140 Insulin plays a key role in re-coupling the IGF-somatotropin axis in the early postpartum dairy cow. S.T. Butler* and W.R. Butler, *Cornell University, Ithaca, NY.*

Negative energy balance associated with the onset of lactation results in hypoinsulinemia, uncoupling of the IGF-somatotropin axis, attenuation of gonadotropin release and delayed first ovulation. Our objectives were to examine the effects of elevated insulin during the immediate postpartum period on circulating IGF-I concentrations, ovarian follicular growth, estradiol secretion and LH pulse profiles. Holstein cows (n=14) were subjected to either a hyperinsulinemic-euglycemic clamp (INS) or saline infusion (CTL) for 96 hours starting on day 10 postcalving. Blood samples were taken on days 8-9 to establish baseline glucose values. Insulin was infused continuously (1 μg/kg BW/hr) via a jugular catheter. Blood samples were collected hourly, and euglycemia was maintained by infusion of exogenous glucose. During infusion, insulin concentrations were increased 8-fold in INS cows over those in CTL cows (2.4 $\pm$ 0.1 vs. 0.3 $\pm$ 0.1 ng/ml; P<0.001), while blood glucose concentrations were not different between the treatments (45.4 $\pm$ 2.2 vs. 41.7 $\pm$ 2.3 mg/dl; P=0.27). Plasma IGF-I increased continuously during the insulin infusion, and reached the highest values at the end of the clamp, being almost 4-fold higher in INS compared with CTL cows (122 $\pm$ 11 vs. 33 $\pm$ 2 ng/ml; P<0.001). Ultrasound measurements of ovarian follicular growth revealed that 2 cows ovulated, both of which were CTL cows. Among non-ovulatory cows, the dominant follicle reached a greater maximum diameter in the INS cows compared with CTL cows (13.4 $\pm$ 0.5 vs. 11.6 $\pm$ 0.6 mm; P=0.04). Excluding ovulatory cows, no difference in plasma estradiol was observed between groups during the infusion period (1.3 $\pm$ 0.2 vs. 1.1 $\pm$ 0.3 pg/ml; P=0.69). Blood samples collected every 10 minutes for 8 hours prior to and at the end of the infusion periods showed no differences between groups in LH pulse frequency, pulse amplitude or area under the curve. In conclusion, insulin appears to be a key metabolic signal in coupling the IGF-somatotropin axis, orchestrating the observed marked elevation in circulating IGF-I.

Key Words: Insulin, IGF-I, Ovary

141 Postpartum nutrition influences concentrations of leptin, IGF-I, and pregnancy rate of primiparous beef cows. N. H. Ciccioli*[1], R. P. Wettemann[1], L. J. Spicer[1], D. H. Keisler[2], C. A. Lents[1], and F. J. White[1], [1]*Oklahoma Agricultural Experiment Station, Stillwater,* [2]*University of Missouri-Columbia.*

The influence of nutrition on plasma concentrations of leptin and IGF-I and reproductive function was determined at the first estrus in Hereford x Angus spring calving primiparous cows. At parturition, cows (BCS = 4.3 $\pm$ 0.1; BW = 385 $\pm$ 17 kg) were blocked by BCS and calving date and randomly assigned to gain 0.45 (M; n = 17) or 0.90 (H; n=17) kg/d for 11 wk. Then, all cows were fed the same (M) diet until the first estrus. A second replication was added to assess pregnancy rate (M; n= 13; H; n=17). Leptin and IGF-I were quantified weekly and progesterone thrice a week in blood plasma. Estrous behavior was detected with Heatwatch® and ovulation was determined using plasma progesterone. The dominant follicle was measured by ultrasonography at 4 to 14 h after onset of estrus. All cows were AI between 12 and 20 h after onset of estrus. During treatment, H cows gained (P < 0.01) BW and BCS and had greater (P < 0.01) concentrations of leptin and IGF-I compared with M cows. From the end of treatment to first estrus, concentrations of IGF-I were greater (P < 0.01) in H cows; however, concentrations of leptin decreased (P < 0.01) in H cows after treatment termination and did not differ (P = .16) from those of M cows through the first estrus. Cows that exhibited estrus and ovulated on or before 19 wk postpartum had greater (P < 0.06) concentrations of leptin on wk 13 to 15 than anestrous cows. During treatment, leptin and IGF-I concentrations were positively correlated (P < 0.05) with changes in BW and BCS. H cows had a larger (P < 0.05) dominant follicle, more mounts (P < 0.06), and a shorter (P = 0.06) interval to estrus than M cows. Pregnancy rate at the first postpartum estrus was greater (P < 0.01) for H (n=34; 82.3 %) than for M (n=30; 60 %) cows. We conclude that increased nutrient intake after calving results in increased concentrations of IGF-I in plasma, and increased follicle size and pregnancy rate at the first estrus.

Key Words: IGF-I, Leptin, Reproduction

142 Concentrations of leptin and insulin like growth factor-I (IGF-I) during acute nutritionally induced anovulation and realimentation. F.J. White*[1], C.A. Lents[1], N.H. Ciccioli[1], R.P. Wettemann[1], L.J. Spicer[1], and D.H. Keisler[2], [1]*Oklahoma Agricultural Experiment Station, Stillwater,* [2]*University of Missouri, Columbia.*

