difference in (a) fraction of DM was observed between ES and USM (42 and 38% respectively, $P > 0.05$). In situ (a) fraction of CP was highest for USM (27%, $P < 0.05$) and had no difference between ES and FT (7 and 11% respectively, $P > 0.05$). Slowly degradable (b) fraction of DM did differ between treatments ($P < 0.05$). USM had the highest (b) fraction (63%) and did not differ between TF and ES (34 and 27% respectively, $P > 0.05$). Slowly degradable (b) fractions of CP had no difference between ES and USM (78 and 72% respectively, $P > 0.05$), were higher than TF (34%). Rate of degradation (c) of DM and CP were significant ($P < 0.05$). TF had a lower (c) fraction of DM and CP (3 and 1% respectively) compared with USM (9 and 9%) and ES (12 and 4%) compared with USM. It was concluded that TF decreased (a) and (b) fraction of DM and CP relative to USM. But, ES led to decreased (a) and increased (b) fraction.

**Key Words:** soybean, extrusion, formaldehyde

**M438 Disappearance of total carotenoids in the rumen and intestine of steers measured using a mobile nylon bag technique.** R. G. Cruz-Monterrosa*[1], I. Guerrero-Legarreta[1], and E. Ramirez-Bribiesca[2], [1]*Universidad Autonoma Metropolitana*, *D.F., Mexico*, [2]*Colegio de Postgraduados*, *Texococo Mexico*.

There are few studies on the disappearance of total carotenoids in tropical forages. However, it could be a contributing factor in the wide variation in bio-availability of carotenoids of natural forages. The disappearance of dry matter (DM) and total carotenoids were measured in 4 Holstein steers (312 kg) using a mobile nylon bag technique. In situ effects and differences between forage in rumen and intestine, and total tract disappearance of DM, and total carotenoids were analyzed using the GLM procedure of SAS with Tukey multiple range test used for the comparison of means. A higher ($P < 0.05$) proportion of the dry matter and total carotenoids in the *Cynodon* spp. disappeared drastically in the rumen during the first 12 h, and after were stabilized from 24 to 72 h. A similar trend ($P < 0.05$) was evident in the disappearance of the total carotenoids in the *Cynodon* spp. Correlation value within *Cynodon* spp. between the disappearance of DM and total carotenoids in the rumen was 0.997 ($P < 0.001$). 53% of the carotenoids in the *Cynodon* spp. disappeared from the duodenal bags in the lower digestive tract when it was not incubated in the rumen. Bags with longer incubation in the rumen contain less carotenoids and therefore the efficiency of disappearance in the small intestine decreased ($P < 0.05$). In the total disappearance of carotenoids content in digestive tract was half when the grass sample was not incubated into the rumen; subsequently, the disappearance of total carotenoids in the small intestine increased in forage samples with significant differences between ruminal hours post incubation ($P < 0.05$). These results show that apparent availability in the total digestive tract was higher to 0.70 of intake. The concentration of total carotenoids in *Cynodon* spp. was 627 mg/kg DM. It is concluded that degradability of total carotenoids contained in *Cynodon* spp. is high and not all absorbed as carotenoids. Thus, the yellow color of beef fat is caused by accumulation of carotenoids in the fat depots.

**Key Words:** carotenoids, *Cynodon* spp., cattle

**M439 The relationship between intestinal digestibility of crude protein and dry matters and the protein fractions with ruminant feedstuffs.** R. Zhou, J. Q. Wang*, F. M. Pan, D. P. Bu, H. Y. Wei, and L. Y. Zhou, *State Key Laboratory*, *Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, P. R. China*.

