RUMINANT NUTRITION VI

0655 Effect of rumen-protected lysine supplementation of corn-protein based diets fed to lactating dairy cows. N. E. Lobos^{*1}, G. A. Broderick², and M. A. Wattiaux³, ¹Dep. of Dairy Science, University of Wisconsin–Madison, Madison, ²Broderick Nutrition & Research, LLC, Madison, WI, ³University of Wisconsin–Madison, Madison.

This trial tested whether rumen-protected Lys (RPL) supplementation would improve the nutritive value of RUP from corn protein. Thirty-two lactating Holstein cows were blocked by DIM and parity into eight squares of four cows in replicated 4 \times 4 Latin squares. Treatments were all supplemental CP from: 1) Soy [67% expeller soybean meal (ESBM) plus 33% solvent soybean meal (SSBM)]; 2) Soy/Corn [33% ESBM, 17% SSBM, 25% corn gluten meal (CGM) plus 25% distillers dried grains plus solubles (DDGS)]; 3) Corn (50% CGM plus 50% DDGS); or 4) Corn/RPL [diet 3 top-dressed with RPL (125 g Ajipro/d, an estimated 20 g absorbed Lys/d)]. Diets contained (DM basis) 22% alfalfa silage, 43% corn silage, 18% ground high moisture and dry corn, 2.4% mineral-vitamin premix, 1.5-3.9% soyhulls, 15% CP, 30-32% NDF, and, as predicted by NRC (2001), equal RDP, RUP and metabolizable protein. Cows within squares were randomly assigned to treatment sequences and fed diets for 4-wk periods before switching; data from the last 2 wk were analyzed using the PROC MIXED of SAS. The Table 0655 reports LS-means. Intake was highest on diet 1, intermediate on diets 2 and 3, and lowest on diet 4; BW change was highest on diet 3, intermediate on diets 1 and 2 and lowest on diet 4. Intakes and BW changes were reflected by differences in Milk/DMI, which were highest on diets 2 and 4 and lowest on diet 3. Milk yield was lower on diet 3 than on diets 1, 2, and 4, and protein yield was highest on diets 1 and 2, intermediate on diet 4, and lowest on diet 3. These results indicated that dilution of soybean meal RUP with that from corn protein did not reduce milk yield and adding RPL to the corn-protein based diet increased milk and protein yields.

Key Words: soybean meal, corn gluten meal, corn distillers dried grains, rumen-protected Lys

 Table 0655. Effect of Dietary CP Source and Rumen-Protected Lys on

 Production

Item	Soy	Soy/Corn	Corn	Corn/ RPL	SE	P > F
DMI, kg/d	27.7ª	27.4 ^{ab}	26.9 ^{bc}	26.8°	0.40	< 0.01
BW change, kg/d	0.03^{bc}	0.59 ^{ab}	0.70^{a}	-0.07°	0.23	0.04
Milk, kg/d	45.8ª	46.1ª	44.3 ^b	45.4ª	1.17	0.01
Milk/DMI	1.66 ^{ab}	1.69ª	1.65 ^b	1.69ª	0.038	0.04
Fat, kg/d	1.87	1.87	1.83	1.83	0.060	0.37
True protein, kg/d	1.36 ^a	1.34ª	1.25°	1.30 ^b	0.029	< 0.01
MUN, mg/dL	10.6	10.6	10.8	11.1	0.25	0.06
abc (P < 0.05)						

 $^{abc}(P < 0.05)$

0656 Effects of a rumen protected lysine (AjiPro-L) supplementation on peripartum disease, reproduction, and lactational performance of dairy cows. J. E. Nocek^{*1}, A. Haruno², M. Miura², T. Takagi², I. Shinzato³, and T. Fujieda², ¹Spruce Haven Farm and Research Center, Auburn, NY, ²Ajinomoto Co., Inc.,, Tokyo, Japan, ³Ajinomoto Heartland, Inc., Chicago, IL