Luteal activity and concentrations of leptin and IGF-I were evaluated during acute nutritional restriction and realimentation of beef heifers. Angus x Hereford heifers (14 mo old; n=19) were housed in individual pens and fed a diet supplying 1.2 x maintenance (M) for 1 wk. Then heifers were randomly allotted on d 0 to either 0.4 or 1.2 M. Heifers were treated with PGF2α on d -10, 0, and 10 to synchronize ovulation. After 30 d, 0.4 M heifers were gradually increased to 1.2 M during 10 d. Blood was collected 23 h after feeding on alternate days during restriction and 3 x per wk during 100 d realimentation. Heifers with progesterone <0.5 ng/mL for 8 d were classified as anovulatory. Seventy percent (7 of 10) of 0.4 M heifers did not ovulate on d 14 while all 1.2 M heifers had normal luteal function. Plasma IGF-I in .4 M heifers decreased from 49 $\pm$ 3 ng/mL on d 0 to 33 $\pm$ 4 ng/mL on d 14; however, 1.2 M heifers had similar concentrations on d 0 and 14 (57 $\pm$ 3; day x diet effect, P < .01). During restriction, heifers on 0.4 M tended (P = 0.1) to have decreased concentrations of leptin (14 % less) in serum compared with 1.2 M. Five of the anovulatory heifers had luteal activity by 16 to 51 d of realimentation at 1.2 M (mean = 35 d); however, two heifers did not ovulate by 100 d. Realimentation with 1.2 M for 2 wk increased concentrations of IGF-I in 0.4 M heifers (day x diet effect, P < 0.05) to concentrations similar to 1.2 M heifers. During realimentation, concentrations of leptin in blood samples taken after 23 h of fasting were similar (P > 0.1) in 0.4 and 1.2 M heifers. Concentrations of IGF-I in plasma decreased with acute nutritionally induced anovulation and were similar to those in control heifers (1.2 M) by 2 wk of realimentation at 1.2 M. Systemic concentrations of IGF-I were more indicative of nutritional status than concentrations of leptin in these acute nutritionally restricted heifers.

Key Words: Beef heifer, IGF-I, Leptin

ASAS/ADSA Ruminant Nutrition: Feed Additives

143 Influence of length and ramification of the alcohol radical of esters of methionine and of 2-hydroxy-4 (methylthio) butanoic acid on methionine bioavailability. J.C. Robert*[1], B.K. Sloan[2], G. Etave[1], and B. Bouza[1], [1]*Aventis Animal Nutrition, Antony, France,* [2]*Aventis Animal Nutrition, Alpharetta, USA.*

In a series of five experiments, Methionine bioavailability from 8 DLmethionine and 3 HMB esters were tested according to the blood kinetics method based on determining the Area Under the Curve (AUC). The alcohol radical varied in number of carbons but all were arranged in a linear form. Smartamine™ M (a methionine coated with a pH sensitive polymer based coating) of which the methionine bioavailability is 80%, was used as a reference. Non lactating rumen cannulated Holstein cows, receiving 10 kg / animal / day of a ration comprising 75% hay and 25% concentrate delivered in equal quantities twice a day, were used. A single dose, 50 g of methionine equivalent, was supplied directly into the rumen at 0800 on day two (D2) for the esters and at 1600 on day one (D1) for Smartamine™M. Blood samples were obtained, on D2 at 0900,1000, 1100, 1300, 1500h and, thereafter, every three hours from 0900 to 1500h on D3 and D4. For Smartamine™M, blood samples were collected every two hours on D2, starting at 0600 until 1000h and, thereafter, every three hours on D3 and D4 from 0600 until 1500h. Blood plasma methionine concentration (BPMC mg/100g) for base line determinations were measured on D1 at 0900,1100 and 1500h. Modelization of the AUC results for BPMC for the 11 esters resulted in the following relationship : Y = 40.6787 exp.(-0.2272 X)(R^2 = 0.89)(SED =6.13) Y = bioavailability as a percentage of methionine equivalent ingested. X = alcohol carbon number. It shows that methionine bioavailability decreases with increasing number of carbons in the alcohol. Three esters with branched alcohol radicals were tested vs. their corresponding linear forms using the same methodology described above. In all cases, methionine bioavailabilities with the corresponding branched alcohol were higher.

Products	Alcohol carbon numbers	Branching	Methionine bioavailability (%)
Methionine n-propyl ester	3	1	20.7
Methionine isopropyl ester	3	2	43.9
Methionine n-butyl ester	4	1	7.9
Methionine sec.-butyl ester	4	2	28
HMB n-butyl ester	4	1	17
HMB sec.-butyl ester	4	2	31

Key Words: Methionine, Bioavailability, Chemical derivatives

144 Investigation of the site of absorption and metabolism of a novel source of metabolisable methionine: 2 hydroxy 4 (methyl thio) butanoic acid isopropyl ester (HMBi). J.C. Robert*, C. Richard, T. D'Alfonso, N. Ballet, and E. Depres, *Aventis Animal Nutrition, Antony, France*.

In 2 tests, HMBi was delivered in a single dose, 50 g of methionine equivalent directly into the rumen. Two and four non lactating rumen cannulated Holstein cows were used in trials 1 and 2, respectively. They were offered 10 kg/cow/day of a ration comprising 75% hay and 25% concentrate delivered in 2 equal meals. Blood samples were obtained after supplementation at intervals of about 2h except in trial 2 where, during the interval 0-2h, more frequent blood samples were taken. Blood plasma HMB (BPHC) and blood plasma methionine concentrations (BPMC) were measured. In the two tests, a peak of HMB was observed (1h) followed by a methionine peak (4h) relative to ruminal delivery of HMBi . The appearance of HMB and methionine in plasma were found to be best described by a gamma function Y= a + bX exp.(- cX) (Y =BPC and X =hours post-supplementation). In trial 2, more frequent measurements during the 1st hour showed the virtually immediate appearance of HMB in peripheral blood after HMBi supply to the rumen. BPHC, zero before supplementation, rose quickly 1.41-1.73-2.05 mg/100g at times 10,20,30 min after supplementation. In parallel, BPMC increase from 0.35 to 0.45,0.66 and 0.83 mg/100g at the same times. This indicates HMBi is being absorbed through the rumen wall, subsequently dissociates to give HMB which is ultimately transaminated into methionine.