The study was to find out the relationship between the degradation and the digestibility and the contents of each protein fractions of 11 feeds common used in China. Chemical contents were determined according to the methods provided by CNCPS Version 5.0. The mobile nylon bag technique was used to investigate the apparent small intestinal digestibility of crude protein (CP) and dry matters (DM).Three dairy cows fitted with permanent ruminal cannulas and T-type duodenal cannulas were used to investigate intestinal digestibility of CP and DM by mobile nylon bag technique in soybean meal (SBM), cottonseed meal (CSM), rapeseed meal (RSM), peanut meal (PM), flaxseed meal (FSM), linseed meal (LSM), distillers dried grains (DDGS), expanded soybean (ES), corn grain (CG), brewer's grains (BG), alfalfa hay (AH), chinese wildrye (CW) and whole corn silage (WCS). The content of undegraded DM after 16 h incubation was affected by the NDF and PB3 fractions ($r^2 = 0.9088$). The content of ruminal undegraded protein (RUP) have a high relationship with contents of the NDF and PA fractions ($r^2 = 0.6227$). The absorbed DM in intestine increased while the feeds have more NDF. The protein content absorbed in the intestine can be mainly predicted by the NDF and PC content ($r^2 = 0.7344$). In our result, we can see that the intestinal absorbed DM and CP have a high relationship with the content of each protein fractions.

**Key Words:** protein fractions, intestinal digestibility, ruminants

**M441 Enzymatic activity of microorganisms attached to solid residues of *Festulolium*, fermentation variables and in vitro kinetics of gas production.** I. Almaraz-Buendía[1], S. S. González-Muñoz*[1], O. Loera[2], L. A. Miranda-Romero[3], M. A. Cobos-Peralta[1], M. Meneses-Mayo[1], B. Alarcón-Zúñiga[3], and R. Bárcena-Gama[1], [1]*Colegio de Postgraduados*, *Montecillo, Edo. de México, México*, [2]*Universidad Autóoma Metropolitana–Iztapalapa*, *México D.F., México*, [3]*Universidad Autónoma Chapingo*, *Chapingo, Edo. de México, México*.

The objective of the present study was to determine the effect of *Festulolium*, harvested either at 28 (T1) or 35 d, on enzymatic activities from microorganism attached to this fermented substrate [IU/g DM; at 39°C and pH 6.8: after 1 h for carboxymethycellulases (CMCases) and after 30 min for xylanases], in addition to VFA, N-NH3 and in vitro DM disappearance (IVDMD). Samples of enzymatic extract (EE) were obtained at 12, 16, 24 and 48 h of fermentation. Xylanolytic activity was determined at substrate concentration ranges ensuring linear release of reducing sugars (R2 = 0.99 for up to 60 min). Maximal gas volume (Vmax), lag phase (L) and fractional rate (S) were calculated with a logistic model and PROC NLIN (SAS). The experimental design was incomplete randomized blocks with a split-plot arrangement and data was analyzed using PROC MIXED (SAS). Enzymatic activity and N-NH3 were similar among treatments ($P \geq 0.05$). The highest CMCase activity ($P \leq 0.05$) was obtained at 12 (T1:8.19, T2:7.75 IU/g DM) and 16 h (T1:7.75, T2:7.65 IU/g DM) of fermentation, whereas maximal xylanolytic levels ($P \leq 0.05$) were observed after 16 (T1:51.09, T2:46.68 IU/g DM) and 24 h (T1:51.23, T2:46.15 IU/g DM). The highest concentration ($P \leq 0.05$) of acetic (30.05 vs 42.92 mM/L), propionic (9.69 vs 12.3 mM/L) and butyric acids (8.03 vs 12.04 mM/L) were detected ($P \leq 0.05$) in T2 after 48 h of fermentation. Both maximal ($P \leq 0.05$) IVDMD and Vmax were also observed in T2, whereas L and S were highest ($P \leq 0.05$) in T1. According to these results, in vitro CMCases and xylanases from microorganism attached to *Festulolium*, harvested either at 28 or 35 d, remain unchanged; however IVDMD and gas

production kinetics did show an effect as a function of harvesting date of *Festulolium*.

**Key Words:** *Festulolium*, enzymatic activity, microbial attachment

**M442 Use of in vitro starch and neutral detergent fiber degradation rates to predict carbohydrate availability.** M. A. Brooks*, N. F. Johnson, R. M. Harvey, and M. S. Kerley, *University of Missouri*, *Columbia*.

