

**T280 Supplementing rumen-protected Met and Lys in low protein diets based on corn distillers grains fed to lactating dairy cows.** N. E. Lobos<sup>\*1</sup>, G. A. Broderick<sup>2</sup>, and M. J. de Veth<sup>3</sup>, <sup>1</sup>University of Wisconsin, Madison, WI, <sup>2</sup>U.S. Dairy Forage Research Center, Madison, WI, <sup>3</sup>Balchem Corporation, New Hampton, NY.

Feeding rumen-protected Met (RPM) and Lys (RPL) may allow feeding lower CP diets to dairy cows, thereby increasing N efficiency and reducing environmental impact. Moreover, RPL supplementation may improve the value of corn distillers dried grains plus solubles (DDGS) because its RUP is limiting in Lys. A trial tested experimental preparations of RPM and RPL that provided 15 g/d of DL-Met and 26 g/d of L-Lys. Forty lactating Holstein cows were blocked by DIM and parity into 8 squares in a replicated 5 × 5 Latin square trial with 5 dietary treatments: 1) low CP control (14.9% CP) without supplement, diet 1) top-dressed with 2) RPM, 3) RPL, 4) RPM + RPL, and 5) high CP control (16.8% CP) without supplement. As fed, all diets contained (DM basis): 31% alfalfa silage, 31% corn silage, 7.7% DDGS, 2.4% mineral-vitamin premix and 31% NDF. Diets 1-4 contained 25% corn grain plus 3.3% solvent soybean meal (SBM); diet 5 contained 21% corn grain, 1.9% solvent SBM, 3.6% expeller SBM, and 1.9% corn gluten meal. Periods were 3-wk (total 15 wk); data from wk-3 were analyzed using Proc Mixed in SAS. Contrasts and LS-means are reported in the table. Relative to the low CP control, feeding higher CP increased DMI and yield of milk and protein, but also increased MUN. No RPM main effect was observed; however, the RPM\*RPL interaction for milk yield was significant because RPM increased milk 1.2 kg/d but milk was unaltered on RPM + RPL. Feeding RPL reduced ECM/DMI because numerically greater DMI had no effect on ECM yield. In this trial, feeding RPM, but not RPL, increased milk yield on low CP diets containing SBM plus DDGS.

Table 1.

Variable	CP, %: 14.9 14.9 14.9 14.9 16.8					Contrasts			
	RPM, g/d: 0	15	0	15	0	CP	RPM	RPL	RPM*RPL
DMI, kg/d	24.2	24.4	25.0	24.2	25.5	<0.01	0.34	0.29	0.08
Milk, kg/d	38.0	39.2	38.7	38.0	40.2	<0.01	0.58	0.54	0.05
Milk/DMI	1.60	1.65	1.59	1.60	1.61	0.58	0.12	0.16	0.28
ECM, kg/d	35.1	35.6	35.0	34.2	36.9	0.07	0.72	0.25	0.31
ECM/DMI	1.47	1.48	1.42	1.42	1.46	0.87	0.80	0.04	0.86
Fat, kg/d	1.41	1.41	1.39	1.34	1.46	0.22	0.53	0.14	0.42
Protein, kg/d	1.09	1.11	1.10	1.09	1.16	0.02	0.92	0.72	0.37
MUN, mg/dL	9.8	10.1	10.2	10.4	13.6	<0.01	0.29	0.07	0.78

**Key Words:** corn distillers dried grains, rumen-protected methionine, rumen-protected lysine

**T281 Performance and diet digestibility of dairy cows supplemented with *Bacillus subtilis* spores.** V. L. Souza<sup>2</sup>, V. A. Silveira<sup>1</sup>, N. M. Lopes<sup>1</sup>, O. F. Zacaroni<sup>1</sup>, R. A. M. Pereira<sup>3</sup>, J. A. de Freitas<sup>\*2</sup>, R. Almeida<sup>2</sup>, and M. N. Pereira<sup>1</sup>, <sup>1</sup>Universidade Federal de Lavras, Lavras, Brazil, <sup>2</sup>Universidade Federal do Parana, Curitiba, Brazil, <sup>3</sup>Empresa de Pesquisa Agropecuaria de Minas Gerais, Lavras, Brazil.