					y =a+b X exp.(- cX)		
Assay	Metabolites	Intervals of time (h)	Peak hours	Max BPC (mg/ 100 g)	Coefficients	R^2	SED
1	HMB	$0 \geq 10$	1.3	1.48	a=0 b=3.13 c=0.7790	0.99	0.08
	Methionine $0 > 25$	4.3	3.26	c=0.231	0.98 c=0.231	0.24	
2	HMB	$0 \geq 24$	1.3	2.64	c=0.7631 c=0.7631	0.78	0.79
	Methionine $0- > 24$	4.4	2.16	a=0.2850	0.93 b=1.1556 c=0.2267	0.22	

Key Words: Methionine, Rumen absorption, Chemical derivative

145 Feeding 2-hydroxy-4-(methylthio)-butanoic acid to transition dairy cows improves milk production but not hepatic lipid metabolism. M. S. Piepenbrink*[1], A. L. Bork[1], M. R. Waldron[1], T. R. Overton[1], M. Vazquez-Anon[2], and M. D. Holt[2], [1]*Cornell University, Ithaca, NY,* [2]*Novus International, Inc., St. Louis, MO*.

Forty-eight Holstein cows entering second or later lactation were utilized to determine the effects of 2-hydroxy-4-(methylthio)-butanoic acid (HMB; Alimet® feed supplement, Novus International, Inc. St. Louis, MO) on milk production and hepatic lipid metabolism during the transition period. Cows were fed one of three diets as TMR starting 21 d before expected calving. These diets contained 0 (CON), 0.1 (+HMB), or 0.21 (++HMB)% HMB. From parturition to 84 DIM, cows were fed diets that contained 0, 0.15, or 0.23% HMB. The CON diets were formulated to be low in Met (1.98 and 1.77% of MP for pre- and postpartum diets) but adequate in Lys (7.23 and 7.00% of MP for pre- and postpartum diets). Prepartum (12.9, 12.8, 12.7 kg/d) and postpartum (18.6, 19.8, 19.7 kg/d) DMI were similar among cows fed CON, +HMB, and ++HMB ($P > 0.20$). Feeding +HMB increased milk yield (42.0, 45.0, and 42.0 kg/d for CON, +HMB, ++HMB; P (quadratic) < 0.05). Percentages of fat, protein, and total solids in milk were not affected by treatment. Trends ($P < 0.15$) for increased yields of 3.5% FCM, lactose, and total solids by cows fed +HMB were related to milk yield. Differences in plasma NEFA (495, 514, 446 μEq/L) and β-hydroxybutyrate (13.72, 12.04, 11.92 mg/dl) were not significant ($P > 0.25$). Liver triglyceride content was similar on d 1 postpartum (7.24, 6.61, 6.75%) and was increased for +HMB on d 21 postpartum [(8.87, 13.68, 11.07%) treatment x d; $P < 0.04$]. Differences in rates of [1-^{14}C]palmitate oxidation [27.9, 23.7, 25.4 nmoles/(hour x g wet weight)], and formation into stored esterified products [318, 316, 338 nmoles/(hour x g wet weight)] were not significant. The data suggest that adding HMB to low Met but adequate Lys diets at 0.1% prepartum and 0.15% postpartum is beneficial for increasing milk production. The underlying mechanism does not appear to be associated with hepatic lipid metabolism as measured in this experiment.

Key Words: HMB, Methionine, Liver

146 Use of milk protein concentrations to estimate the "methionine bioavailability" of two forms of 2-hydroxy-4-methylthio butanoic acid (HMB) for lactating cows. C. G. Schwab*[1], N. L. Whitehouse[1], A. M. McLaughlin[1], R. K. Kadariya[1], N. R. St-Pierre[2], B. K. Sloan[3], R. M. Gill[3], and J. C. Robert[4], [1]*University of New Hampshire, Durham,* [2]*The Ohio State University, Columbus,* [3]*Aventis Animal Nutrition, Alpharetta, GA,* [4]*Aventis Animal Nutrition, Antony, France*.

Forty multiparous Holstein cows (58 to 167 DIM) were assigned randomly to a balanced split-plot 5 x 5 Latin square design involving two replicates of four squares. Experimental periods were 14 d with the last 7 d for measurements. Each square consisted of five concentrations of a single methionine (Met) source in a Met-deficient diet. The four Met sources were: 1) Smartamine M-TM (SmM, Aventis Animal Nutrition), 2) HMB, 3) the isopropyl ester of HMB (HMBi), and 4) a combination of HMB and HMBi (HMB/HMBi). Treatment levels were (g Met equivalents/d per 25 kg of DMI): SmM (0, 10, 15, 20, and 25), HMB and HMBi (0, 15, 20, 25, and 30), and HMB/HMBi (0/0, 5/10, 8.3/16.7, 11.7/23.3, and 15/30). Because treatment levels within source were not spaced equally, appropriate orthogonal coefficients were generated using PROC IML of SAS. Corrected LSM for milk protein percentages were: SmM (2.99, 3.08, 3.15, 3.15, and 3.13; quadratic effect, $P < 0.01$), HMB (3.04, 3.02, 3.03, 3.06, and 3.03), HMBi (3.05, 3.11, 3.16, 3.17, and 3.19; linear effect, $P < 0.001$), and HMB/HMBi (3.07, 3.13, 3.12, 3.16, and 3.18; linear effect, $P < 0.001$). Inspection of the LSM indicated that a broken line model of response to Met sources was more adequate than smooth function models. Thus, PROC NLIN of SAS was used to identify the optimum level of supplementation (breakpoint) and the slope of the dose-response relationships prior to breakpoint for SmM, HMBi, and HMB/HMBi. Based on differences of slope, and the assumption that 80% of the Met in SmM is available, the bioavailability of Met from HMBi and HMB/HMBi was estimated to be 42% and 34%, respectively. Results indicate that HMB provided little or no Met for milk protein synthesis and that HMBi was 50% or more as effective as SmM.