The purpose of this in vitro experiment was to compare degradation rates ($K_d$) of ruminally degradable starch and neutral detergent fiber (NDF) of various feedstuffs, then use these data to predict carbohydrate (CHO) release. The feedstuffs, ground corn (GC), corn bran (CB), corn starch (CS), dried distiller's grains (DDG), soy hulls (SH), and ground alfalfa (AL), were fermented in rumen inoculum. Carbohydrate degradation was determined at 0, 4, 8, 12, 16, 24, 36, and 48 h for each feed. Undigested feed was separated via differential centrifugation (1,000 × g, 15 min) and dried at 55°C. Samples were analyzed for total starch and NDF. The time point at which degradation reached extent (no further starch or NDF was degraded) was set to 100%. Carbohydrate mass at each time point was calculated as proportion of starch or NDF remaining. Data were analyzed using proportion of potential degradation as the dependent variable with flask as the experimental unit to determine homogeneity of slope with the means adjusted to time as a covariate. Pair-wise comparisons for similarity were then done of each feed using a significance level of $P > 0.01$. Slope of the data was indicative of degradation rate over time. All $R^2$ values were >0.80, and since these data were evaluated as proportions and were corrected for extent of degradation, all intercepts values were similar to 1.0. The starch $K_d$ values for AL, CB, CS, DDG, GC, and SH were 5.01, 6.08, 4.80, 5.51, 2.31, and 4.14% $h^{-1}$, respectively. Analysis showed GC had a slower $K_d$ ($P < 0.01$) than all other feeds, which were not different from each other ($P > 0.01$). The NDF $K_d$ values for AL, CB, CS, DDG, GC, and SH were 2.18, 2.24, 2.16, 2.18, 2.44, and 2.31% $h^{-1}$, respectively. Analysis showed no differences in NDF $K_d$ ($P > 0.01$). Digestion of CHO in the rumen occurred at similar rates independent of feedstuff. Similarity of starch and NDF $K_d$ across feeds, when adjusted for passage rate, makes possible prediction of CHO digestibility in the rumen by ruminal microbes and prediction of microbial growth and efficiency potential.

**Key Words:** starch, NDF, ruminant

**M443 Effect of lysozyme-adapted *Lactobacillus acidophilus* on fermentation in an artificial rumen system (Rusitec).** M. L. He[1,3], T. A. McAllister*[1], and L. M. Rode[2], [1]*Agriculture and Agri-Food Canada, Research Centre*, *Lethbridge, AB, Canada*, [2]*AB Sage Biosciences Inc.*, *Edmonton, AB, Canada*, [3]*University of Saskatchewan*, *Saskatoon, SK, Canada*.

Direct-fed microbials are increasingly being used as an alternative to subtherapeutic antibiotics in dairy and beef production because of their potential to enhance animal performance without promoting the emergence of antibiotic-resistant bacteria. Lysozyme is an enzyme that hydrolyzes the 1,4-β-linkages between N-acetylmuramic acid and N-acetyl-D-glucosamine residues in the peptidoglycan of gram-positive bacteria. The cell wall of lysozyme-adapted bacteria may exhibit increased rigidity and thereby contribute to their perseverance in highly competitive environments such as the rumen ecosystem. The Rumen Simulation Technique (Rusitec) was used to evaluate the effect of a lysozyme-adapted (L- BT1386) or a non-adapted strain (BT1386) of *Lactobacillus acidophilus*, as well as strain La 14, on ruminal fermentation. Ruminal fluid for the Rusitec was collected from 3 Holstein cows fed a 90% concentrate:10% silage diet. Sixteen fermenters (920 mL capacity; dilution rate 0.029/h) were provided fresh substrate daily (8 g steam-rolled barley grain + 2 g silage; DM basis). Four fermenters per treatment were inoculated with $5 \times 10^{10}$ cfu/d of the specified strain or with buffer only. Compared with non-inoculants, L-BT1386 increased ($P = 0.04$) gas production (mL/d) and BT1386 increased ($P = 0.02$) VFA and ammonia production. Methane emissions (mL/d) were numerically greater ($P = 0.09$) in inoculated fermenters than in non-inoculants, as were disappearances (at 48 h) of DM, ADF, NDF and N from barley grain and silage ($P = 0.20$ to 0.66). Protozoal numbers were similar among treatments ($P = 0.21$) but decreased ($P < 0.05$) over time (d 9 to 18). Measured fermentation parameters did not differ between L-BT1386 and BT1386. Studies using strain-specific molecular probes are underway to determine if exposure to lysozyme increased the numbers and persistence of BT1386 within the Rusitec.