This experiment evaluated the supplementation of dairy cows with *B. subtilis*. Trial 1 used 18 Holsteins (246 DIM) in a crossover design, with 39-d periods, and a 10-d washout between periods. Treatments were orally dosed daily: 3x10<sup>9</sup> viable spores/d of *B. subtilis* C-3102 (Calpis Co. Ltda, Tokyo, Japan) or placebo. Cows were individually fed a TMR: 50% corn silage, 4.1% Tifton hay, 20.2% soybean meal, 11.2% high

moisture corn, and 10.2% citrus pulp. Diet digestibility was evaluated by total collection of feces during d 33 to 35. The NDF digestibility was 52.6% of intake for *B. subtilis* and 55.8 for placebo ( $P = 0.28$ ), OM digestibility was 72.1 and 72.6, respectively ( $P = 0.69$ ). No treatment effect on digesta passage rate and chewing activity were detected ( $P > 0.21$ ). Milk yield was 25.2 kg/d for placebo and 25.4 for *B. subtilis* ( $P = 0.66$ ), DMI was 18.3 ( $P = 0.91$ ). There was no treatment effect on milk solids and MUN ( $P > 0.17$ ). Milk SCC was 97,000 cells/mL for *B. subtilis* and 99,000 for placebo ( $P = 0.91$ ). Proportion of cows with positive *B. subtilis* culture in feces was 22% for placebo and 67% for *B. subtilis*. In trial 2, 30 cows (161 DIM) received the treatments for 16 weeks, in a covariate adjusted randomized block design. Milk samples were obtained weekly. Body weight and condition score were evaluated at 4-week intervals. The TMR contained 42.4% corn silage, 21.8% soybean meal, 15.6% ground corn, and 15.8% citrus pulp. Data was analyzed as repeated measures over time with Mixed of SAS. *B. subtilis* increased the daily secretions of milk (25.3 vs. 23.6 kg,  $P = 0.02$ ) and protein (0.816 vs. 0.763 kg,  $P = 0.01$ ), and had no effect on fat and lactose secretions ( $P > 0.35$ ). The milk and protein yield response was consistent along the entire trial ( $P > 0.90$  for the interaction of week and treatment). Milk SCC was 952,000 for *B. subtilis* and 747,000 for placebo ( $P = 0.56$ ). *B. subtilis* tended to decrease MUN from 20.8mg/dL to 19.3 ( $P = 0.06$ ). There was no treatment effect on body weight and condition score ( $P > 0.33$ ). The supplementation of *B. subtilis* spores increased milk and protein yield when the trial was performed with high SCC cows, but the mechanism for the response was not elucidated.

**Key Words:** *Bacillus subtilis*, direct-fed microbial, probiotic

**T282 Milk fatty acids composition of dairy ewes fed increasing levels of an unprotected CLA (UnCLA) supplement.** D. R. M. Alessio<sup>1</sup>, M. Baldin<sup>1</sup>, R. Dresch<sup>1</sup>, J. Souza<sup>2</sup>, M. A. S. Gama<sup>3</sup>, M. P. Soares<sup>4</sup>, and D. E. Oliveira<sup>\*5,1</sup>, <sup>1</sup>Centro de Ciências Agroveterinárias, UDESC, Lages, SC, Brasil, <sup>2</sup>Esalq/USP, Piracicaba, SP, Brasil, <sup>3</sup>Embrapa, CNPGL, Juiz de Fora, MG, Brasil, <sup>4</sup>Instituto Federal Catarinense, Araquari, SC, Brasil, <sup>5</sup>Centro de Educação Superior do Oeste, UDESC, Chapecó, SC, Brasil.

This study aimed to evaluate the changes in milk fatty acids (FA) composition in ewes fed increasing doses of UnCLA (29.9% of trans-10, cis-12 CLA as methyl esters). Twenty-three primiparous Lacaune ewes with 40 ± 10 DIM, milk yield of 1.73 ± 0.26 kg/d and 52.1 ± 5.0 kg of BW were fed the following dietary treatments during 14d in an 28-d experimental period: Control (C): 30g of Megalac-E, n = 5; T10: 20g of Megalac-E plus 10g of UnCLA, n = 6; T20: 10g of Megalac-E plus 20g of UnCLA, n = 5 and T30: 30g of UnCLA; n = 7. The fat supplements were mixed into the concentrate (1.0 kg/d) and fed individually in 2 equal meals after a.m. and p.m. milkings. Ewes grazed paddocks of a tropical pasture as the only source of forage. Milk samples were collected on the 14th d of experimental period and analyzed for FA profile. Data were analyzed as a completely randomized design using the REG procedure of SAS. The desaturase indexes and the concentration of ≤C16 FA were linearly decreased, whereas the concentration of >C16, trans-10 C18:1 and the sum of trans-C18:1 FA was linearly increased as the CLA dose increased (Table 1). Moreover, treatments T10, T20 and T30 resulted in 191, 445 and 745% increases in milk trans-10, cis-12 CLA content and 131, 320 and 424% increases in trans-10, cis-12 CLA secretion into milk fat, respectively. The transfer efficiencies of trans-10, cis-12 CLA from diet into milk were 2.16, 1.97 and 1.63% (SE = 0.16;  $P = 0.39$ ) for T10, T20 and T30, respectively. Incremental inclusion of UnCLA changed in a linear manner the milk fatty acid profile to a greater proportion of trans monounsaturated, longer chain fatty acids.