Key Words: Methionine, HMB, Ruminant

147 Performance of high producing dairy cows fed methionine hydroxy analog or D, L-methionine in a total mixed ration during early lactation. K. Uchida[1], P. Mandebvu[2], C. J. Sniffen*[2], C. S. Ballard[2], and M. P. Carter[2], [1]*Zen-Noh National Federation of Agricultural Co-operative Associations, Tokyo, Japan,,* [2]*W. H. Miner Agricultural Research Institute, Chazy, NY.*

Effect of feeding two methionine supplements was compared using high producing Holstein cows during early lactation. Pregnant cows housed in a free-stall barn at the Miner Institute in northeastern New York, were blocked and at calving, were assigned randomly to one of two TMR containing a liquid form of methionine hydroxy analog (MHA; Novus Intl., Atlanta, GA), or D,L-methionine (D,L-Met), and group-fed for ad libitum intake. Cows spent 3315 days in the fresh group (FG), after which they were moved to the high producing group (HG) where they stayed up to 8 wk postpartum. The TMR had on DM basis a forage to concentrate ratio of 40 to 60% for FG cows with predicted DMI of 21.7 kg/d, and 42 to 58% for HG cows with predicted DMI of 26.5kg/d. All TMR contained on DM basis 33% NDF and 18% CP. The TMR were formulated using the CPM Dairy# model to meet the methionine and lysine factorial requirement, and the methionine and lysine ratios of 2.20 and 6.89% of metabolizable protein, respectively. The concentrations of methionine and lysine, respectively, in all TMR were 0.25, 0.75 (% of DM). In order to provide the same amount of methionine postruminally from the two methionine supplements, cows fed TMR containing MHA and D,L-Met, respectively, were fed approximately 14.2 g of MHA and 25.7 g of D,L-Met for FG and 17.1 g of MHA and 31.0 g of D,L-Met for HG, assuming a rumen escape value of 40% for MHA and 22% for D,L-Met. The average daily DMI by the group-fed cows across treatments was 222.5 kg/cow for the FG, and 272.0 kg/cow for HG. Cows that were fed TMR containing MHA and D,L-Met, respectively, had similar ($P>0.18$) milk yield (49.0, 49.8 kg/d; SE=0.32), milk fat (4.03, 4.21%; SE=0.051), milk CP (3.13, 3.21%; SE=0.019), linear SCC (4.08, 4.55; SE=0.072), body condition score (3.17, 3.17; SE=0.019), days to first service (68.7, 65.1; SE=6.61), and first service conception rate (36.9, 35.0%) during wk 1 to 8 postpartum, and different milk fat (3.64, 3.93%; SE=0.07, P=0.07) during wk 5 to 8 postpartum. Average milk yield was 45.5 kg during wk 1 to 4 postpartum and 53.3 kg during wk 5 to 8 postpartum. In conclusion, D,L-Met performed as well as MHA in promoting milk yield and contents of milk fat and CP when fed at levels aimed at achieving similar amounts of methionine postruminally as supplied by MHA.

Key Words: dairy cow, methionine hydroxy analog, milk yield and reproductive performance

148 Effect of two levels of crude protein and supplementation of methionine on performance of dairy cows. C. Leonardi*[1], L.E. Armentano[1], and M. Stevenson[2], [1]*University of Wisconsin-Madison,* [2]*Degussa Canada Ltd., Ontario, Canada.*

Sixteen lactating Holstein cows (4 primiparous and 12 multiparous) were used in a 4 x 4 Latin Square, with periods of 35 days. At the beginning of the study animals averaged 95 DIM and produced 44.5 kg/d of milk. The effect of supplemental methionine at two levels of CP (16.1 vs. 18.8%) was tested. The two levels of protein and methionine supplementation were such that the lower level of CP supplemented methionine was sufficient to cover the requirements of amino acids, according to the Mepron Dairy Ration Evaluator ver. 2.1 (Degussa Huls Corp., Allendale NJ). The high level of protein (HP) was selected to meet the amino acids requirement without the supplementation of methionine. A low protein (LP) diet without methionine was added as negative control and the high protein diet plus methionine (HPM) as a positive control. All four diets contained 16.2% alfalfa silage, 38.6% corn silage, 16.2 % of corn grain, 8.1% soybean roasted, 5.7 % cottonseed, 2% soyplus, 1% blood meal, 0.8% animal fat, and 1.8 % minerals and vitamins mix (DM basis). The remaining 9.6% of the diet was either corn grain plus urea in the low protein diet, or soybean meal in the high protein diet. We anticipated an interaction where low protein responded to methionine more than the high protein diet, but no interactions were significant. The only methionine main effect was an increase in milk protein %, while increased dietary protein decreased milk protein %. Milk fat was depressed across diets. Low dietary NDF (27.2 % of DM), and high levels of vegetable oil (4.5 % fatty acids) may have been the cause of the low milk fat test. These dietary conditions may also have caused microbial protein synthesis to differ from the values predicted by the model used. A main effect of dietary protein was to elevate milk fat concentration and yield.