We used 108 multiparous cows to examine the effects of feeding AjiPro-L (Ajinomoto Co., Inc., Tokyo) 21 d pre- through 21 d postpartum and then withdrawal. Cows were assigned to one of four pre/postpartum regimens: a) Control:Control (C/C), b) 100 g AjiPro-L:Control (A/C), c) Control:150 g AjiPro-L (C/A), and d) 100 g AjiPro-L:150 g AjiPro-L (A/A). All cows started their treatment regime 21 d before expected calving date through 21 d postpartum (phase 1). Upon completion of the 21 d postpartum period, all cows were moved to a common group and fed the same diet without AjiPro-L (phase 2, 21-63 DIM). Individual DMI and milk yield were measured daily, and milk components weekly throughout both phases. Body weights, BCS, health incidence were recorded. During phase 1, DMI was not affected in the prepartum period by AjiPro-L inclusion. Postpartum, cows on A/C consumed more (P = 0.05) DM than C/C and C/A, with A/A cow not being different. Milk yield was highest (P = 0.04) for cows receiving A/A compared to C/C, with other treatments not being different. Milk fat yield was higher (P = 0.04) for cows supplemented with A/A compared to C/C and C/A, but not different from A/C. Cows receiving the C/A had higher milk protein percentage than A/C and A/A and not different from C/C. Milk fat percentage and milk protein yields were not affected by treatment regimens. During phase 2, no differences in body weight, BCS or milk yields were observed. Milk fat yield was higher (P = 0.05) for A/C and A/A cows compared to C/A with C/C not being different. Milk protein percentage was higher (P = 0.02) for C/A cows than those on C/C or A/A regime, with A/C not being different. Cows on A/C had higher (P = 0.05) milk fat percentage than C/C and C/A, not being different than A/A. Cows on A/C exhibited lower incidences of displaced abomasum and ketosis than other regimens. These results suggest that AjiPro-L supplementation both pre- and postpartum generally resulted in the most consistently positive production performance immediate postpartum than other regimens. It was also suggested that feeding AjiPro-L only prepartum had some specific effects on postpartum health status and production performance.

Key Words: lysine, milk production, transition period

0657 Effect of strategic ration balancing with use of Prolak and USA-Lysine on the efficiency of milk protein production and environmental impact. J. H. Harrison¹, J. Jarrett², Y. Chen^{*3}, L. VanWieringen⁴, B. Chalupa⁵, F. Sun³, P. Ndegwa³, D. Wilks⁶, and H. S. Joo³, ¹Washington State University, Puyallup, ²Prince Agri, Quincy, IL, ³Washington State University, Pullman, ⁴Washington State University, Sunnyside, ⁵University of Pennsylvania, New Bolton Center, ⁶EPL Feeds, Dixie, WA.

The objective of this study was to evaluate the effect of a diet supplemented with a high quality protein and rumen protected lysine source on the efficiency of milk production and environmental impact in a commercial dairy herd. The general herd diet was reformulated with the Agricultural Model and Training System model and use of Prolak and USA-lysine. The control and treated diet had similar dietary CP concentration (17.7% vs. 17.6%). Cows were completely randomized to two groups with 163 cows each pen and milked three times per day. The groups had similar average days in milk (135 vs. 135) and parities (2.99 vs. 2.84) before initiation of the trial. Respective diets were fed in a 40 d switch back design trial with two periods. Milk weight (all three milkings), and milk (sample from one of three milkings) and manure samples were obtained at the end of second and third weeks of each period for analysis. Cows fed the two diets had similar pen DMI (24.2 vs. 24.1 kg/d), but the reformulated diet supported more milk yield (43.0 vs. 44.9 ± 0.15 kg/d), milk protein yield $(1.27 \text{ vs. } 1.33 \pm 0.01 \text{ kg/d})$, milk fat yield $(1.51 \text{ vs. } 1.57 \pm 0.01 \text{ kg/d})$ kg/d), and increased ratio of milk true protein to total protein intake (29.4 vs. 31.2%) and ratio of total milk protein to total protein intake (34.3 vs. 36.5%). The concentration of MUN was higher when cows were fed reformulated diet (16.5 vs. 18.7 mg/dl). Cows fed the reformulated diet consumed 2.6% less N, but produced 4.7% more milk N, and excreted 6.2% more predicted urinary N and 24% less calculated fecal N. Manure from cows fed the reformulated diet had lower ammonia (NH3) flux (145 vs. 130 mg*h-1*m-2). This study illustrates that diets supplemented with high quality protein could improve efficiency of milk production and N utilization, and reduce the environmental impact.