**Key Words:** direct fed microbials, rumen fermentation, rumen simulation technique

**M444 Development of a PCR assay for the detection of *Zymomonas mobilis* in distillers grains.** M. A. Rasmussen* and F. H. Benahmed, *U.S. Food and Drug Administration, Center for Veterinary Medicine, Office of Research*, *Laurel, MD*.

The objective of this project was to develop a PCR assay for the detection of *Zymomonas mobilis* (Zm) in distillers grains (DG). This bacterium can produce ethanol at rates and concentrations exceeding those of yeast. This has made Zm, an attractive alternative or supplement to *Saccharomyces cervisiae* for ethanol production. Naturally occurring Zm can only ferment glucose, fructose and sucrose and as a result there has been an ongoing effort to modify the microbe to broaden its substrate range. However, the use of Zm as a primary fermentative microorganism may alter the nutritional content of DG and the extent of its use for ethanol production is currently unknown. Therefore, we developed an assay for DG that can be used to determine if Zm is being used for ethanol production. PCR is a useful and sensitive technique that can detect the bacterium even if nonviable or nonculturable. Reference strains of Zm were obtained from the ARS culture collection in Peoria, IL and were cultivated in YP media under oxygen limiting conditions. Overnight cultures were serially diluted into buffer supplemented with Tween 20. Zm cells were harvested and tested directly by PCR. Primer sequences specific for Zm targeted a 900 bp rRNA overlapping region of the 16s gene Z16p3 and the 23s gene Z23p5. For tests in the relevant matrix, the assay was tested on DG samples inoculated with serial dilutions of Zm cells. A single PCR band was visualized in all Zm strains investigated and no band was observed in the negative control DG matrix. Additionally, plate culture was used for assay optimization and confirmation. The limit of detection for the PCR assay was determined to be 10 cfu/ml of the DG wash/suspension buffer or 100 cfu/g of DG. Current activities are directed at optimizing the assay for use with DG field samples to determine if Zm is present. In conclusion, this assay can be used to test for Zm and is more rapid and sensitive than traditional culture methods.

**Key Words:** distillers grains, *Zymomonas*, bioethanol

**M445 Intake prediction using *n*-alkanes in beef cattle fed a mixture of switchgrass and alfalfa hay.** S. J. Chavez*, C. Baum-Lane, E. Leonard, J. Burns, and G. B. Huntington, *North Carolina State University*, *Raleigh*.

The objectives of the study were to use *n*-alkanes to predict intake in beef cattle fed mixed forage and to predict intake of forage constituents based

on forage alkane concentrations. In May and June 2009, 11 Angus-cross steers and 1 heifer (BW = 283 ± 25 kg) were housed under a roof on expanded metal flooring with access to 6 feeding stations. Cattle were fed 1 kg of a soyhull and corn supplement once daily at one feeding station that was sprayed with dotriacontane (C32) and hexatriacontane (C36). Cattle consumed approximately 300 mg each of C32 and C36. Cattle were offered a switchgrass:alfalfa (3:1 as fed) mixed hay at 0.6% BW at each feed station. Hay was ground in a hammer mill to minimize sorting. Periods consisted of one week adaptation followed by 14 d of feeding supplement with added alkanes. Fecal grab samples were collected between 0800 and 1000 during the last 5 d. Fecal samples were stored frozen and oven-dried to constant weight at 60°C. Hay and fecal samples were saponified, alkanes were extracted with heptane, and samples analyzed by gas chromatography for alkane concentrations. One steer was removed from data analysis due to inconsistent eating and rear leg inflammation and another was removed for highly variable fecal ratios of hentriacontane (C31) and C32. Tetratriacontane (C34) was used as an internal standard for saponification and extraction with recoveries ranging from 80 to 90%. When feed and fecal ratios of C31 and C32 were used in prediction equations, predicted daily DMI (5.96 ± 0.70 kg) was not different ($P < 0.35$) from measured intake (5.51 ± 0.68 kg). Using simultaneous equations and concentrations of nonacosane (C29) and C31 in the hays, the predicted ratio of switchgrass:alfalfa (3.09:1) was similar to the actual ratio. Using C36, predicted digestibility was 58.7 ± 2.2% for the switchgrass and alfalfa hay. Alkanes can be used to predict intake in cattle fed a mixed forage diet and also predict the proportion of each forage consumed.

