The soybean meal group (SBMG; n = 17) was supplemented daily with 362 g of soybean meal (SBM)/animal. The low cottonseed meal group (CSML; n = 17) was supplemented with a mixture including 181 g of SBM and 227 g cottonseed meal (CSM; 0.09 % FG). The high cottonseed meal group (CSMH; n = 16) was supplemented with 454 g CSM/animal. The daily intake of FG/animal for SBMG, CSML, and CSMH was 0.0, 0.20, and 0.41 g, respectively. Beginning 8/14/03, BW, BCS, and serum blood samples were collected weekly, until 11/20/03. Ultrasonography, for pregnancy detection, was performed for all does on 11/20/03 and 12/15/03. The SBMG (-49.83±4.08 g/animal/d) lost less (P<.01) BW than CSML (-73.84±4.08 g/animal/d) or CSMH (-77.01±4.08 g/animal/d). Average daily BW loss did not differ (P>.1) between CSML and CSMH. Body condition score (5.36±0.09), pregnancy rates (100%), and time between weaning and conception did not differ (P>.1) among treatments. Doe serum progesterone concentrations were reduced (P<.05) in CSMH relative to SBMG and CSML. Among lactating does, BW and BCS at weaning was correlated (P<.01, R= -0.50 and P<.05, R= -0.41, respectively) with time between weaning and conception. Despite reductions in BW gains and serum progesterone concentrations, consumption of CSM (8.1 mg FG/kg BW; 0.41 g FG/animal/d) did not affect reproductive performance of fallow deer.

336 Soybean hulls for finishing meat goats. M. H. Poore*, J. A. Moore, A. T. Maye, and J.-M. Luginbuhl, *North Carolina State University*.

Soyhulls (SH) have been used at high levels in goat diets, but feeding method has not been studied. Twenty-four wethers, at least 75% Boer (21 kg), were allotted to 4 treatments (trt). All trt used orchardgrass hay (10.5 % CP and 65.9% NDF). Treatment 1 (hay) was 100% hay, trt 2 (HFcorn) was 33% hay and 67% hand-fed corn/SBM mix (12.5% CP and 11.0% NDF), trt 3 (HFSH) was 33% hay and 67% hand-fed SH (12.3% CP and 64.2% NDF), and trt 4 (FCSH) was hay at 0.75% of live wt with free choice SH. One goat on HFcorn was removed due to low intake. All goats were started slowly on concentrate (conc) with ad lib hay. Initially, loose SH were fed, but after the first wk most goats were refusing their SH, so they were changed on d 12 to a pelleted source which was acceptable to all goats. Once all goats began consuming their conc at 33% of diet DM, they were fed 50% conc for 3 d and then 67% conc for 3 d. Following that, trt 4 received SH free choice and hay intake was restricted. All goats were on final diets by d 28. Ruminal samples were taken by rumenocentesis 4 h after feeding on d 84, and goats were harvested on d 85. Contrasts determined were hay vs. the other 3 trt (C1), HFcorn vs. HFSH (C2), and HFSH vs. FCSH (C3). Due to differences in dressing %, final live wt was adjusted using carcass wt and the average dressing % for all trt to calculate ADG. The study showed that all conc trt outperformed hay, that HFcorn was similar to HFSH, and that there was little difference between HFSH and FCSH. Therefore, SH are a viable feed for finishing goats, and can be fed free-choice.

Item	Hay	HFcorn	HFSH	FCSH	SEM	C1*	C2*	C3*
DMI, g/d	557	685	764	709	25.5	.01	.05	.13
Carcass wt, kg	9.3	14.6	14.5	14.3	.44	.01	.88	.76
Loineye, sq cm	11.0	14.6	15.1	17.1	.81	.01	.28	.24
KPH, %	1.55	3.60	2.66	2.72	.34	.01	.08	.90
Dressing %	40.5	51.0	50.4	53.2	.85	.01	.65	.03
Adj ADG, g/d	-19.6	100.7	103.7	92.8	9.17	.01	.82	.40
Gain:feed	-.036	.145	.135	.130	.011	.01	.57	.77
Ruminal pH	6.37	5.94	6.08	6.23	.090	.01	.28	.24
Ruminal A:P	3.52	1.85	3.83	4.49	.210	.99	.01	.01

*P value for contrast

Key Words: Meat Goats, Soybean Hulls, Carcass

Ruminant Nutrition: Dairy - Protein & Amino Acids

337 Effects of parity and levels of protein on production response and n-balance in holsteins. S. A. Flis* and M. A. Wattiaux, *University of Wisconsin-Madison*.