Key Words: protected lysine, bypass protein, milk production

0658 Effect of strategic ration balancing with use of Prolak and MetaboLys on the efficiency of milk protein production and environmental impact. P. Ndegwa¹, J. H. Harrison², D. Wilks³, L. VanWieringen⁴, Y. Chen^{*1}, W. Chalupa⁵, F. Sun¹, and H. S. Joo¹, ¹Washington State University, Pullman, ²Washington State University, Puyallup, ³EPL Feeds, Dixie, WA, ⁴Washington State University, Sunnyside, ⁵University of Pennsylvania, New Bolton Center.

The objective of this study was to evaluate the effect of reduced dietary CP concentration on efficiency of N utilization and environmental impact in a commercial dairy herd. The general herd diet was reformulated with the Agricultural Modeling and Training System model and use of Prolak and MetaboLys. Reformulation reduced the dietary CP concentration (17.4 vs. 15.9%) and resulted in a DCAD of 31.3 vs. 21.2 mEq/100 g DM. Cows were completely randomized to two groups with 155 cows each and milked three times per day, and the two groups had the same average days in milk (DIM = 149) before initiation of the study. Respective diets were fed in a 40-d switch back design trial with two periods. Milk weight, milk, and manure samples were obtained at the end of the second and third week of each period. The manure samples were incubated in closed chambers for 19 d to measure ammonia (NH3) flux. Average pen DMI was similar (25.1 vs. 25.3 kg/d). The reformulated diet supported 0.6 kg less milk production (44.3 vs. 43.7 ± 0.16 kg/d), less milk protein yield (1.27 vs. 1.25 ± 0.01 kg/d) and the same amount of fat yield (1.48 vs. 1.46 ± 0.01 kg/d), but increased the ratio of true protein/total CP intake (28.4 vs. 30.6%) and milk CP/total CP intake (33.3 vs. 38.6%). The concentration of MUN was lower for cows fed the reformulated diet (13.4 vs. 11.56 mg/dl). When the cows were fed reformulated diet, they consumed 9% less N, produced 1% less milk N, but excreted 14% less predicted urinary N and 15.4% less calculated fecal N. The manures from the reformulated diet had a lower NH3 flux (147 vs. 137mg*h ¹*m⁻²). This study illustrated that the lower N diet could improve the efficiency of N utilization and reduced the environmental impact, but may influence the milk production.

Key Words: milk protein, lysine, rumen undegradable protein

0659 Evaluation of diets formulated with soybean-based products, blood meal, or rumen-protected lysine to meet MP lysine demands of lactating dairy cows.
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Objectives were to determine if production of early to midlactation dairy cows was maintained when fed diets formulated to be similar in grams of metabolizable protein (MP) lysine and methionine but lower in CP. Diets were formulated using: 1) soy-based protein (positive control; PC, 17.7% CP, 204 g/d MP-lys), 2) blood meal and soy-based protein (BM; 16.4% CP, 203 g/d MP-lys), or 3) rumen-protected lysine and soy-based protein (RPL; 16.2% CP, 205 g/d MP-lys). Diets formulated to desired MP lysine (204 g/d) were compared with a negative control diet lower in MP lysine (LL; 16.2%) CP, 193 g/d MP-lys) created by removing dietary RPL. Diets were formulated using AMTS (Version 3.4.7.1). Sixteen Holstein cows blocked by parity (12 multiparous) and production were assigned one of four treatments arranged in a 4×4 Latin square design with 28-d periods. The model included: treatment, square, treatment \times square, and period. Treatments were prepared as four separate concentrate mixes that were combined with common amounts of corn silage (36% of diet DM) and alfalfa hay (19%) among diets. Milk production was similar (P > 0.10), comparing BM and RPL with PC or LL, but tended (P = 0.06) to be greater for BM over RPL. Cows fed BM and RPL had lower (P = 0.04) DMI than LL; however, ECM was sustained which led to an increase in feed efficiency comparing BM and RPL with LL. Performance of lower CP diets, BM, and RPL, were similar to PC, but performance from LL suggests that MP supply was sufficient across treatments.

Key Words: amino acid, lysine, metabolizable protein

Table 0659.