-4 Treatments		-7 Effect (P-value)				
	LP	LPM	HP	HPM	SEM	CP	Met
DMI, kg/d	22.3	21.8	22.1	23.3	.8	.21	.49
Milk yield, kg/d	42.7	41.0	42.2	42.8	1.6	.51	.63
Protein, %	3.21	3.28	3.13	3.23	.08	.01	<.01
Protein, g/d	1361	1330	1311	1373	45	.93	.71
Fat, %	2.27	2.39	2.71	2.65	.12	<.01	.74
Fat, g/d	949	958	1099	1116	59	<.01	.69
MUN[1], mg/dl	10.44	10.60	14.53	14.17	.94	<.01	.88
MUN, g/d	4.44	4.29	5.96	6.10	.5	<.01	.98
Casein N/total N	.666	.641	.694	.691	.026	.13	.57

[1]MUN=milk urea nitrogen

Key Words: Crude protein, Methionine

149 Effects of rumen undegradable protein digestibility and supplemental methionine on production parameters and nitrogen efficiency of Holstein cows in early lactation. S. Noftsger* and N. St-Pierre, *The Ohio State University.*

A production trial followed by a collection trial was conducted to assess the effects of post-ruminal undegradable protein (RUP) digestibility, metabolizable protein supply and Met supplementation on production efficiency and N utilization of lactating dairy cows in early lactation. Treatments were: 1)18.5 % crude protein (CP) with low estimated intestinal digestibility RUP (Control); 2) 18.5 % CP with high digestibility RUP (HiCP); 3) 17 % CP with high digestibility RUP (LowCP); and 4) 17 % CP with high digestibility RUP and supplemental Met (LowCP + Met). Thirty-six multiparous and 24 primiparous cows were assigned at random to one of the four dietary treatments 3 weeks post-freshening. At week 13, six cows from each treatment were placed in metabolic stalls for a digestibility study, each cow remaining on the same treatment as during the production trial. Treatments had a significant effect on dry matter intake (21.7, 23.3, 23.2, 23.6; SE = .05 kg/d), milk production (40.8, 46.2, 42.9, 46.6; SE = 0.7 kg/d), protein production (1.20, 1.38, 1.28, 1.44; SE =0.02 kg/d) and milk protein content (2.95, 2.99, 3.00, 3.09; SE = 0.03 %) for Control, HiCP, LowCP, and LowCP + Met respectively in the production trial. Nitrogen balance results are reported in the following table. Lowering the CP in diets in early lactation in conjunction with selection of RUP sources selected for higher estimated post-ruminal RUP digestibility can improve the efficiency of N utilization. The supplementation of Met did not improve the efficiency of N utilization during the digestibility study, in contrast to what was estimated during the production trial.

	Control	HiCP	LowCP	LowCP + Met	SEM	p-value
N intake (g/d)	759	734	681	679	27.9	.031
N feces (g/d)	279	271	257	263	10.9	.292
N urine (g/d)	268	259	215	224	19.3	.067
N milk (g/d)	223	217	223	216	9.4	.909
N retained (g/d)	-0.6	-13.2	-15.6	-23.3	18.3	.838
App. N digestibility (%)	63.7	62.9	61.9	60.8	1.8	.320
N excreted/N milk	2.44^{a}	2.44^{a}	2.09^{b}	2.24^{ab}	.092	.039

Key Words: RUP digestibility, methionine, N efficiency

150 Ruminal escape and response of serum methionine to 25 and 50 grams of methionine hydroxy analog in dairy cows. K. M. Koenig*[1], M. Vazquez-Anon[2], C. D. Knight[2], and L. M. Rode[1], [1]*Agriculture and Agri-Food Canada, Research Centre, Lethbridge, AB, Canada,* [2]*Novus International, Inc., St. Louis, MO, US.*

Ruminal escape of various amounts of methionine hydroxy analog [D,L-2-hydroxy-4-(methylthio)-butanoic acid (HMB), Novus International Inc., St. Louis, MO] was measured in an experiment designed as a 4 × 4 Latin square using four lactating dairy cows with cannula in the rumen. The cows were fed a diet composed of corn silage, alfalfa haylage, rolled barley grain, canola meal, and blood meal, three times per day. The cows were fed the liquid analog each day for one week prior to the initiation of the experiment. On the day of the experiment, each

cow received an oral bolus dose of 0, 25, or 50 g of the liquid analog (Alimet® feed supplement, 88% HMB) or 51.2 g of a dry calcium salt of the analog (86% HMB; MHA®) mixed with 0.5 kg of ground barley grain. After a period of 30 min, any remains of the carrier grain and treatment were placed in the rumen. A liquid phase marker (Co-EDTA) was administered as a bolus dose into the rumen at the time of oral administration of the methionine analogs. Rumen contents and blood serum were collected at 0, 1, 3, 6, 9, 12 and 24 h relative to the time of dosing. Rumen samples were analyzed for Co and HMB, and serum was analyzed for free methionine. Fractional rate constants for the passage of the liquid marker (k_p) and the decline of HMB concentration in the rumen (k_{rHMB}) were determined by non-linear regression. Liquid passage from the rumen was similar among the four analog treatments (0.136 ± 0.022 /h; mean ± SEM). Ruminal escape of HMB as a percentage of the dose (100% × k_p/k_{rHMB}) did not differ between cows receiving 25, 50, and 51.2 g of the methionine analogs (42.5, 41.0, and 34.9 ± 9.0%, respectively) and averaged 39.5%. Serum methionine concentration peaked at 3 and 6 h after dosing and increased in proportion to the amount of the analog administered. It was concluded that 39% of the methionine hydroxy analog escaped rumen degradation, the percentage of the dose that escaped the rumen was not affected by the amount or form of the methionine analog fed, and the analog that escaped ruminal degradation was absorbed and metabolized to methionine.