**Table 1.** Milk fatty acid responses to increasing levels of UnCLA

g/100 g FA	C	T10	T20	T30	SE	P <sup>1</sup>
<b>Summary</b>						
<C16	32.0	29.3	26.3	27.9	0.69	0.018
C16 + C16:1	27.4	24.5	24.1	23.2	0.47	0.001
>C16	40.5	46.0	49.5	48.7	1.01	0.001
<b>Desaturase Index</b>						
14:1/14:0+14:1	0.010	0.008	0.008	0.006	0.001	0.001
16:1/16:0+16:1	0.025	0.025	0.022	0.020	0.001	0.017
18:1/18:0+18:1	0.559	0.532	0.501	0.473	0.010	0.001
CLA/18:1 t11+CLA	0.319	0.271	0.258	0.256	0.009	0.010

<sup>1</sup>Significance of linear responses. Quadratic responses were not significant ( $P > 0.05$ ).

**Key Words:** CLA, dairy ewes, milk fatty acid composition

**T283 Effect of monensin and tallow on methane estimation and protozoan and bacterial populations in dairy cows rumen.** A. R. Castillo-Gonzalez<sup>\*1</sup>, M. E. Burrola-Barraza<sup>1</sup>, J. A. Ortega-Gutierrez<sup>1</sup>, M. I. Rivas-Martinez<sup>2</sup>, and A. Chavez-Martinez<sup>1</sup>, <sup>1</sup>Facultad de Zootecnia y Ecología, Chihuahua, Chihuahua, México, <sup>2</sup>Colegio de Postgraduados, Texcoco, Edo. de México, México.

The objective was to evaluate the effect of monensin and/or tallow in the diet of lactating cows on the populations of protozoa and bacteria and the estimation of methane. The ruminants have a significant contribution to the greenhouse gases, due to the production of methane. Recent efforts have been made to explore the effect of different additives (monensin, tallow, etc) on rations and its effect on rumen's microbial populations responsible of methanogenesis. The basal diet was formulated in a ration F:C (Forage:Concentrate) of 40:60. The treatments were: The control diet without additive (T1), control diet plus monensin (3.3g/d, T2), control diet plus tallow (3.0%, T3) and control diet plus monensin and tallow (3.3 g/d, 3.0%, T4). The animals were fed 2 times daily (0800 and 1500 h) and milked twice a day (0400 and 1300 h). An experimental design of 4 × 4 Latin square were used. Samples were collected from 13 to 15 d. Quantification of protozoa, bacteria and VFA's (to estimate the production of methane with the Wollin equation) were conducted. Data were analyzed using PROC MIXED of SAS; means comparison was made using orthogonal contrasts. Results showed that protozoan population exhibited changes upon the inclusion of different additives to rations ( $P \leq 0.06$ ) among treatment, the combination of monensin and tallow resulted in the lowest protozoan population size ( $P \leq 0.0001$ ) with an average of  $5.49 \pm 0.07 \text{ Log}_{10}$ . Bacteria population size did not change ( $P \geq 0.83$ ); however, it was observed a reduce on the population size from 1.3 to 0.68 fluorescence unity (FU). Meanwhile, methane estimate was reduced ( $P \leq 0.05$ ), from  $57.3 \pm 1.1 \text{ mmol/mL}$  in the control diet (T1) to  $53.2 \pm 1.1$ ,  $55.4 \pm 1.1$  and  $54.4 \pm 1.1 \text{ mmol/mL}$  in T2, T3 and T4, respectively. In conclusion, the most efficient additive to reduce methane production was monensin (3.3%).

**Key Words:** additives, ionophores, ruminal microorganisms

**T284 Hepatic transcriptomics in dairy cows supplemented with SmartamineM or MetaSmart during the peripartur period.** J. S. Osorio<sup>\*1</sup>, P. Ji<sup>1</sup>, S. L. Rodriguez-Zas<sup>1</sup>, D. Luchini<sup>2</sup>, R. E. Everts<sup>1</sup>, H. A. Lewin<sup>1</sup>, J. K. Drackley<sup>1</sup>, and J. J. Loo<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, <sup>2</sup>Adisseo, Alpharetta, GA.