Item	PC	BM	RPL	LL	SEM	Contrast ¹
DMI, kg/d	26.16	25.64	25.46	26.66	0.61	В
Milk, kg/d	39.47	40.44	38.81	39.50	1.08	d
ECM	39.35	40.43	39.37	39.88	1.36	-
ECMFE	1.53	1.59	1.59	1.52	0.05	a, B
Fat, %	3.47	3.54	3.59	3.55	0.12	-
Fat, kg/d	1.35	1.42	1.39	1.40	0.06	-
Protein, %	3.17	3.06	3.10	3.13	0.08	A, b
Protein, kg/d	1.25	1.23	1.20	1.23	0.05	-
MUN, mg/dL	16.59	13.92	14.03	13.46	0.33	A, b

¹ Contrasts: A = PC vs. BM + RPL; B = BM + RPL vs. LL; D = BM vs. RPL. Uppercase = P < 0.05, lowercase = 0.05 < P < 0.10.

0660 The plasma free amino acid dose response technique: A proposed approach for determining lysine bioavailability of ruminally protected lysine products. N. L. Whitehouse^{*1}, A. F. Brito¹, and C. G. Schwab², ¹University of New Hampshire, Durham, ²Schwab Consulting, LLC, Boscobel, WI.

While most companies provide estimates of lysine (Lys) bioavailability for their products, values are often obtained with different techniques, and there are little to no data comparing efficacy and cost-effectiveness of different products. Our objective is to propose that the plasma free amino acid (AA) dose–response technique be accepted as the standard-ized method for evaluating rumen protected Lys (RP-Lys) products, because its animal-derived estimates of bioavailability are obtained under conditions similar to commercial use. Bioavailability estimates are calculated by dividing the

slope of the regression line relating changes in plasma Lys to increased feeding of RP-Lys divided by the slope of the Lys infusion regression line. Eleven dose-response Latin square trials using 66 lactating, ruminally cannulated multiparous Holstein cows (d in milk = 60 through 315; milk yield = 25 to 62 kg/d at the start of the study) were conducted. Abomasally infused and fed amounts of Lys ranged from 0 to 84 g/d and periods from 4 to 21 d. The RP-Lys products were mixed with 1 kg of total mixed ration, placed in tubs, and fed 30 min before each of the three daily feedings. Product not consumed within 20 min was delivered via the ruminal cannula. Diets were formulated (NRC, 2001) to be adequate in metabolizable protein-Met but varied in predicted metabolizable protein-Lys (5.04-6.81%). One to four blood samples were taken from the tail vein for 1 to 3 d in each period. Basal plasma Lys concentrations ranged from 2.14 to 5.62% of total AA without infusions, from 2.33 to 5.67% of total AA for fed RP-Lys, and from 2.94 to 8.62% of total AA with infusions. Results corroborate previous research that showed a positive linear relationship between both infused and fed RP-Lys and plasma Lys concentrations. Regression analysis (by trial and cow) indicated the two factors that most affected the magnitude of the response slopes, and hence the technique's precision, were the cows basal level of plasma Lys (i.e., intercept, the lower the better) and the cows plasma Lys response to infused Lys (measured by the magnitude of their response to the highest level of infused Lys). Stage of lactation and milk production did not affect plasma Lys response. It is concluded that the plasma free AA dose response technique is sensitive to increasing amounts of absorbed Lys and therefore is an appropriate technique for evaluating RP-Lys supplements.

Key Words: technique, bioavailability, lysine

0661 Effects of maternal nutrition and rumen-protected arginine supplementation on pregnant and non-pregnant ewe and postnatal lamb serum amino acids. J. L. Peine^{*1}, G. Jia¹, M. Kapphahn¹, S. T. O'Rourke¹, A. M. Meyer², L. P. Reynolds¹, and J. S. Caton¹, ¹North Dakota State University, Fargo, ²Div. of Animal Sciences, University of Missouri, Columbia, MO.