Key Words: HMB, Methionine, Ruminal escape

151 Carbohydrate fermentation and nitrogen metabolism of a finishing diet by ruminal microbes in continuous cultures as affected by ethoxyquin and(or) supplementation of monensin and tylosin. H. Han*[1], H. S. Hussein[1], H. A. Glimp[1], D. H. Saylor[2], and L. W. Greene[3], [1]*University of Nevada - Reno,* [2]*Solutia Inc.,* [3]*Texas A&M University.*

Long-term feedlot studies have shown positive effects (i.e., improved ADG and gain/feed; reduced morbidity and mortality) of dietary supplementation with ethoxyquin (AGRADO®). This may be due to improving the antioxidant capacity at the ruminal, post-ruminal, or post-absorption levels. This study was designed to investigate the potential antioxidant role of ethoxyquin at the rumen level. A finishing diet (12.5% CP; DM basis) was formulated to contain (on a DM basis) 77.5% flaked corn, 10% corn cobs, 10% protein/vitamin/mineral supplement, and 2.5% animal fat. In a randomized complete block design experiment, the treatments were arranged as a 2 × 2 factorial. The main factors were 2 ethoxyquin treatments (without or with 150 ppm) and 2 monensin/tylosin treatments (without or with monensin and tylosin at .0028 and .0014% of dietary DM, respectively). Eight dual-flow continuous culture fermenters were used in two experimental periods (blocks; 8 d each with 5 d for adjustment and 3 d for sample collection) to allow for 4 replications for each treatment. No interactions (P > .05) were detected for any of the measurements evaluated. Therefore, results of the main factors were summarized. Ethoxyquin supplementation improved (P < .05) true digestibility of DM (from 40.7 to 47.1%) and OM (from 38.8 to 45.0%) but it did not alter (P > .05) concentrations of total VFA (averaging 131.4 mM) or acetate (averaging 58.8 mM). Ethoxyquin decreased (P < .05) propionate concentration from 51.1 to 42.4 mM and increased (P < .05) butyrate concentration from 18.4 to 22.9 mM. Digestion of total nonstructural carbohydrates was not altered (P > .05) by the treatments and averaged 86%. With the exception of increased (P < .05) concentration of propionate (from 42.0 to 51.5 mM) and decreased (P < .05) concentration of butyrate (from 25.9 to 16.3 mM), no effects were detected for monensin/tylosin. Ruminal N metabolism including efficiency of bacterial protein synthesis (averaging 21.2 g N/kg OM truly digested) was not affected (P > .05) by the treatments. Results suggest positive effects of ethoxyquin on ruminal digestion of DM and OM and unique changes in VFA production.

Key Words: Continuous culture, Ethoxyquin, Beef cattle

152 Comparison of different methods of administration on the effect of fibrolytic enzymes on digestive processes in lactating cows. J.D. Sutton*[1], R.H. Phipps[1], D.E. Beever[1], D.J. Humphries[1], G.F. Hartnell[2], and J.L. Vicini[2], [1]*University of Reading, UK,* [2]*Monsanto Co, St Louis, MO.*

To clarify the site of action of fibrolytic enzymes extracted from *Trichoderma longibrachiatum*, four multiparous Holstein x Friesian cows with cannulas in the rumen and proximal duodenum were used in a 4 x 4 Latin square experiment with 5-wk periods. From lactation wk 5, the cows were given ad libitum a TMR composed of (DM) 57% forage (3:1 maize silage:grass silage) and 43% concentrates. The TMR contained (g/kg DM): 274 NDF, 295 starch, 180 CP. Treatments were TMR alone (T1) or TMR with the enzymes added (2 g/kg TMR DM) either sprayed on the TMR 1 h before the morning feed (T2), sprayed only on the concentrate the day before feeding (T3), or infused into the rumen for 14 h/d (T4). Xylanase and endoglucanase activities of the enzymes were 26,483 and 2,645 mol.min^{-1}.g^{-1}. There was no significant effect on feed intake (T1-T4: 20.7, 21.1, 20.9, 20.4 kg DM/d, s.e.m. 0.43) or milk yield (34.0, 35.5, 34.5, 35.0 kg/d, s.e.m. 0.52) but both were highest on T2. Rumen digestibility of DM (0.321) and starch (0.843) was unaffected. Digestibility of NDF was lowest on T2 in the rumen (0.449, 0.335, 0.402, 0.492, P<0.05) but highest on T2 post-ruminally (0.090, 0.247, 0.161, 0.006, P<0.05). Total tract digestibility was highest on T2 for DM (0.714, 0.724, 0.707, 0.714, P<0.05) and starch (0.970, 0.977, 0.970, 0.972, P<0.05) but treatment differences were non-significant for NDF (0.503). Maize silage stover passage rate out of the rumen was increased by all enzyme treatments (0.025, 0.030, 0.033, 0.032 h^{-1}, P<0.05) but gut transit time was also increased (12.5, 16.7, 16.1, 15.9 h, P<0.05) so the decline in total tract retention time (61.2, 58.3, 54.5, 55.7 h, s.e.m. 2.57) was not significant. Although inclusion of these enzymes by all three methods altered digesta kinetics, it was only when they were sprayed on the TMR that diet utilisation was improved with higher DM digestibility and a numerical increase in milk yield.

