

Evaluation of Ruma Pro (a calcium-urea product) on microbial yield and efficiency in continuous culture

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OBJECTIVES

To evaluate effects of RumaPro on fermentation parameters in continuous cultures operated to simulate the rumens of lactating cows (Experiment 1) and close-up dry cows (Experiment 2).

PROCEDURES

The major differences between lactating cows and close-up dry cows, in terms of rumen function, are primarily in quantity of feed intake and in rumen turnover rates. In these studies, the following conditions were used:

	Experiment 1	Experiment 2
Item	(Lactating)	(Close-up)
Liquid dilution rate, %/hr	12	10
Solids retention time, hr	22	27
Feed intake, g/d	100	60
Feeding frequency, times/day	2	2
pH	Monitored	Monitored

Diet composition for a close-up dry cow should be similar to that of a lactating cow after peak production. Because in this study the lactation ration was balanced for peak production, the close-up ration had a slightly lower nutrient content. Composition and analysis of the lactation and close-up rations are in Table 1 and 2, respectively.

Experiment 1, the lactation study, had four diets: a control in which soybean meal (SBM) and urea were the major protein sources, and three experimental diets. In the first experimental diet, RumaPro replaced the urea in the control; in the second experimental diet, RumaPro replaced the urea and 16.5% of the SBM, and in the third experimental diet, RumaPro replaced the urea plus 100% of the SBM.

Three close-up diets were used; a control, with urea and SBM, and two experimental diets in which RumaPro replaced the urea along with 35 and 94% of the SBM.

TABLE 1. Diet Composition and Analyses-Lactation Rations
(% Dry Matter Basis)

Ingredients	Control	RumaPro 1	RumaPro 2	RumaPro 3
Corn Silage	27.78	27.65	27.80	33.14
Haylage	18.52	18.43	18.54	23.15
Ground Corn	31.85	31.71	33.83	35.19
SBM 44	19.07	18.99	15.94	-----
Urea	0.37			-----
MgO	0.111	0.111	0.111	0.222
TM Salt	0.222	0.221	0.222	0.185
Limestone	0.556	0.553	0.371	-----
Dicalcium Phosphate	0.222	0.221	0.222	0.611
Sodium Bicarbonate	1.30	1.29	1.30	1.30
RumaPro	-----	0.829	1.67	6.20

Analyses				
Crude Protein	17.39	17.67	17.52	18.09
Soluble Protein, %CP	26.63	32.31	32.76	72.01
Neutral Detergent Fiber	27.89	28.51	28.25	30.54
Acid Detergent Fiber	17.90	18.71	18.84	20.44
Nonstructural Carbohydrate	37.67	38.62	41.70	37.48
Starch	34.42	36.91	39.98	35.75
Sugar	3.25	1.71	1.72	1.73
Ether Extract	2.88	2.91	2.71	2.82
Ash	6.18	6.31	6.04	6.32
Calculated NFC ¹	45.66	44.60	45.48	42.23

¹Non Fiber Carbohydrate

The major differences in analyses between the lactation and close-up rations were that the close-up rations had less total protein and nonstructural carbohydrate, and more fiber than did the lactation rations.

Each treatment was fermented in triplicate. The results were summarized and analyzed statistically as two separate experiments. Statistical analyses were performed using SAS.

In Experiment 1 comparisons were:

Control vs the mean of all RumaPro levels
 RumaPro 1 vs RumaPro 2
 RumaPro1 + 2 vs RumaPro 3

In Experiment 2, comparisons were:

Control vs the mean of both Ruma Pro levels
 RumaPro 1 vs Ruma Pro 2

RESULTS

Experiment 1

Digestion coefficients are shown in Table 3. Digestibilities of dry and organic matter were equal to that of the control for all treatments, with the numbers favoring RumaPro level 2 in which all the urea and 16.5% of the SBM were replaced by RumaPro.

Responses of fiber digestion, both NDF and ADF, were similar to the control for RumaPro levels 1 and 2. Fiber digestion for RumaPro level 3, however, was significantly lower than that of levels 1 and 2, and, since levels 1 and 2 were equal to the control, it can be assumed that level 3 was significantly lower than the control as well. All levels of RumaPro were equal to the control in supporting nonstructural carbohydrate (NSC) digestion. As a result of the combined fiber and NSC digestion, the total carbohydrate actually digested in g/d were greatest for RumaPro level 2, which was significantly higher than level 1.