	Treatment					P-Value		
	C	U	D	UD	SEM	D*	U**	UxD
DMI kg/d	22.7	24.4	21.1	23.6	0.9	0.07	<.01	0.57
NI g/d	618	707	596	712	24	0.62	<.01	0.46
Milk kg/d	38.8	40.6	37.4	38.6	1.2	0.04	0.06	0.71
Fat kg/d	1.28	1.42	1.35	1.22	0.08	0.25	0.89	0.02
%Fat	3.32	3.52	3.65	3.24	0.17	0.80	0.37	0.02
Prot kg/d	1.00	1.07	1.00	0.99	0.05	0.18	0.30	0.20
%Prot	2.59	2.66	2.68	2.61	0.11	0.76	0.97	0.14
TMN g/d	175	189	169	181	8.31	0.21	0.06	0.92
Fecal N g/d	209	232	190	208	10	<.01	<.01	0.68
UN g/d	194	213	202	223	7.35	0.26	0.02	0.88

*P-value for D and UD vs. U and C
**P-value for U and UD vs. D and C

Eight Holstein cows (4 primiparous and 4 multiparous) were used in a replicated 4x4 Latin Square to determine milk production response and N balance when diets had no excess of RUP or RDP (C), 10% RUP excess (U), 10% excess RDP (D), or 10% excess of both RUP and RDP (UD) according to NRC 2001. Solvent soybean meal and soyPLUSTM made up 6.6 and 5.1, 6.1 and 7.5, 11.8 and 1.2, or 11.3 and 4.1% for the C, U, D, and UD diets, respectively. Diets were fed as a TMR with 25% alfalfa silage, 25% corn silage (DM basis) and corn grain as the primary source of dietary starch. During each 21day period, milk yield and DMI were recorded daily, and in the last 3 days of each period fecal and urinary N (UN) excretion were determined from total collection. Dietary CP averaged 17, 18, 17.6, and 18.7% for the C, U, D, and UD diet, respectively. DMI, milk and protein yield were lower in primiparous than in multiparous cows ($P < 0.05$). N intake (NI) was 535 and 782 g/d for primiparous and multiparous cows, respectively ($P < 0.01$). Total milk N (TMN g/d), fecal N and UN were lower for primiparous than for multiparous cows ($P < 0.01$), but N balance did not differ with parity (P = 0.2). Highest DMI and NI were in diets with excess RUP (see Table). Milk yield was lower on diets with excess RDP (D and UD) ($P < 0.05$). Fecal N, UN, and N balance (data not shown) were higher in diets with excess RUP. N balance was positive for all treatments. Through the trial little change in BW was observed. Results indicated that greater efficiency of N utilization on farm could be obtained by balancing rations for first and later lactation cows separately.

Key Words: Nitrogen, N-Balance

338 Site of digestion in dairy cows fed different sources and amounts of crude protein. I. R. Ipharraguerre*[1], J. H. Clark[1], and D. E. Freeman[2], [1]*Department of Animal Sciences, University of Illinois, Urbana*, [2]*Department of Veterinary Clinical Medicine, University of Illinois, Urbana*.