We used a newly-developed bioinformatics tool (Dynamic Impact Approach, DIA; M. Bionaz, P. Kathiravan, S. Rodriguez-Zas, W. Hurley,

and J. Loo, PLoS One; <http://dx.doi.org/10.1371/journal.pone.0032455>) that allows visualizing the dynamic adaptations of biological pathways to evaluate the impact of Methionine (M) supplementation on liver transcriptome. Twenty 4 multiparous Holstein cows were fed a control diet (ME, n = 8; 1.47 Mcal/kg DM prepartum and 1.67 Mcal/kg DM postpartum), ME plus MetaSmart (MS, n = 8; Adisseo France S.A.S.), or ME plus Smartamine M (SA, n = 8; Adisseo France S.A.S.). All cows received a common diet (1.24 Mcal/kg DM) during the far-off period [-50 to -21 d in milk (DIM)]. Treatments started at -21 DIM and continued through 30 DIM. MetaSmart (0.19% of DM prepartum and 0.18% of DM postpartum) and SA (0.07% of DM prepartum and postpartum) were top-dressed on the ME diet. Percutaneous liver biopsies for microarrays using a 13,000-gene oligonucleotide microarray were performed at -10, 7, and 21 DIM. Analysis of variance with a false discovery rate (FDR) correction resulted in 2,664 differentially expressed genes (DEG) with a treatment × day (T × D) interaction (FDR < 0.10, uncorrected  $P = 0.02$ ). For the DIA analysis the whole data set with Entrez gene IDs, FDR, fold-change, and post-hoc P value between the 3 treatments at each time point were uploaded. A cut-off of FDR = 0.05 and p-value = 0.05 was applied during analysis. The greatest degree of change in terms of up- and downregulated DEG due to T × D occurred with MS vs. ME, e.g., more than half DEG were downregulated by MS at -10 but at 7 DIM ca. 500 DEG were downregulated and > 300 downregulated. The pattern at 21 DIM resembled closely that at -10 DIM. The DIA analysis revealed that MS vs. ME led to an inhibition of cyanoamino acid metabolism at -10, 7, and 21 DIM but it activated taurine/hypotaurine metabolism at 7 DIM. Another novel feature of MS vs. ME was the activation at 7 DIM of riboflavin and nicotinamide metabolism and pantothenate (CoA) biosynthesis. Analysis of DEG due to T × D within treatments revealed that carbohydrate metabolism was among the top-impacted pathways due to ME and SA particularly at 7 vs. -10 DIM when flux of gluconeogenesis and TCA cycle were markedly activated. In contrast to cows fed MS, both ME and SA led to marked activation of glutathione metabolism at 7 vs. -10 DIM. Overall, results from the bioinformatics analysis seem to suggest that the 2 sources of M elicit distinct effects on the liver transcriptome. Such adaptations might be of biological relevance in terms of liver function, dry matter intake, and optimal milk production.

**Key Words:** systems biology, pathway analysis, genomics

**T285 Production response of lactating dairy cows in a confinement operation to a commercial probiotic.** J. K. Bernard<sup>\*</sup> and N. A. Mullis, University of Georgia, Tifton.

Thirty-six lactating Holstein cows from the Dairy Research Center at the University of Georgia -Tifton Campus were used in a 10 wk randomized design trial to determine the effects of feeding a commercial probiotic (ProDairy, Donaghys Industries Ltd., Christchurch, New Zealand) on dry matter intake, milk yield and milk composition of lactating Holstein cows. The probiotic is a blend of non-viable lactobacillus species and fermentation extracts including amino acids, vitamins, amylase, and cellulose. During the first 2 wk of the trial all cows were fed the control diet and data collected were used as a covariate in the statistical analysis. At the end of wk 2, cows were assigned randomly to one of 2 treatments (control (CONT) or supplemental probiotic (PRO) for the following 8 wk. A basal diet was fed to cows once daily behind Calan gates as a TMR in amounts to provide at least 5% refusal. PRO was added to the TMR at a rate of 10 mL/cow/d and mixed for 10 min before feeding. There were no differences in DMI between treatments; 25.3 and 25.4 kg/d for CONT and PRO, respectively. Cows fed PRO have increased yield (kg/d) of milk ( $P = 0.001$ ), protein ( $P = 0.05$ ), and SNF ( $P = 0.002$ ) compared with CONT; 32.7, 0.94 and 2.74 and 30.9, 0.91,