Since there were no significant effects of the treatments on dry or organic matter digestion, it is not surprising that there were no differences in total VFA produced/day, as shown in Table 4. There were, however, changes in molar proportions of fatty acids, particularly acetic and butyric. The combined treatments had a significantly higher level of acetate than did the control, and the proportion of acetate appeared to increase with each increase in RumaPro. Butyrate responded in exactly the opposite manner, decreasing with increasing RumaPro. In the rumen, acetate and butyrate are in equilibrium. It appears that acetate is increased at the expense of butyrate, which is probably beneficial, as butyrate can be toxic in ruminants. Propionate was largely unaffected by RumaPro, but there was a trend ($p=.08$) for RumaPro level 3 to have higher propionate than levels 1 and 2. Acetate-propionate ratio was not significantly affected by treatments.

Average daily pH was higher for the RumaPro diets than for the control, largely due to the effects of level 3. A plot of pH over time after feeding is presented in Figure 1. At 2 hours post-feeding, level 2 maintained a higher pH ($p=.02$) than did the control or level 1, but no differences among the control and RumaPro levels 1 and 2 were found for the remainder of the time after feeding. RumaPro level 3 actually increased pH after feeding compared to the other treatments, and maintained a higher pH throughout the feeding period. In spite of the higher pH for level 3, fiber digestion was significantly lower for level 3 compared to levels 1 and 2 (Table 3). The higher rumen pH caused by RumaPro may well be responsible for the higher acetate found on the treatments. The reason, however, for the higher pH caused by RumaPro is not clear, but it certainly is a positive effect of the product.

Nitrogen partitioning and microbial growth are shown in Table 5. Protein digestion was unaffected by RumaPro, except at the highest level, where digestion tended ($p=.13$) to be higher than for levels 1 and 2. Although not statistically significant, levels 1 and 2 had numerically lower protein digestion than did the control. When RumaPro was substituted for urea only (level 1), the result was the least ammonia production for all treatments and lower microbial growth than the control. This demonstrates that the availability of RumaPro nitrogen was slower than that from urea. The addition of RumaPro nitrogen at a higher level (level 2) was sufficiently available to enhance protein digestibility, increase ammonia slightly and bring microbial protein back to the control level. Protein digestion was highest at the highest level of RumaPro, and should have provided more N for microbial growth, but this was not the case. Much of the N was lost as ammonia, which was significantly greater than on all other diets ($p=.01$). This resulted in the lowest by-pass of feed-N as well as the lowest microbial N production of the three treatment levels. The inefficient N use for microbial growth is clearly shown in the Feed N efficiency value of 74.9%, which is considerably lower than for all other diets. *(continued)*

The efficiency of conversion of organic matter or carbohydrates to microbial mass also was only affected by the highest level of RumaPro. Thus, as with most responses seen, RumaPro at level 2 was superior to levels 1 and 3, and was equal to SBM in supporting microbial growth and metabolism.

TABLE 3. Digestion Coefficients for Experiment 1 – Lactation Rations

Nutrient	Diets				P=% Digested		
	Control	RumaPro 1	RumaPro 2	RumaPro 3	Cont vs Trts	RumaPro 1 vs 2	RumaPro 1, 2 vs 3
Dry Matter	78.3	77.7	78.4	80.1	NS	NS	NS
Organic Matter	57.4	55.2	61.6	59.7	NS	.08	NS
Neutral Detergent Fiber	37.1	37.5	35.6	31.2	NS	NS	.01
Acid Detergent Fiber	37.2	38.1	38.4	27.9	NS	NS	.01
Nonstructural Carbohydrate	83.8	77.1	80.9	84.3	NS	NS	NS
Total Carbohydrate digested grams/d	42.2	40.8	45.1	41.3	NS	.02	NS