Key Words: Enzymes, Dairy Cows, Forage

153 Effects of liquid feed supplementation and (or) celluloytic enzymes on dry matter disappearance of either legume or grass hay. G. V. Pollard*[1], W. T. Wright[1], T. C. Bramble [1], C. R. Richardson[1], and C. W. Cobb[2], [1]*Texas Tech University, Lubbock,* [2]*Loveland Ind., Inc., Greeley, CO.*

The objective of this research was to determine if pH of a suspension liquid feed product and (or) cellulolytic enzyme supplementation increases the digestibility of forage as indicated by in vitro dry matter disappearance (IVDMD). Liquid feed suspensions (32% CP, as fed) and cellulolytic enzymes (CE) were added as aqueous solutions and sprayed directly onto ground (1mm) .5 g samples of alfalfa, wheat, or grass hay. Treatments were negative control (CON); enzyme only (ENZ); liquid supplement, pH 4 (LS); liquid supplement plus enzyme (LE); liquid supplement plus enzyme buffered to pH 5 (LE5). Levels of CE were equivalent to 185 g per 907 kg of forage DM and liquid feed supplement levels were equivalent to intakes of 1 kg as fed/hd/d. The IVDMD of samples was determined following a 24 h incubation in 100-mL in vitro tubes containing 50-mL of a buffer and rumen fluid mixture. Rumen fluid was obtained from a cannulated steer fed a roughage diet. Alfalfa hay samples had greater (P < .05) overall IVDMD than either wheat or grass hay samples. Alfalfa hay IVDMD was improved (P < .05) by ENZ, LS, and LE, however, alfalfa hay was unaffected (P > .05) by LE5. Wheat hay samples were unaffected (P > .05) by dietary treatments, however LE5 tended (P = .09) to improve IVDMD over CON. Also, ENZ tended (P = .08) to have lower IVDMD when compared to LS in wheat hay, and reduced (P < .05) IVDMD in alfalfa hay. In contrast, grass hay samples were unaffected (P > .05) by either LS or ENZ, while both LE and LE5 improved (P < .05) IVDMD. Treatments containing liquid feed supplements and (or) CE resulted in greater overall IVDMD in both alfalfa and grass hay samples. These results indicate that the addition of CE and liquid feed supplements either separately or in combination, improves digestibility of alfalfa and grass forage. However, buffering liquid feed supplements containing CE from pH 4 to pH 5 appears to be unwarranted.

Key Words: Enzymes, Liquid feed, Dry matter disappearance

154 Effects of ruminant feed enzyme additives on digestibility evaluated in vitro. G. R. Bowman*[1], K. A. Beauchemin[2], and J. A. Shelford[1], [1]*University of British Columbia, Vancouver, Canada,* [2]*Agriculture and Agri-Food Canada, Lethbridge, Canada.*

A study was conducted to determine if fibrolytic enzymes increase feed digestion by ruminants. A revolving digestion incubator and mesh bags (9.5 X 19.5 cm) were used to evaluate the dry matter digestibility (DMD,%) and neutral detergent fiber digestibility (NDFD,%) of a TMR for dairy cows consisting of barley silage and barley grain or the same

TMR treated with a fibrolytic enzyme product (Promote, Agribrands Intern., St. Louis, MO). The objectives were: 1) determine whether enzymes improve DMD or NDFD, and 2) establish whether increased digestibility was due to feed-treatment effect or an enhancement of the hydrolytic capacity of the ruminal fluid. Two cannulated lactating Holstein cows were fed a sequence of four TMR: no enzymes (-E), enzymes added to the concentrate portion of the TMR (C+E), enzymes added to the supplement portion of the TMR (S+E), or enzymes added to the premix portion of the TMR (P+E). The same cows served as rumen fluid donors, which was composited by diet. Fresh, unground TMR was incubated in vitro for 12 or 48 h. At 12 h, when rumen fluid from the respective diets was used, -E showed the lowest DMD (41.2 ^{a}), followed by S+E (43.2 ^{ab}), C+E (44.7 ^{b}), and P+E (44.8 ^{b}). However, there were no differences ($P > 0.05$) among treatments for NDFD at 12 h (20.08±12.0). There was no effect of treatment ($P > 0.05$) at 48 h for DMD (59.83±3.8) or NDFD (40.88±9.3). When the -E TMR was incubated with the various inoculates, there was no improvement in its DMD or NDFD at 12 or 48 h. We did not test the effects of incubating the enzyme-treated TMR in ruminal fluid from cows fed -E. This study indicates that fibrolytic enzymes increase DMD during the early stages of digestion. However, enzyme supplementation has no effect on feed digestibility at longer incubation times, which may indicate that enzymes improve rate, rather than extent, of digestion. Enzymes must be in physical contact with the feed to improve digestibility.

Key Words: Enzyme, Digestibility, in vitro

155 The effect of different levels of yeast culture inclusion in the concentrate diet on calf performance. R.J. Fallon*[1] and B. Earley[1], [1] *Teagasc.*

The recent banning of a number of in-feed antibiotic digestive enchancers within the European Union has generated the need to find suitable non-antibiotic alternatives. Yeast culture (YeaSacc 1026) as a supplement has been evaluated in several growth studies in calves, steers and bulls. There is however, little information available on the optimum level of YeaSacc 1026 for inclusion in the calf concentrate ration. The aim of this study was to determine the optimum level of YeaSacc 1026 inclusion. The following experiment was undertaken using 80 Friesian male calves (average initial weight of 54 kg) to determine the optimum inclusion rate of YeaSacc 1026 in a barley soyabean meal ration. Calves were allocated at random to 1 of 4 treatments: 1) 0, 2) 0.625, 3) 1.25 and 4) 2.5 kg YeaSacc 1026per tonne of ration. The concentrate ration was available ad libitum throughout the 84 day experimental period and the calves were offered 25 kg of calf milk replacer by bucket over the initial 42 day period. Calf liveweight gain in the period 1-42 days was 0.58, 0.65, 0.65 and 0.68 (s.e.d 0.029) kg/d for treatments 1 through 4, respectively. The corresponding liveweight gain for the period 1 to 84 days were 0.84, 0.92, 0.92 and 0.89 (s.e.d 0.026) kg/day, respectively. Concentrate intake (1 to 42 day) were 25, 29, 30 and 31 (s.e.d 2.1) kg, respectively the corrosponding values for the period 43 to 84 days was 129, 138, 138 and 140 kg. It was concluded that the inclusion of 0.625 or 1.25 kg of YeaSacc 1026 per tonne of ration increased calf liveweight gain by 5 kg in the period 1 to 84 days.