Six multiparous Holstein cows cannulated in the rumen and duodenum that averaged 70 DIM were used in a 6x6 Latin square design with a 2x3 factorial arrangement of treatments. Two sources of CP (soybean meal (SBM) and a mixture of SBM and a commercial blend of high rumen-undergradable (RUP) CP sources) and three contents of dietary CP (about 14, 16, and 18%) were combined into six dietary treatments. Each source of CP supplied about 50% of the CP mixture used to formulate the high RUP diets. On DM basis, diets contained 25% corn silage, 20% alfalfa silage, 10% cottonseed, 26.7 to 37% corn grain, and 4 to 13.5% CP supplement. Diets were fed twice daily for ad libitum intake. Intakes of DM, OM, and NDF, and OM truly digested in the rumen were unaffected by treatments (P >.05; mean = 23.2, 21.7, 6.8, and 8.9 kg/d, respectively). As dietary CP increased from 14 to 18%, starch intake (8.0, 7.1, and 6.7 kg/d) and apparent ruminal (3.4, 2.8, and 1.6 kg/d) and total tract digestion (7.6, 6.7, and 6.4 kg/d) decreased (P <.01). At 14% CP, starch intake and total tract digestion (+.7 and +.6 kg/d) were higher for the RUP diet than the SBM diet, but the opposite occurred at 16% CP (.8 and .8 kg/d, P <.01). Across CP sources, increasing CP in the diet from 14 to 18%, increased N intake (510, 612, and 668 g/d, P <.01), ruminal outflow of nonamonnia N (521, 536, and 617g/d, P <.03), nonamonnia nonmicrobial N (224, 264, and 365 g/d, P <.01), Lys (157, 171, and 220 g/d, P <.02), total essential AA (1177, 1253, and 1531 g/d, P <.02), and total AA (2525, 2688, and 3265 g/d, P <.02). Across CP percentages, replacing SBM with RUP in the diet increased Met intake (57 vs. 63 g/d, P <.01) but did not affect Met flow to duodenum (61 vs. 64 g/d, P >.05). Ruminal outflow of microbial N (mean = 270 g/d) and milk yield (mean = 30.1 kg/d) were unaffected by treatments (P >.05). Data suggest that SBM-corn-based diets containing about 16% CP can quantitatively optimize the supply of N to duodenum of dairy cows consuming large amounts of feed.

Key Words: Nutrient flows, Protein, Dairy cows

339 Performance of lactating dairy cows fed different sources and amounts of crude protein. I. R. Ipharraguerre* and J. H. Clark, *Department of Animal Sciences, University of Illinois, Urbana.*

Sixty multiparous Holstein cows were involved in a 210-d lactation trial using a completely randomized design with a 2x3 factorial arrangement of treatments. Two sources of CP (soybean meal (SBM) and a mixture of SBM and a commercial blend of high rumen-undergradable (RUP) CP sources) and three concentrations of dietary CP (about 15, 17, and 19%) were combined into six dietary treatments. Each source of CP supplied about 50% of the CP mixture used to formulate the high RUP diet. On DM basis, diets contained 25% corn silage, 20% alfalfa silage, 10% cottonseed, 26.7 to 37% corn grain, and 4 to 13.5% CP supplement. Diets were fed twice daily for ad libitum intake. Across the 210 d and treatments, DM intake (DMI); yields of milk, 3.5% FCM, fat, and true protein (TP); FCM/DMI; and body condition score (BCS) averaged 24.6, 37.8, 36.8, 1.26, and 1.10 kg/d; 1.51; and 2.82, respectively. Most of these variables, however, showed a significant (P< .05) interaction between concentration and source of CP. At 19% CP, cows fed SBM had greater DMI, yields of FCM, fat, and TP (+2.5, +2.2, +.11, and +.05 kg/d), but lower FCM/DMI (.06) and BCS (.27) than cows fed RUP. At 17% CP, cows fed RUP had higher yields of FCM, fat, and TP (+2.9, +.13, +.05 kg/d) and FCM/DMI (+.06) than cows fed SBM. At 15% CP, cows fed SBM had higher DMI (+.6 kg/d) but lower yield of TP (.07 kg/d) and FCM/DMI (.06) than cows fed RUP. Across CP sources, cows fed 15% CP compared with cows fed 17 and 19% CP showed lower (P< .05) yields of FCM and TP (mean = 35.8 and 1.06, 36.7 and 1.13, 37.9 and 1.13 kg/d, respectively) and tended (P< .07) to have lower FCM/DMI (mean = 1.49, 1.49, and 1.55, respectively). Across CP percentage, cows fed RUP showed higher FCM/DMI than cows fed SBM (mean = 1.54 vs. 1.48). As dietary CP increased from 15 to 19%, milk urea nitrogen increased on average from 8.89 to 14.48 mg/dl. Data indicate that CP content of the diet of lactating dairy cows can be decreased without decreasing milk and milk component yields if the source and amount of dietary CP and carbohydrate are properly matched.