**Key Words:** cattle, alkane, intake

**M446 A comparison of methods to evaluate in vitro intestinal digestibility.** D. A. Ross*, M. M. McCullouch, and M. E. Van Amburgh, *Cornell University*, *Ithaca, NY*.

Various assays are used to evaluate the intestinal digestibility of dry matter (DM) and nitrogen (N) in ruminant feeds (Casamiglia and Stern, 1995; Gargallo et al. 2006). The objective of this study was to evaluate in vitro (IV) intestinal digestibility of DM and N of 7 feeds (fishmeal, corn silage, alfalfa silage, 2 soy products and 2 DDG) following in situ (IS) or ruminal IV procedures. We wanted to determine if ruminal IV or IS exposure or length of exposure altered the IV intestinal digestibility and further, if the pore size of the incubation bag affected variation of the assay. Samples (0.5g) were placed in 3 pore size (15, 25 or 50 µm) bags (5 × 5 cm) and incubated for 12-h or 24-h ruminal digestion either IS or IV (Daisyincubator, Ankom, Macedon, NY) followed by IV intestinal digestion (modified Gargallo et al. 2006). Empty bags were incubated for correction. Data were analyzed as a factorial design using GLM in SAS and Tukey's method to separate means. Incubation time in the rumen or rumen fluid affected digestibility, but there was no difference in IV intestinal digestibility based on ruminal exposure time (Table 1). Digestibility of DM and N_DM were significantly lower in the 25 µm pore bags; however no differences among bag pore sizes were detected for IV intestinal digestion parameters. The effect of bag pore size for ruminal IV or IS digestion was not linear, due to characteristics of the bag material and will be discussed.

**Table 1.** The mean (± SD) values for digestible DM and N

| | RDM | RNDM | TDM[1] | TNDM[1] |
|---|---|---|---|---|
| n | 164 | 163 | 159 | 159 |
| RumDigTime | | | | |
| IV12 | 0.43 ± 0.13[a] | 0.68 ± 0.15[a] | 0.78 ± 0.18 | 0.98 ± 0.04 |
| IV24 | 0.43 ± 0.13[†] | 0.71 ± 0.15[a] | 0.80 ± 0.17 | 0.98 ± 0.03 |
| IS12 | 0.43 ± 0.13[a] | 0.59 ± 0.16[b] | 0.82 ± 0.18 | 0.98 ± 0.05 |
| IS24 | 0.53 ± 0.17[b†] | 0.70 ± 0.18[a] | 0.85 ± 0.15 | 0.98 ± 0.04 |
| BagPore,µm | | | | |
| 15 | 0.47 ± 0.13[a] | 0.69 ± 0.14[a] | 0.82 ± 0.17 | 0.99 ± 0.02 |
| 25 | 0.39 ± 0.13[b] | 0.58 ± 0.17[b] | 0.78 ± 0.18 | 0.97 ± 0.05 |
| 50 | 0.52 ± 0.15[a] | 0.74 ± 0.15[a] | 0.84 ± 0.16 | 0.98 ± 0.04 |

[ab]Means with different letters are significant, $P < 0.05$.

[†]Common superscript show trends, $P < 0.10$.

[1]Cumulative digestion.

**Key Words:** in vitro, in situ, intestinal digestion

**M447 The role of ADIN in determining nutrient availability in new co-products from bio-ethanol processing.** W. G. Nuez-Ortín* and P. Yu, *Department of Animal and Poultry Science, College of Agriculture and Bioresources, University of Saskatchewan*, *Saskatoon, SK, Canada*.