Our hypothesis was that rumen-protected arginine supplementation would increase levels of serum amino acids in both non -pregnant and pregnant ewes and their offspring, and thereby help overcome negative effects of nutrient restriction during the last two-thirds of gestation. To test this hypothesis, two studies were conducted with Rambouillet ewes penned individually in a temperature-controlled facility: Study 1 was a dose titration study to determine the most effective dose of rumen-protected arginine, and Study 2 tested our hypothesis in pregnant ewes and their lambs. Study 1 used non-pregnant primiparous ewes (n =60) randomly assigned to one of four treatments: a control group receiving no supplement (0), and groups receiving 90mg/kg BW (90), 180mg/kg BW (180), or 360mg/kg BW of rumen-protected arginine supplement (360). After 15 d of supplementation, ewes receiving 180 had greater serum ornithine (P = 0.05) and arginine (P = 0.05), and tended to have greater aspartate (P = 0.08)than ewes receiving 90, and 180 were similar to 360 fed ewes (P > 0.55). In Study 2, multiparous ewes (n = 32) were allocated to trhee treatments at 54 ± 3.9 d of gestation: 100% of requirements (control, CON), 60% of control (restricted, RES), or RES plus a 180 mg/kg BW rumen-protected arginine supplement once daily (RES-ARG). Ewes were maintained on treatments through parturition, when lambs were removed from dams and reared independently. At 128 d of gestation, CON ewes weighed more (P < 0.001) than RES and RES-ARG ewes, and had greater (P \leq 0.001) BCS. In addition, serum ornithine, citrulline, aspartate, arginine, and methionine were greater ($P \le 0.03$) in CON than RES and RES-ARG ewes, suggesting that arginine supplementation could not overcome differences in nutritional plane. Lambs from CON ewes had greater (P = 0.03) birth weights than RES, with lambs from RES-ARG ewes being intermediate and similar in weight to other treatments (P > 0.08). At birth, lambs from RES-ARG ewes had greater serum arginine (P =0.04) and lysine (P = 0.04) than those from RES ewes, suggesting effects on amino acid transport from dam to fetus. These results support our hypothesis that maternal rumen-protected arginine supplementation could increase serum levels of amino acids in offspring. Additionally, arginine supplementation of dams may be able to increase fetal circulating arginine without altering maternal circulating arginine.

Key Words: arginine, amino acids, developmental programming

0662 Intestinal digestibility of amino acids in fluid- and particle-associated rumen bacteria determined using a precision-fed cecectomized rooster bioassay.
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Microbial protein represents the majority of metabolizable protein absorbed by ruminant animals. Enhanced understanding of the AA digestibility of rumen microbes will improve estimates of metabolizable protein. The objective of this experiment was to determine the digestibility of AA in fluid- (FAB) and particle-associated bacteria (PAB) using the precision-fed cecectomized rooster bioassay. Bacteria were isolated from four ruminally cannulated lactating Holstein cows fed an 89% forage diet (DM basis). Samples of FAB and PAB were fed to nine cecectomized roosters to determine standardized digestibility of AA. Data were analyzed using PROC MIXED (SAS, 2004). Bacterial N composition (average = 8.3%) was similar to previous literature values. Total AA digestibility was (mean \pm SE) 76.8 \pm 2.4 and 75.5 \pm 2.2% for FAB and PAB, respectively. Amino acid digestibility did not differ between FAB and PAB (P > 0.05). There were differences in essential (EAA) and non-essential AA digestibility within bacterial type when compared with mean essential or non-essential AA digestibility values (P < 0.05). Arginine, His, and Met were greater than the mean EAA digestibility in FAB, whereas Trp and Val were lower. Arginine and Lys were greater than the mean EAA digestibility in PAB, whereas Phe and Val were lower. Compared with previous literature and relative to NRC (2001) estimates of AA digestibility in microbes, the precision-fed cecectomized rooster assay is an acceptable in vivo model to determine AA digestibility of rumen bacteria.

Key Words: amino acid, digestibility, rumen bacteria

0663 Performance by holstein steers offered hay and supplement with or without added methionine.
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Recently, amino acid supplementation by forage-fed cattle has been examined in greater detail. Methionine can restrict productivity of many ruminant animals, chiefly because microbial protein, the main source of metabolizable protein in forage-based ruminant diets, is most limiting in methionine. The objective of this study was to evaluate steer performance when offered hay and a supplement with or without added methionine as MFP (MFP). On Oct. 24, 2013, a total of 90 $(230 \pm 2.2 \text{ kg body weight})$ Holstein steers were stratified by body weight within five blocks and were allocated randomly to one of two treatments: 1) control supplement (C; 15 replications) or control supplement plus MFP (15 replications). Each replication had access to a 0.4-ha pasture with limited forage available for grazing and were offered ad libitum access to medium quality hay, water, and shelter. A soybean hull and wheat middling based, pelleted supplement was offered daily at 0.5% of body weight for each replication; in addition the supplement contained minerals, and vitamins. Treatment was provided at 1.17% of supplement DM resulting in an average intake of approximately 15 g/d of MFP. Hay offered did not differ (P = 0.62) across treatments. Initial, d 14, and d 28 body weights did not differ ($P \ge 0.11$) across treatments, but d 42 and d 56 body weights were greater ($P \le 0.05$) from pastures receiving MFP compared with C. Average daily gain and gain at d 14 did not differ (P > 0.27) across treatments. However, d 28 ADG and gain tended (P < 0.07) to be greater from MFP compared with C and were greater ($P \le 0.03$) at d 42 and d 56 from MFP compared with C. Through 56 d, steers in pastures offered MFP gained 5.0 \pm 2.1 kg/hd more (P < 0.03) body weight than steers fed C. It is concluded that, when steers were offered medium quality hay, providing a supplement that contained MFP showed improved body weights, ADG, and total gain through the backgrounding phase.