Key Words: Performance, Protein, Dairy cows

340 Effects of different protein supplements on nitrogen utilization in dairy cows. I. Lactation performance and ruminal metabolism. A. F. Brito*[1] and G. A. Broderick[2], [1]*University of Wisconsin, Madison,* [2]*US Dairy Forage Research Center, Madison, WI.*

Sixteen multiparous and 8 primiparous Holstein cows (8 with ruminal cannulae) were randomly assigned to six 4 × 4 Latin squares to investigate the effects of RDP source on lactation performance and ruminal metabolism. All diets contained (% of DM): 20.7% alfalfa silage and 35.1% corn silage. The following protein supplements were added to the basal diet (% of DM): urea (1.9%; diet A), solvent soybean meal [(SSBM; 12.1%); diet B]; cottonseed meal [(CSM; 14.1%); diet C]; or canola meal [(CM; 16.1%); diet D]. Diets contained 16.6% CP. Yield of milk, fat, protein, and 3.5% FCM were significantly higher for cows fed diets supplemented with SSBM, CSM, or CM. Feeding CM resulted in numerically greater responses, although not always statistically different from SSBM and CSM. Cows fed diet A had the lowest DMI while cows on diet D had the highest; diet C was intermediate but not different from diet B. Milk fat content, total VFA, acetate, and propionate were similar among all diets whereas milk protein content was highest on diets B and D and lowest on diets A and C. Ruminal pH and butyrate also did not differ across diets (data not shown). Overall, use of true proteins improved N utilization versus NPN. However, greater protein yield indicated that SSBM and CM were more effective than CSM.

Item	A Urea	B SSBM	C CSM	D CM	SED	*P*>F
DMI, kg/d	22.1^{c}	24.2^{b}	24.7^{ab}	24.9^{a}	0.4	<0.01
Milk yield, kg/d	33.3^{b}	40.0^{a}	40.6^{a}	41.2^{a}	1.1	<0.01
FCM, kg/d	30.7^{b}	37.1^{a}	36.7^{a}	38.8^{a}	1.2	<0.01
Milk fat, %	3.24	3.09	2.94	3.14	0.16	0.32
Milk fat, kg/d	1.01^{b}	1.22^{a}	1.18^{a}	1.29^{a}	0.06	<0.01
Milk protein, %	2.90^{b}	3.15^{a}	2.97^{b}	3.12^{a}	0.05	<0.01
Milk protein, kg/d	0.91^{c}	1.23^{a}	1.18^{b}	1.27^{a}	0.04	<0.01
Ruminal NH_3, mM	7.8^{a}	5.9^{b}	5.7^{b}	5.9^{b}	0.53	<0.01
Ruminal total VFA, mM	88.7	92.1	90.3	89.9	2.85	0.68
Ruminal acetate, mM	53.1	55.7	56.3	54.7	1.60	0.22
Ruminal propionate, mM	19.9	21.6	19.0	20.6	1.26	0.22

Key Words: Protein Supplements, Milk Yield, Dairy Cows

341 Effects of different protein supplements on nitrogen utilization in dairy cows. II. Digesta flow and bacterial protein synthesis. A. F. Brito*[1] and G. A. Broderick[2], [1]*University of Wisconsin, Madison,* [2]*US Dairy Forage Research Center, Madison, WI.*

Eight multiparous Holstein cows fitted with ruminal cannulae were randomly assigned to two 4×4 Latin squares to investigate the effects of RDP source on digesta flow and bacterial protein synthesis. All diets contained (% of DM): 20.7% alfalfa silage and 35.1% corn silage. The following protein supplements were added to the basal diet (% of DM): urea (1.9%; diet A), solvent soybean meal [(SSBM; 12.1%); diet B]; cottonseed meal [(CSM; 14.1%); diet C]; or canola meal [(CM; 16.1%); diet D]. Diets contained 16.6% CP. Digesta flow (Co-EDTA and $YbCl_2$) and bacterial markers (^{15}N) were continuously infused into the rumen. The omasal sampling technique was used to collect digesta from the omasal canal. Both DM and OM flows were lowest for cows fed diet A but similar among diets B, C, and D. Apparent ruminal dry matter digestibility (ARDMD) was highest on diet A, lowest on diet C, and intermediate on diets B and D. Organic matter apparently digested in the rumen (OMADR) and organic matter truly digested in the rumen (OMTDR) did not differ among diets. Fluid-associated bacteria (FAB), particle-associated bacteria (PAB), total bacterial flow, and bacterial efficiency were all higher in cows fed SSBM, CSM, or CM. Overall, bacterial nonammonia nitrogen (NAN) flow and efficiency were increased by feeding true protein supplements rather than a NPN source (urea).