Figure 1. Fermentation pH
RumaPro Project - Lactation Diets

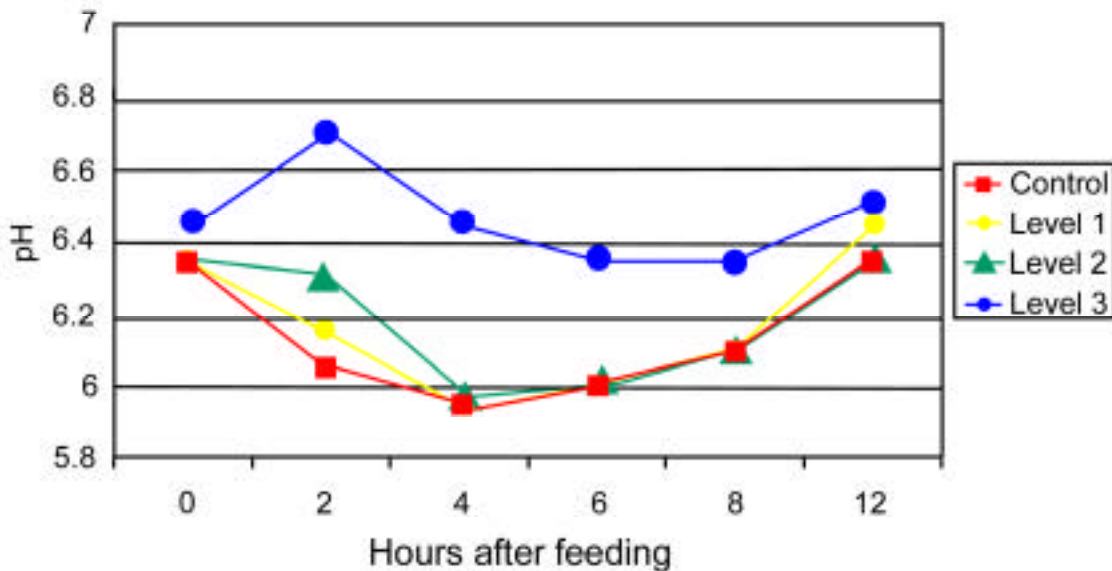


TABLE 4. Volatile Fatty Acid Production, Molar Ratios and pH for Experiment 1 – Lactation Rations

Item	Diets				Cont vs Trts	P=% Digested		
	Control	Ruma Pro 1	Ruma Pro 2	Ruma Pro 3		Ruma Pro 1 vs 2	Ruma Pro 1, 2 vs 3	
Total VFA, mm/day	352	353	362	356	NS	NS	NS	
Molar Proportions, %NS.08NS	57.4	55.2	61.6	59.7	NS	.08	NS	
Acetic	47.3	48.0	53.8	55.8	.04	.06	.06	
Propionic	27.2	26.4	24.6	30.7	NS	NS	.08	
Isobutyric	0.67	0.65	0.66	0.52	NS	NS	NS	
Butyric	21.5	20.6	17.0	9.1	.01	.02	.01	
Isovaleric	1.02	1.19	1.34	1.14	NS	NS	NS	
Valeric	2.28	3.15	2.68	2.57	NS	NS	NS	
pH, average/day	6.14	6.17	6.18	6.46	.03	ns	.01	
Acetate/Propionate ratio	1.74	1.82	2.20	1.91	NS	NS	NS	

TABLE 5. Nitrogen Partitioning, Microbial Growth and Microbial Efficiency for Experiment 1 – Lactation Rations

Item	Diets				Cont vs Trts	P=% Digested		
	Control	Ruma Pro 1	Ruma Pro 2	Ruma Pro 3		Ruma Pro 1 vs 2	Ruma Pro 1, 2 vs 3	
Nitrogen Intake, g/day	352	353	362	356	NS	NS	NS	
Nitrogen Digested, %	87.2	81.2	85.3	89.0	NS	NS	.13	
Non-ammonia N, g/day	2.88	2.97	2.87	2.48	.02	.07	.01	
Ammonia N, mg/dl	6.12	6.10	6.72	21.35	.01	NS	.01	
By-pass N, g/day	0.40	0.59	0.456	0.35	NS	NS	NS	
Microbial N, g/day	2.48	2.62	2.42	2.13	.10	NS	NS	
Efficiencies:								
Mic N/kg OMD ¹	46.1	43.8	41.8	38.2	.09	NS	.12	
Mic N/kg CHOD ²	59.2	55.9	55.3	51.7	.01	NS	NS	
Feed N ³	92.3	93.3	91.4	74.9	.01	NS	.01	

¹Microbial N produced/kg organic matter digested

²Microbial N produced/kg carbohydrate digested

³Digested feed N converted to microbial N, %

EXPERIMENT 2**TABLE 2. Diet Composition and Analyses-Close-up Ratios (% Dry Matter Basis)**