Key Words: methionine, backgrounding, steers

0664 Effects of feeding slow release NPN and microbial fermentation extracts on lactation performance of high-producing dairy cows. F. Díaz-Royón^{*1}, A. D. Garcia¹, K. F. Kalscheur², and K. Mjoun³, ¹Dairy Science Dep., South Dakota State University, Brookings, ²South Dakota State University, Brookings, ³Alltech, Brookings, SD.

The objective of this study was to examine the effects of partial substitution of soybean meal with a product containing slow release NPN and microbial fermentation extracts [(OPT); Optimase Alltech Inc., Nicholasville, KY)] in diets with two forage concentrations. Sixteen lactating Holstein dairy cows (four primiparous and 12 multiparous) were randomly assigned to a 4×4 Latin square in a 2×2 factorial. High and low forage diets contained respectively 61% (HF) and 46% forage (LF), with (O) and without OPT (NO). Forage consisted of 75% corn silage and 25% alfalfa hay (DM basis). Experimental diets containing OPT (125 g/cow/day) were designed to partially replace soybean meal 48 (0.625 kg) with forage fiber [corn silage (0.225 kg) and alfalfa hay (0.075 kg)] and non-forage fiber [soybean hulls (0.200 kg)]. Feeding cows HF versus LF reduced DMI, milk, ECM, protein %, and protein yield, but increased fat % (Table 0664, P < 0.05). Feeding OPT decreased (P = 0.033) DMI 1.05 kg/d compared to diets without OPT. Milk yield decreased (P =0.046) slightly with OPT inclusion; however, 4%FCM and ECM were not affected. Feeding OPT did not affect milk fat percentage and yield, lactose percentage, and total solids percentage and yield. Milk protein yield decreased (P = 0.013) 3.5% in cows fed OPT. OPT fed both in HF and LF diets as a partial replacement to soybean meal resulted in similar lactation performance (4% FCM and ECM) and feed efficiency despite a decrease in DMI and milk yield.

Key Words: slow-release-NPN, microbial fermentation extracts, dairy cows

Table 0664.

	HF		L	LF		P value ¹	
Item	NO	0	NO	0	SEM	F	0
DMI, kg/d	26.4	25.5	29.5	28.3	0.76	< 0.001	0.033
Milk, kg/d	41.2	40.0	43.5	42.5	1.44	< 0.001	0.046
4% FCM, kg/d	42.3	40.9	43.0	42.7	1.25	0.165	0.362
ECM, kg/d	44.8	43.1	46.1	45.6	1.25	0.038	0.247
Fat, %	4.22	4.20	3.96	4.06	0.13	0.024	0.648
Fat, kg/d	1.72	1.66	1.71	1.72	0.06	0.681	0.590
Protein, %	3.07	3.00	3.14	3.14	0.05	< 0.001	0.058
Protein, kg/d	1.25	1.19	1.35	1.32	0.03	< 0.001	0.013
ECM:DMI	1.71	1.70	1.57	1.61	0.04	0.001	0.624

¹ There were no interactions forage by OPT.

0665 Concentration of soluble non-ammonia nitrogen and related transporter expression in nonmesenteric gastrointestines of dairy cows. Y. M. Xie¹, Q. B. Xu^{*1}, Y. M. Wu¹, and J. X. Liu², ¹Institute of Dairy Science, Zhejiang University, Hangzhou, China, ²Zhejiang University, Hangzhou, China.