Item	A urea	B SSBM	C CSM	D CM	SED	*P*>F
DMI, kg/d	21.4^{b}	23.6^{a}	24.1^{a}	24.7^{a}	1.0	0.03
DM flow, kg/d	13.0^{b}	15.1^{a}	16.2^{a}	16.5^{a}	0.7	<0.01
OM intake, kg/d	20.0^{b}	21.8^{ab}	22.2^{a}	22.8^{a}	1.0	0.05
OM flow, kg/d	10.7^{b}	12.4^{a}	13.4^{a}	13.3^{a}	0.6	<0.01
ARDMD, %	39.5^{a}	36.2^{b}	32.4^{c}	34.6^{bc}	1.2	<0.01
OMADR, kg/d	9.3	9.4	8.8	9.4	0.5	0.55
OMTDR, kg/d	14.2	15.7	15.8	16.0	0.7	0.12
FAB NAN flow, g/d	173^{b}	194^{ab}	180^{b}	205^{a}	11	0.05
PAB NAN flow, g/d	199^{b}	235^{a}	249^{a}	234^{a}	16	<0.01
Total bacteria NAN flow, g/d	371^{b}	429^{a}	428^{a}	439^{a}	21	<0.01
Bacterial efficiency, g/kg OMTDR	27.7^{b}	30.6^{a}	31.5^{a}	31.2^{a}	0.8	<0.01

Key Words: Protein Supplements, Bacterial NAN Flow, Dairy Cows

342 Evaluation of prediction equations for estimating urinary output of nitrogen in lactating dairy cows.

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Previously developed prediction equations need to be validated with other data sets to verify their accuracy across various diets and environments. The objective of this study was to evaluate previously published prediction equations that estimate urinary nitrogen excretion from lactating dairy cows. A data set was assembled from total urine collection studies that used multiparous Holstein cows (n = 418). The studies were conducted by Washington State University, University of California-Davis, The Ohio State University, and the University of Wisconsin. Milk production ranged from 1.4 to 86.1 kg/d. Dietary crude protein concentration averaged 17.5±5.8 and urinary nitrogen (UN) ranged from 63 to 499 g/d. Examples of previously published equations that were evaluated included UN (g/d) = N intake (g/d) x 0.83 milk N (g/d) 97 (J. Dairy Sci. 82:1261-1273) and UN (g/d) = 0.0259 x BW (kg) x MUN (mg/dl) (J. Dairy Sci. 84:2284-2294). Equations were evaluated by plotting residuals versus predicted values. The linear regression line of these plots was used to determine if mean or slope biases were present for any of the equations. The mean bias was not significant for any of the equations evaluated. However, there were significant slope biases for each of the equations. The new regression equation developed using this data set was UN (g/d) = 0.09 x DIM 0.94 x Milk (kg) + 0.20 x BW (kg) + 6.07 x DMI (kg) + 4.70 x MUN (mg/dl) + 11.42 x Dietary CP (%) - 281.53. Evaluation and validation of prediction equations is important to develop equations that will more accurately estimate urinary nitrogen excretion from lactating dairy cows.