<u>Ingredient</u>	<u>Control</u>	<u>RumaPro 1</u>	<u>RumaPro 2</u>
Corn Silage	38.61	41.97	44.35
Haylage	25.74	25.70	26.90
Ground Corn	21.45	21.41	24.35
SBM44	13.94	8.99	0.87
Urea	0.257	-	-
RumaPro	-	1.93	4.35
Analyses			
Crude Protein, % CP	31.39	41.56	72.77
Neutral Detergent Fiber	15.08	14.30	15.41
Soluble Protein	35.60	35.56	36.43
Acid Detergent Fiber	23.78	23.47	24.81
Nonstructural Carbohydrate	31.78	34.47	36.63
Starch	28.62	32.02	35.04
Sugar	3.16	2.45	1.59
Ether Extract	2.82	3.18	3.33
Ash	3.53	4.85	5.16
Calculated NFC ¹ ;1;39.67	42.97	42.11	39.67

¹Non Fiber Carbohydrate

Experiment 2

Digestion coefficients are shown in Table 6. As was seen in Experiment 1, no effects on dry matter or organic matter digested due to treatment were noted. Fiber digestion, however, was increased ($p=.08$) by both levels of RumaPro compared to the control, and the response appeared to increase with increased level of RumaPro in the diet. Although RumaPro caused a slight, but statistically significant, decrease in NSC digestion, the combined fiber and NSC digested was greater ($p=.03$) for the RumaPro diets than for the control diet, when determined as total carbohydrate digested in g/d.

Effects on volatile fatty acids were similar to those seen in Experiment 1, which was an increase in acetate and a decrease in butyrate, as shown in Table 7. In Experiment 2, however, the increased acetate was sufficient to cause significant increases in the acetate-propionate ratio for both treatments.

Although the average pH did not differ due to the treatments (Table 7), both treatments resulted in higher fermentation pH ($p=.10$) compared to the control at hours 2, 4 and 6 post-feeding (Figure 2). This is consistent with the higher fiber digestion seen with both RumaPro treatments.

Nitrogen partitioning is presented in Table 8. Nitrogen digestion was not affected by treatment. Flow of non-ammonia N, by-pass feed N and microbial N all were lower for the average of the treatments than for the control. These results were primarily caused by the responses to the highest level of RumaPro. Responses to the lower level of RumaPro appeared similar to those of

the control. As seen in Experiment 1, ammonia was low for the RumaPro diet at level 1, indicating very slow release of N, probably at a rate similar to that of SBM. As a consequence of the higher carbohydrate digestion and no increase in microbial yield on the RumaPro diets, carbohydrate and organic matter efficiencies were lower than for the control. As seen in the previous experiment, the highest level of RumaPro resulted in higher ammonia losses and lower feed N efficiency compared to either the control or the lower level of RumaPro.