The objectives of this study were to investigate the role of acid detergent insoluble nitrogen (ADIN) in determining the nutrient availability in the new co-products of bio-ethanol processing and the relationship between ADIN content and nutrient utilization of the bioethanol co-products in ruminants. The corn DDGS, wheat DDGS and blend DDGS (70% wheat: 30% corn), and wheat and corn samples with 3 to 5 different batches were obtained during 2007–2008. The results showed that compared with the grains, DDGS contained higher ($P < 0.05$) acid detergent insoluble crude protein (ADICP) level from 1.2 to 7.6%CP vs. 0 to 0.1% crude protein (CP) in the grains. However, all ADICP levels were lower than 13% of total CP across DDGS samples, revealing no effects ($P > 0.05$) on protein ruminal digestion with a correlation between ADICP and rumen-degradable protein (RDP): R = −0.08, $P = 0.74$. However, the difference in the ADICP content was reflected in largely numerical differences in the intestinal digestibility of rumen-undegradable protein (RUP) in vitro, which ultimately affected ($P < 0.05$) the intestinal availability of RUP (ARUP) as well as the predicted total post-ruminal protein supply and availability (the correlation between ADICP and metabolizable protein (MP): R = −0.74, $P < 0.05$). This suggests a higher sensitivity to low ADICP levels in the rumen than that in the small intestine in the DDGS samples.

**Key Words:** nutrient utilization and availability, bio-ethanol co-products, ADIN

**M448 A comparison of models used to estimate kinetics of in vitro degradation of alfalfa hay dry matter.** C. A. Old*[1] and D. A. Sapienza[2], [1]*California Chapter of the American Registry of Professional Animal Scientists*, *LeGrand, CA,* [2]*Sapienza Analytica, LLC*, *Des Moines, IA*.

To determine more accurately alfalfa hay quality California ARPAS undertook a project to estimate metabolizable energy, rate, site and extent of degradability of selected chemical and proximate entities and use these as part of a set of prediction equations in near infrared (NIR) spectrophotometry. This study had in vivo, in vitro and in silico components based on 150 samples of alfalfa hay collected during the

2008 growing season in California. The diversity of samples can be seen from the range of crude protein and acid detergent fiber content, the former was 13 percentage points and the latter 30 percentage points. Samples for in vitro analysis were ground through a 6-mm screen and incubated from 1 to 120 h. Losses of N were calculated from 16-h amino acid losses and averaged 63%. Disappearance of individual amino acids ranged from a low of 57% (Leu) to a high of 75% (Pro). Variation in losses among samples was less than variation among amino acids. Dry matter losses, at 120 h, averaged 80.2%. Kinetics of in vitro dry matter degradation were estimated using a heterogeneous, stochastic model assuming a gamma distribution of rates (HS): $C_t = D(1+\beta[t-\tau])^{-\alpha}+I$, a single exponential deterministic model (SE): $C_t = C_0e^{-kt}$ and a biexponential deterministic model (BE):$C_t = C_{01}e^{-k1t}+ C_{02}e^{-k2t}$. $C_t$ is the residue at time t, D is the degradable fraction, β and α are shape parameters of a gamma distribution, τ is lag time, I is the undegradable fraction,$C_{0n}$ is compartment n at time 0 and $k_n$ is the rate constant for compartment n. Average residual sum of squares for models HS, SE and BE were 0.012, 4.29 and 0.160, respectively. As a result of this study, kinetics of alfalfa hay degradation will be described in the NIR model using a heterogeneous, stochastic model.

**Key Words:** alfalfa hay quality, kinetic models, amino acids

**M449 Application of near infrared spectroscopy to estimate composition of NuPro.** G. A. Harrison*, M. D. Meyer, E. C. Taylor, and K. A. Dawson, *Alltech*, *Nicholasville, KY*.