Key Words: Rrine, Nitrogen, MUN

343 Meta analysis of the influence of different sources of methionine on the milk protein content of dairy cows.

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Several experiments have been carried out to study the influence of HMBi (2- hydroxy – 4 (methyl thio) butanoic acid isopropyl ester) on the milk protein content, at different doses and in comparison with HMB and Smartamine^{TM} M (Smartamine^{TM} M (Sm M) Adisseo) a coated form of methionine. The aim of this work was to compare the efficiency of HMBi on milk protein content vs other sources of methionine. A meta analysis was performed on a database including data from seven experiments. The rations were formulated to be adequate in term of metabolisable lysine supply. The data set was weighted by the number of cows in the experiment and the study effect was tested in the analysis. A first meta analysis was carried out in order to predict the increase of the milk protein content (MPC, g/kg) in relation to the quantity of methionine distributed to the cows (MQ ; g /animal/day). This took into account the methionine percentage of each product (70% for HMBi, 78% for Smartamine^{TM} M and 88 % for HMB). The relationships were: for HMBi: MPC = 0.277 ln MQ (R^2 =0.83;SRD=0.12) ; for HMB: MPC = 0.001 ln MQ (R^2 =0;SRD=0.15) and for Sm M: MPC = 0.343 ln MQ (R^2 =0.85 ; SRD=0.14). Furthermore the following estimates of methionine bioavailability (50% for HMBi, 40% for HMB and 80% for Sm M) were used to calculate the quantities of metabolisable methionine supplied. A second meta analysis was performed in order to predict the increase of MPC depending on the quantity of metabolisable methionine (MMQ, g/animal/day). The relationships were: for HMBi and Sm M: MPC = 0.357 ln MMQ (R^2 =0.77; SRD= 0.11) ; for HMB: MPC = 0.002 ln MMQ (R^2=0 ; SRD=0.15). Thus, the efficiency of utilization of both HMBi and Smartamine^{TM} M at equivalent quantities of metabolisable methionine were not found to be significantly different. Moreover this meta analysis illustrates that only by conjugating HMB with isopropanol was sufficient HMB absorbed to increase the supply of metabolisable methionine necessary to increase MPC.

Key Words: Methionine, Ruminant, Nutrition

344 Use of changes in plasma sulfur amino acid concentrations to compare the ability of methionine (Met) products to provide absorbable Met to lactating dairy cows fed a Met-adequate diet.

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This experiment is part of an ongoing effort to develop an accurate and repeatable approach for determining differences among Met products in their ability to provide absorbable Met to lactating cows. Fifteen multiparous Holstein cows (212 ± 162 DIM) were blocked by milk yield, DIM, and milk protein content and assigned randomly to one of three 5 x 5 Latin squares. The basal diet was formulated to meet NRC (2001) requirements for energy and nutrients, including content of Met in metabolizable protein (MP). Adequacy of Met was achieved by adding Smartamine^{TM} M to the basal diet to increase Met in MP from 1.76% to 2.14% to achieve an approximate 1:3 ratio with lysine in MP (6.51%). Each square consisted of top-dressing the basal diet with five amounts (treatments) of a single Met source. The three Met sources were Smartamine^{TM} M, Met-Plus^{TM} and Mepron M85$^{®}$. Products were added to the basal diet to provide levels of Met supplementation of 0, 0.012, 0.024, 0.036 and 0.048% of DM. Treatments had no effect on concentration of total free AA in deproteinized plasma. Percent of Met and Met+cysteine (Cys) in total plasma AA for the five treatments were: Smartamine^{TM} M (1.95, 2.06, 2.23, 2.48, 2.44; linear, $P < 0.01$ and 3.21, 3.42, 3.62, 3.84, 3.89; linear, $P < 0.01$), Met-Plus^{TM} (1.88, 1.91, 1.99, 2.05, 1.98; $P = 0.27$ and 3.25, 3.40, 3.41, 3.41, 3.49; $P = 0.168$) and Mepron M85$^{®}$ (1.81, 2.16, 2.11, 2.03, 2.23; linear, $P = 0.03$ and 3.29, 3.65, 3.57, 3.48, 3.62; $P = 0.168$). Concentrations of Met and Met+Cys in total plasma AA were regressed on levels of Met supplementation. The slopes for Met-Plus^{TM} and Mepron M85$^{®}$ were divided by the slope for Smartamine^{TM} M to obtain a Met-bioavailability relative to that of Smartamine^{TM} M. The described approach indicates that Met-Plus^{TM} was 24% and 28% as effective as Smartamine^{TM} M and Mepron M85$^{®}$ was 51% and 28% as effective as Smartamine^{TM} M using changes in plasma Met and Met+Cys concentrations, respectively, in providing absorbable Met to lactating cows.