Key Words: Yeast Culture, YeaSacc, Calves

156 The effect of different levels of YeaSacc 1026 inclusion on the lifetime performance of cattle offered an ad libitum concentrate ration. R.J. Fallon*[1] and B. Earley[1], [1] *Teagasc.*

The recent banning of a number of in feed antibiotic digestive enchancers within the European Union has generated the need to find suitable non-antibiotic alternatives. Yeast culture (YeaSacc 1026) as a supplement has been evaluated in several growth studies in calves, steers and bulls. There is however, little information available on the optimum level of YeaSacc 1026 for inclusion in the diet on the lifetime performance of bulls offered an ad libitum concentrate diet. The aim of this study was to determine the optimum level of YeaSacc 1026 inclusion. The following experiment was undertaken using 80 Friesian male calves (average initial weight of 54 kg) to determine the optimum inclusion rate of YeaSacc 1026. Calves were allocated at random to 1 of 4 treatments: 1) 0, 2) 0.625, 3) 1.25 and 4) 2.5 kg YeaSacc per tonne of ration using individual bulls as the experimental unit. The concentrate ration consisted of rolled barley, soya bean meal plus minerals and vitamins was available throughout the 329 day experimental period. The crude protein content was 168 g/kg to 12 weeks and 151 g/kg there after. In addition the animals were offered a fixed daily allowance of straw (80g/kg of concentrates). Bull liveweight gain in the period 1 to 168 days was 1.11, 1.14, 1.20 and 1.15 (s.e.d 0.022) kg/day from treatments 1 through 4, respectively. The corresponding liveweight gains for the period 169 to 329 days were 1.32, 1.36, 1.34 and 1.36 (s.e.d 0.043) respectively. The corresponding concentrate dry matter intakes were 548, 562, 583 and 565 for the period 1 to 168 days and 1122, 1161, 1208, 1143 for the period 169 to 329 days. The corresponding carcass weights were 233, 239, 242 and 239 kg. It was concluded that the 1.25 kg/tonne inclusion rate for YeaSacc 1026 resulted in a response in the 1 to168 day period,but was not significant

Key Words: Yeast Culture, Cattle, Dose Response

157 The effects of feeding a fungal extract (Amaferm) to ewes 60 days prepartum through weaning on milk protein, fat, lactose and yield. S. L. Campbell*, S. P. Jackson, A. D. Herring, M. L. Galyean, and D. R. Niemann, *Texas Tech University Lubbock,TX/US.*

The objectives of this study were to determine differences in milk protein, fat, lactose and milk yield in ewes either fed whole corn and CSM (control) or the same diet plus Amaferm (AMF), which contains the fungal extract *Aspergillus orzaye*. Sixty-two Rambouillet ewes from ages 3 to 7 yr. were randomly assigned to four pens, and treatments were randomly assigned to each pen. Milk protein, fat, and lactose were measured in milk samples taken at parturition (time 0), 6 h and every 12 h thereafter for the first 36 h and additionally at 3 wk and 6 wk. Milk yield was measured at 3 and 6wk postpartum. Milk yield was not affected by treatment at the 3 or 6 wk milking time. Milk protein decreased linearly through lactation. Percent milk protein was 11.63, 10.99, 9.19, 7.06, 6.38, 5.12, and 5.12 at time 0, 6 h, 12 h, 24 h, 36 h, 3 wk, and 6 wk, respectively. Percent milk fat increased from 8.50 at time 0 to 9.69 at 6 h, then decreased to 8.75 by 36 h., and 5.06 at 3 wk., but increased to 5.34 by 6 wk postpartum. Percent milk lactose increased linearly through lactation (3.28, 3.54, 3.90, 4.33, 4.46, 5.37, and 5.31 at time 0, 6h, 12 h, 24 h, 36 h, 3 wk, and 6 wk, respectively). There were no differences in milk protein between ewes fed AMF or the control diet over the entire lactation, but there was a trend at 6 h, suggesting the control group had a slightly higher milk protein content (P = .06) than the AMF group. Ewes on AMF had higher milk fat than ewes on the control diet at 0 h. (P = .004) and 3 wk (P = .02) and tended to have higher milk fat at 6 h (P = .10). Milk lactose was lower in ewes consuming AMF at 6 h. (P = .02) and tended to be lower at 12 h (P = .08) than for ewes in the control group. In this study, Amaferm addition to the diet did not affect milk yield, but it increased milk fat and decreased milk lactose at certain stages of lactation.

Key Words: Amaferm, Sheep , Lactation

PSA Environment and Management: Broilers

158 Impact of Aflatoxin in the feed on Coccidial infection in broiler chicks. V.G. Stanley[1], D. Spiller*[1], W. Kruger[2], and A. Sefton[3], [1] *Prairie view A&M University,* [2] *Texas A&M University,* [3] *Alltech, Guelph Canada.*

An experiment was conducted to examine the effects of aflatoxin on male broiler chicks infected with Eimeria acervulina. The experimental design was a 2 x 2 factorial, consisting of two levels of aflatoxin (0; 3 mg/kg) and two levels of coccidia (0; 500,000 oocyts/chick). Chicks were fed an aflatoxin-treated or non-aflatoxin-treated diet (control) from 1 d of age through 28 d of age. At 14 d of age, the chicks were infected with Eimeria acervulina. At 14 d post-infection the effects of treatments were assessed on weight gains, relative organs (liver, pancreas, gizzard, proventriculus, heart and cecal) weights, and gross lesion scores. Blood was collected for serum chemistry values, and enzyme activities. Weight gain in chicks fed the aflatoxin-treated diet infected with E. acervulina was significantly lower (P < .05) than the control by 26.8 percent. Chicks fed an aflatoxin-treated diet had a 20.46 percent