The properties of near infrared spectroscopy (NIR) make this technology attractive in quality assurance programs. Provided reliable reference methods are utilized to determine component concentrations in calibration samples, NIR can potentially replace routine wet chemistry. The objective of this project was to determine the feasibility of using NIR to estimate the composition of NuPro (an extract of a select yeast strain containing a combination of amino acids, peptides, nucleotides, inositol, and glutamic acid) in the Alltech Quality Assurance Program. From an initial 239 NuPro samples, 100 calibration samples and 20 test samples were randomly selected with equal representation from each of 2 manufacturing sites. Samples were scanned in triplicate using a Bruker MPA FT-NIR Spectrometer (Bruker Optics, Inc., Billerica, MA). For each component, 3 calibration models were developed using cross validation and various spectral preprocessing methods. Multiple criteria were utilized to determine the best fit model including coefficient of determination (R2), root mean square error of cross validation (RMSECV), and residual prediction deviation (RPD). Additionally, NIR-predicted values of test samples were compared with values measured by reference methods. Bias and percentage of samples with predicted values within 5 and 10% of measured were calculated. A 2-tailed paired *t*-test was used as the final criterion. The best fit DM model had an $R^2$ of 84.6, RMSECV of 0.479, and RDP of 2.55 with 100% of samples were within 5%. The best fit N model had an $R^2$ of 93.0, RMSECV of 0.072, and RDP of 3.77 with 100% of samples within 5%. For total nucleotides, the best fit model had an $R^2$ of 86.6, RMSECV of 0.603, and RDP of 2.73 with 60 and 80% of samples within 5 and 10%, respectively. Statistical analyses by paired *t*-test found no difference between measured and predicted values for DM, N, C, H or total nucleotides ($P > 0.10$). For quality assurance purposes, composition of NuPro can be estimated through the use of NIR models.

**Key Words:** near infrared spectroscopy, quality assurance

**M450 Ability of NIR to predict crude fat, fatty acids and unsaturated fatty acids in total mixed ration fed to dairy cattle.** S. Weaver*[1], R. Ward[1], and R. A. Patton[2], [1]*Cumberland Valley Analytical Services*, *Maugansville, MD*, [2]*Nittany Dairy Nutrition*, *Mifflinburg, PA*.

It is known that formation of CLA due to consumption of high levels of linoelic and linolenic acids may have a negative effect on milk fat production depending on the rumen environment. Determination of the amounts and types of fatty acids (FA) in total mixed rations (TMR) would allow nutritionists to better control these elements. At present determination of fat and FA percent as well as individual fatty acid amounts is expensive and time consuming. We postulated that NIR has the potential to rapidly and inexpensively predict the amount of fat, percentage of FA, and the FA composition of this fat in TMR. A data set of 89 individual TMR samples were analyzed for crude fat by the AOAC method for feeds (2003.05). Fatty acids were analyzed as methyl esters on a Restek 30 m capillary column (90% biscyanopropyl / 10% phenylcyanopropyl polysiloxane) using a Perkin Elmer Autosystems GC with a flame ionization detector using the method of Sukhija and Palmquist (1988. J. Agric. Food Chem. 36:1202–1206). Comparisons of observed versus NIR predicted values were by the MSPE method described by Bibby and Toutenburg (1977). NIR predicted crude fat values were close to values for chemically defined fat (observed mean = 4.68, predicted mean = 4.74, RMSPE = 0.45 with 96% random error). The prediction of total FA was even more precise (observed mean = 3.63, predicted mean = 3.63, RMSPE = 0.2 with 99% random error). However, predictions of individual unsaturated fatty acids, although reasonable for mean values, displayed considerable regression bias. This suggests that further research may yield better NIR prediction equations for individual fatty acids.

**Key Words:** fatty acids, TMR, NIR

**M451 Evaluation of models to predict passage rate in cattle.** S. J. Krizsan*[1], S. Ahvenjärvi[2], and P. Huhtanen[1], [1]*Department of Agricultural Research for Northern Sweden, Swedish University of Agricultural Sciences*, *Umeå, Sweden*, [2]*MTT-Agrifood Research Finland, Animal Production Research*, *Jokioinen, Finland*.