Dietary protein is gradually degraded from peptide-bound amino acids (PBAA) into free amino acids (FAA) and ultimately into ammonia by the rumen microbes. Both PBAA and FAA are milk protein precursor, and the rumen and small intestines are the main sites where milk protein precursor is produced and absorbed. This work was designed to investigate the concentrations of PBAA, FAA and soluble protein and the expression of peptide transporter (PepT-1) and amino acid transporters in the rumen, omasum, and duodenum of dairy cows. The digesta and tissues were collected from six healthy Holstein dairy cows (BW = 643 ± 67.4 kg and milk yield = 13.4 \pm 2.05 kg/d) immediately after the animals were slaughtered. The FAA was assessed by amino acid (AA) analyzer, PBAA was by quantification of AA before and after acid-hydrolysis by 6M HCl, and soluble protein was by bicinchoninic acid. Concentrations of FAA, PBAA, and soluble protein are shown in Table 0665. Concentration of all portions of non-ammonia nitrogen was the highest in the duodenum. The PBAA was the largest component of soluble non-ammonia nitrogen in all the digesta from rumen, omasum, and duodenum, indicating that peptides may be the main form absorbed in these sites. Abundance of PepT-1 mRNA was consistent with the concentrations of PBAA from rumen to duodenum. Expression of all genes was greater in the duodenum than in the rumen and omasum, suggesting that the duodenum is the major nonmesenteric site where peptides are transported and absorbed.

Key Words: gastrointestine; milk protein precursor; peptide-bound amino acids

Table 0665. Concentrations of soluble non-ammonia nitrogen (mg N/100ml) in rumen, omasum, and duodenum (n = 6)

			Duo-		
	Rumen	Omasum	denum	SEM	P-values
Free amino acid	22.4 ^B	49.9 ^B	189.8 ^A	24.5	< 0.01
Peptide-bound amino acids	40.0 ^B	73.3 ^B	285.5 ^A	35.8	< 0.01
Soluble protein	15.1 ^в	34.6 ^B	168.4 ^A	16.4	< 0.01
Total	77.5 ^в	157.7 ^в	643.6 ^A	65.8	< 0.01
Peptide-bound/Total (%)	51.6	46.5	44.4		

^{A,B} Values with different letters differ significantly (P < 0.01).

0666 Role of proton-coupled oligopeptide transporter **1** in small peptide absorption in the bovine forestomach. Q. B. Xu^{*1}, Y. M. Wu¹, H. Y. Liu¹, and J. X. Liu², ¹Institute of Dairy Science, Zhejiang University, Hangzhou, China, ²Zhejiang University, Hangzhou, China.

In this work, transepithelial transport of glycylsarcosine (Gly-Sar) in bovine forestomach cells was determined to investigate the role of proton-coupled oligopeptide transporter 1 (PEPT1) in small peptide absorption. Primary cultured omasal epithelial cell (OEC) and rumen epithelial cell (REC) monolayers derived from newborn Chinese Holstein male calves were grown in transwell set-up. The transepithelial electrical resistance of transwell model reached plateau after a week, and that of OEC monolayers was higher than that of REC monolayers (2468 \pm 216 vs. 1918 \pm 33 Ω ·cm²). Fluorescein sodium was used to measure the permeability of monolayers. After 150 min, transmittances of fluorescein sodium across OEC and REC monolayers were 0.63 and 0.19%, respectively, whereas that of control (blank filter) was 12.0%. The monolayers were then used for incubation with various concentrations of Gly-Sar in apical side at various pH values or at 37°C and 4°C for various times. In addition, mRNA of PEPT1 was detected in OECs and RECs. The cells were also incubated in absence or presence of 0.5 mM diethylpyrocarbonate, an inhibitor of PEPT1. Transportation of Gly-Sar was dependent on concentration, incubation time, temperature and pH value. Accumulation of Gly-Sar in basolateral side (pH 7.4) increased with the increasing level of Gly-Sar and reached a plateau at 5 mM. Transportation of Gly-Sar by OEC monolayers was higher at 37°C compared to 4°C (P < 0.05), with an optimal pH of 6.0 to 6.5, and inhibited by diethylpyrocarbonate. In addition, accumulation of Gly-Sar in basolateral side of OEC monolayers was greater than that of REC (P < 0.05), indicating that the OECs have greater ability to transport Gly-Sar than RECs. In summary, the PEPT1 plays an important role in small peptide absorption in bovine forestomachs.

Key Words: bovine forestomach epithelial cells, oligopeptide transporter 1, transport