**Figure 2. Fermentation pH
RumaPro Project-Close-up Diets**

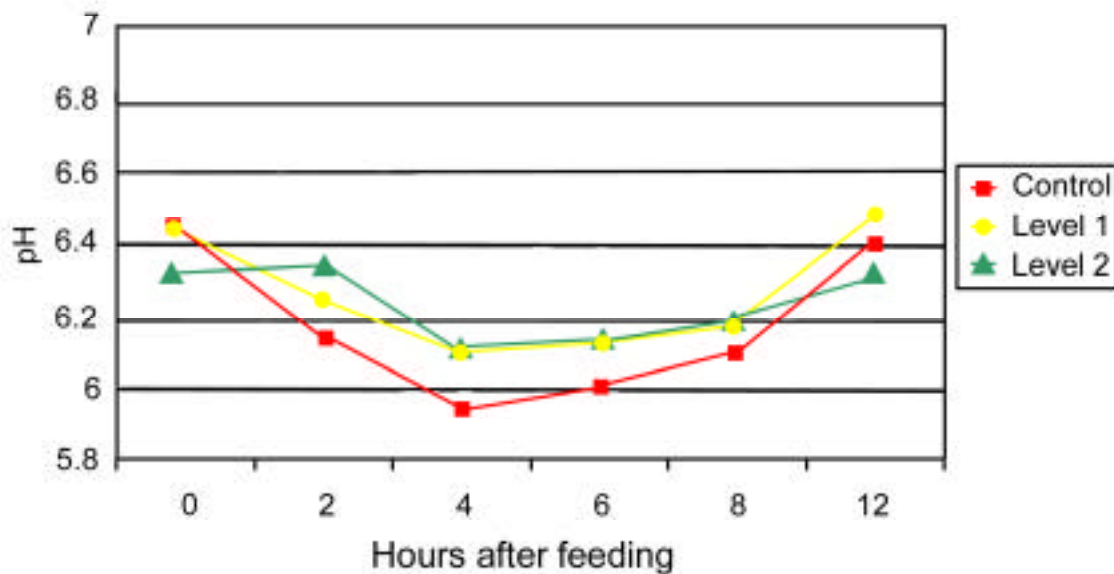


TABLE 6. Digestion Coefficients for Experiment 2 – Close-up Rations

Nutrient	Diets			P=% Digested	
	Control	RumaPro 1	Ruma Pro 2	Cont vs Trts	RumaPro 1 vs 2
Dry Matter	77.3	73.1	76.0	NS	NS
Organic Matter	53.4	55.6	58.0	NS	NS
Neutral Detergent Fiber	27.2	33.7	37.1	.08	NS
Acid Detergent Fiber	35.3	32.4	53.6	.08	.07
Nonstructural Carbohydrate	90.9	92.0	87.4	.05	.01
Total Carbohydrate digested Grams/d	22.1	25.4	26.5	.03	NS

TABLE 7. Volatile Fatty Acid Production, Molar Ratios and pH for Experiment 2 – Close-up Rations

Nutrient	Diets			P=%Gain	
	Control	Ruma Pro 1	Ruma Pro 2	Cont vs Trts	Ruma Pro 1 vs 2
Total VFA, mm/day	238	235	229	NS	NS
Molar Proportions, %					
Acetic	55.2	60.6	61.6	.02	NS
Propionic	23.4	20.2	21.8	NS	NS
Isobutyric	0.60	0.77	0.49	NS	.01
Butyric	16.3	13.7	11.5	.01	.01
Isovaleric	1.88	2.20	2.19	NS	NS
Valeric	2.61	2.53	2.47	NS	NS
pH, average/day	6.186	6.26	6.26	NS	NS
Acetate/Propionate ratio	2.39	3.04	2.83	.10	NS

TABLE 8. Nitrogen Partitioning, Microbial Growth and Microbial Efficiency for Experiment 2 – Close-up Rations

Nutrient	Diets			P=%Gain	
	Control	Ruma Pro 1	Ruma Pro 2	Cont vs Trts	Ruma Pro 1 vs 2
Nitrogen Intake, g/day	2.67	2.54	2.72	-	-
Nitrogen Digested, %	72.7	66.3	70.0	NS	NS
Non-ammonia N, g/day	1.57	1.52	1.46	.07	NS
Ammonia N, mg/dl	4.81	4.23	10.02	NS	.01
By-pass N, g/day	0.47	0.55	0.52	NS	NS
Microbial N, g/day	1.11	0.97	0.94	.07	NS
Efficiencies:					
Mic N/kg OMD ¹	36.2	30.7	28.6	.13	NS
Mic N/kg CHOD ²	49.7	37.2	34.5	.02	NS
Feed N ³	89.1	89.1	77.2	NS	.02

¹Microbial N produced/kg organic matter digested

²Microbial N produced/kg carbohydrate digested

³Digested feed N converted to microbial N, %

IMPLICATIONS AND CONCLUSIONS

Responses to RumaPro were similar when included in both lactation and dry cow rations. Relative to the soybean meal-urea control, results of these studies indicate important responses to RumaPro in several aspects of rumen function. Because of the apparently slow rate of N release, RumaPro is best substituted for a portion of the natural protein in the diet, in this case, soybean meal. Indications from both studies put the level of substitution between 16.5% and 35% of the SBM nitrogen. At the levels of substitution of 16.5 to 35% of SBM nitrogen, RumaPro improved total carbohydrate digestion, increased rumen pH, increased acetate proportion and decreased butyrate production; all positive responses, which have implications in terms of improved milk fat production. At these levels, the use of RumaPro nitrogen was equal to that of SBM as a source of N for microbial growth. This is shown by both microbial nitrogen production and the efficiencies of conversion of feed nitrogen to microbial nitrogen.

The increases in rumen pH may well be at least partly responsible for these responses. It was initially thought that the rumen pH was increased by extensive ammonia release. The low daily averages for ammonia cast doubt on this possibility, however. Further, in our experiences, no feed (including urea at high levels) has resulted in a pH increase after feeding as happened in both Experiment 1 and 2. It is suspected that RumaPro may have a strong buffering effect independent of ammonia.

It can be concluded, based on these studies that:

1. RumaPro is a slow-release source of N.
2. It may be substituted for soybean meal at 16.5 to 35% of SBM nitrogen, on an equal N basis.
3. When substituted at these levels it is equivalent, but not superior to, SBM nitrogen for supporting microbial growth.
4. RumaPro should not be used to replace all the natural protein supplement; however, the absolute upper limit to substitution was not determined in this study.