The passage rate ($k_p$) prediction equations in the National Research Council (2001) and the Cornell Net Carbohydrate and Protein System (CNCPS) were evaluated with compiled data of rumen evacuation studies. Data were comprised of 172 treatment means from 49 studies conducted in Europe and in the USA. A total of 145 diets were fed to dairy cows and 27 to growing cattle. The primary objectives of the trials included in the database were to study dietary effects on digestion and passage kinetics of fiber fractions. The concentration of indigestible NDF (iNDF) was determined by long-term ruminal incubations or in vitro incubations in rumen fluid. National Research Council (NRC) and CNCPS give separate $k_p$ prediction equations for concentrate and forage feed. Therefore, an aggregated $k_p$ was calculated according to: $k_p$ (%) = 100 × Flux of indigestible component into the compartment (kg/h) / Rumen pool of indigestible component (kg). Preferably intake of iNDF of forage and concentrate (n = 165), else intake of NDF from forages and concentrates (n = 7) were used to calculate the aggregated $k_p$. Rumen pool sizes of iNDF and NDF were defined separately for concentrate and forage feed and total pool size was estimated from the sum. Mixed model regression analysis including a random study effect was used to investigate the relationships between NRC and CNCPS predictions and observed $k_p$ of iNDF. Prediction equations were evaluated by regressing residual values on the predicted values. Relationships between predicted and observed $k_p$ were $y = 0.53(\pm0.187) + 0.41(\pm0.0373)x$ and $y = 0.58(\pm0.162) + 0.46(\pm0.0377)x$ for the NRC and CNCPS models, respectively. Residual analysis of the NRC and CNCPS models resulted

in both significant ($P < 0.001$) mean biases of −2.40 and −1.70% and linear biases of −0.59 and −0.53, respectively. This evaluation suggested that both the NRC and CNCPS models grossly overestimated ruminal particulate matter $k_p$.

**Key Words:** cattle, evaluation, passage rate

**M452 Net portal absorption of energy nutrients in ruminants: Assessment of prediction models.** C Loncke*[1], P Nozière[1], G Kraft[1], I Savary-Auzeloux[1], J Vernet[1], H Lapierre[2], D Sauvant[3], and I Ortigues-Marty[1], [1]*INRA, UR 1213*, *Theix, France*, [2]*Agriculture and Agri-Food Canada*, *Sherbrooke, QC, Canada*, [3]*INRA-AgroParisTech, UMR 791*, *Paris, France*.

In ruminant nutrition, the evolution of feed evaluation systems toward nutrient-based systems is an important challenge. Net portal appearance (NPA) of total VFA, acetate (C2), propionate (C3), butyrate (C4), glucose, β-hydroxybutyrate (BHBA), and lactate can be estimated with response equations based on ration intake and composition characterized according to INRA Feed Tables for Ruminants and derived from meta-analyses on the FLORA database. The present objective was to evaluate the models using the results of a recent study, conducted on 6 growing lambs, catheterized at the portal level. The lambs were fed 3 diets made of 30% hay and 70% of 1 of 3 concentrates, according to a Latin Square design. The control diet (C) was designed to offer a balanced and adequate supply of protein and ME for growing lambs according to INRA allowances. Low nitrogen (LN) and energy (LE) diets presented a 23% and 20% deficit in protein and ME supply respectively compared with the C diet. To evaluate the prediction ability of the models, observed (Y) and predicted NPA (X) was compared using a GLM model including a within-animal effect. The slopes obtained were equal to 0.88 ± 0.13, 0.78 ± 0.21, 1.06 ± 0.22, 0.88 ± 0.22, 0.80 ± 0.25, 1.30 ± 0.50, respectively for total VFA, C2, C3, C4, BHBA and lactate NPA and proved not different from 1. Moreover the intercepts of all models were not different from 0 ($P > 0.2$). The slope (12.4 ± 2.4) of the relation between observed and predicted values of glucose NPA showed a lower adjustment probably due to larger uncertainties on the ruminal starch degradability required for the prediction. In most cases the best prediction was observed for the C diet probably because diet compositions of LN and LE were not fully included in the meta-design range of validity used to establish the models. In conclusion, this work suggested that the energy nutrients NPA could be predicted with a good accuracy.

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**Key Words:** meta-analyses, energy nutrients, ruminants