Key Words: Lactating Dairy Cows, Rumen Protected Methionine, Plasma Amino Acids

345 A simplified *in vitro* incubation medium with the potential to evaluate amino acid degradation in ruminants.

F. L. Mould*, R. Morgan, and K. Kliem, *Deprtment of Agriculture, The University of Reading.*

Most *in vitro* assays use a modified Goering and Van Soest [1970] incubation medium [GVS]. However as some of the components are hazardous, difficult to obtain [developing countries] or of little impact [e.g. trace minerals] their omission should be examined. Hungate [1966] considered osmotic potential and buffering capacity to be more vital than composition, while Tilley and Terry [1963] argued that sufficient trace elements and growth stimulants were provided by the inoculum or substrate and that fermentation maintained anaerobic conditions. A modified [MOD] N-free medium based on GVS was prepared without the reducing solution or trace minerals and with NH_4HCO_3 replaced with $NaHCO_3$. The Reading Pressure Technique was used to compare these media with respect to the fermentation of maize starch and where MOD was supplemented with urea up to N levels equivalent to that supplied by GVS. The rumen fluid inoculum provided 10 mg N per flask. Cumulative gas values 4, 8, 12 and 24 h post-inoculation of 5.3, 27.6, 68.9 and 103.6 and 6.0, 32.8, 77.7 and 112.7 ml were recorded for GVS and MOD + nitrogen, respectively. The similarity between the two profiles indicates that a highly simplified, non-reduced medium is sufficient. With the urea dose titration, peak rate of gas release occurred 10 h post-inoculation and corresponded with the time point with the greatest differentiation between levels. Using the assumption that the urea-N was totally available, a highly significant regression between these 10 h gas values and supplemental N [r^2=0.929, P>0.0001] was obtained. This indicates that gas release kinetics from the fermentation of a N-free substrate can be used as a complement assay to estimate supplemental N availability. Further, the comparison of gas profiles from known N sources with those of amino acids should permit the extent of their utilisation by rumen microorganisms to be determined, and subsequently the degree to which commercial preparations confer amino acids protection against rumen degradation to be assessed.

Key Words: In Vitro, Simplified Medium, Amino Acids

346 Ruminal degradation of amino acids assessed using a complement *in vitro* technique. F. L. Mould*, K. Kliem, and R. Morgan, *Department of Agriculture, The University of Reading*.

Lysine and methionine are considered the first limiting amino acids in ruminant nutrition, however consistent treatment responses have generally only been found with maize-based diets offered to high-yielding dairy cattle. The potential loss of "protein value" or decreased efficiency of use following microbial degradation led to the concept of protecting free amino acids from rumen microbial degradation. However the extent of this degradation is unclear. A novel complement *in vitro* methodology was used to examine amino acid utilisation by rumen microorganisms. This technique uses the highly significant relationship between supplemental nitrogen availability and the rate of fermentation gas release. Nine amino acids [Ala, Gln, Glu, Gly, His, Lys, Phe, Met and Ser] were compared to urea at 8, 12 and 16 mg N g^{-1} maize starch. Each flask contained 1.0 g starch with the rumen fluid inoculum supplying 10 mg N. Each treatment was replicated five times with 16 h cumulative gas values obtained using the Reading Pressure Technique. Fermentation gas release [ml] for the three N inclusion levels [8, 12 and 16 mg] were 114, 118, 159, 97, 90, 85, 104, 92, 131 and 182; 120, 126, 174, 89, 91, 85, 102, 95, 136 and 216; and 111, 132, 197, 91, 86, 89, 91, 103, 135 and 246 for the amino acids, respectively. Compared with the control [0 mg N] value of 85 ml these results suggest that Gly, Hist, Lys and Phe were stable, Met marginal with only Gln and to a lesser extent Glu, Ala and Ser labile. AA-N utilisation, estimated by expressing gas release less the control values, relative to that of urea gave efficiencies of 0.243, 0.314, 0.712, 0.064, 0.035, 0.007, 0.125, 0.084, 0.391 for Ala, Gln, Glu, Gly, His, Lys, Phe, Met and Ser, respectively. If these results are confirmed *in vivo* where factors such as rumen absorption and outflow are considered, it is hypothesised that, provided adequate quantities are supplied [e.g. Met 120 % of requirement] "rumen protection" may not be needed to deliver sufficient levels of specific free amino acids post-ruminally.

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