

## Ruminant Nutrition: Ruminal Fermentation

**491 A meta-analysis of the effects of fumarate on ruminal methanogenesis.** E. M. Ungerfeld\* and R. A. Kohn, *University of Maryland, College Park*.

The objective of this analysis was to understand the effects of fumarate addition on ruminal fermentation. Because the chemical reduction of fumarate to succinate can draw electrons away from ruminal methanogenesis, fumarate has been studied as a potential feed additive to decrease methane (CH<sub>4</sub>) production in ruminants. A meta-analysis of the effects of added fumarate on CH<sub>4</sub> production by ruminal batch cultures was carried out using 74 treatment means from 9 experiments in 8 different studies. Treatment means were weighted by the reciprocal of their variance. All regressions included the random effect of experiment. Interactions between the random effect of experiment and independent variables were not significant and were removed from models. The average decrease in CH<sub>4</sub> was of 0.037 μmol/μmol of added fumarate, which is considerably lower than 0.25, the theoretical decrease calculated from stoichiometries of the pathways involved. One reason fumarate was not very effective at decreasing CH<sub>4</sub> was because only an average of 48% of added fumarate appeared to be converted to propionate. Secondly, the uptake of reducing equivalents by the conversion of fumarate to propionate was almost entirely offset by the release of reducing equivalents from 20% of added fumarate that was converted to acetate. Therefore, fumarate addition must have directly or indirectly stimulated other electron sinks to result in the small net decrease in CH<sub>4</sub>. Succinate accumulation, which was generally not measured, may account for some of the decrease in CH<sub>4</sub> observed. Fumarate addition does not seem to stimulate another potential electron sink, microbial biomass. Thermodynamic calculations indicate that the conversion of added fumarate to both propionate and acetate initially was feasible. Thus, along with electron incorporation, the addition of electron-accepting intermediates may also result in the release of electrons through alternate pathways. Thermodynamic considerations should be taken into account when designing these strategies for CH<sub>4</sub> abatement through external electron acceptors.

**Key Words:** Rumen, Methane, Fumarate

**492 Implications of a carbon balance study: Organic acid and protein supplies change with fermentable carbohydrate:protein ratio.** M. B. Hall\* and P. J. Weimer, *USDFRC, USDA-ARS, Madison, WI*.

Three concentrations of sucrose (Suc, trt) with one of isolated neutral detergent fiber (isoNDF) from bermudagrass were fermented together in vitro with rumen inoculum to evaluate the effects of Suc concentration on partition of carbon (C) in Suc into fermentation products. Nitrogen sources were inoculum, isoNDF and casein hydrolysate and NH<sub>4</sub>HCO<sub>3</sub> in the medium. Yield of C in fermentation products [organic acids (OA:acetate, propionate, butyrate, lactate), microbial crude protein (MCP), CO<sub>2</sub>, CH<sub>4</sub>, glycogen] from C from fermented Suc was evaluated at peak MCP production from Suc (detected at 4 to 8 h of fermentation; Suc almost entirely fermented at this point). Yield of total products, MCP and OA decreased or tended to decrease linearly with increasing Suc. This may be a function of decreased catabolic efficiency of the microbes with increasing Suc as evidenced by increasing yields of lactate, but increased energy spilling is also a possibility. Values > 1.0 indicate incorporation of C from the medium, likely from the inoculum and casein hydrolysate. When corrected for estimates of microbial cell C not in MCP, product C from

Suc for the greatest Suc trt was 0.96. Even with C from MCP excluded, yield of C in other products per unit of Suc utilized still differed among trt (1.03, 0.77, and 0.61 from lowest to greatest Suc inclusion; linear effect of Suc, P = 0.017; SED = 0.054). In this study, the ratio of available protein to fermentable Suc decreased with increasing Suc. Published studies report increased yield of MCP in vitro and increased ruminal acid concentrations in vivo as the ratio of degradable protein:fermented carbohydrate was increased. Results of this C balance support the premise that the ratio of available N to Suc and direct effects of Suc concentration altered partitioning of C into products. This has implications for prediction of ruminal pH and supply of nutrients from ruminal fermentation.

**Table 1. Yield of C in products from fermented Sucrose C (mg/mg)**

Trt	Suc C, mg	Total product C, mg	Product yield	MCP yield	OA yield	Glycogen yield	Gas yield
Suc65	27.4	32.8	1.28	0.257	0.822	-0.025	0.230
Suc130	54.7	51.6	0.98	0.215	0.602	-0.029	0.193
Suc195	82.1	63.5	0.81	0.204	0.493	-0.029	0.147
SED	---	3.80	0.047	0.020	0.068	0.047	0.038
p-values	---	0.015	0.010	0.121	0.040	0.934	0.158

Number in Trt = Suc mg fermented with 130 mg isoNDF; Gas as CO<sub>2</sub> + CH<sub>4</sub> predicted from theoretical stoichiometries based on OA production; p-values describe linear effect of Suc determined with orthogonal contrasts. PROC MIXED model = fermentation run, suc, run\*suc.

**Key Words:** Rumen fermentation, Carbohydrate, Degradable protein

**493 Effects of grain induced subacute ruminal acidosis on ruminal lipopolysaccharide and inflammation in Holstein cows.** G. N. Gozho, J. C. Plaizier\*, and D. O. Krause, *University of Manitoba, Winnipeg, MB, Canada*.

The effects of grain induced subacute ruminal acidosis (SARA) on free ruminal lipopolysaccharide (LPS) and the acute phase proteins serum amyloid A (SAA) and haptoglobin (Hp) in peripheral blood serum were determined in four mid lactation Holstein dairy cows using a repeated switchover design with four 7 day experimental periods. During each period, SARA was induced in two animals for 5 subsequent days by replacing 25% of their total mixed ration on a dry matter basis with grain pellets consisting of 50% ground wheat and 50% ground barley, and two other cows acted as controls. The control cows received a total mixed ration containing 50% of dry matter as concentrate. Rumen pH was monitored continuously using indwelling probes. During one day in each period blood was collected at 9 am and at 3 pm, and rumen fluid was collected at six hour intervals. Data was evaluated by SAS Mixed procedure for repeated measures with treatment (SARA or control), sampling time and their interaction as fixed effects, and cow and experimental period as random effects. On average, inducing SARA reduced average daily rumen pH from 6.24 to 6.01, increased the duration of time below rumen pH 5.6 from 187 to 309 min/d, increased LPS from 22,908 to 147,910 EU/mL, increased SAA in blood serum from 286.8 to 498.8 μg/mL, but did not affect the Hp, milk yield and dry matter intake. Rumen pH data and the milk fat contents suggest that, although inducing SARA reduced

rumen pH, control cows also experienced a mild form of this disease. Concentrations of LPS in blood serum of control and SARA cows was lower than the detection limit of <0.01 EU /mL. Results suggest that inducing SARA in mid lactation dairy Holstein cows increases the lysis of gram-negative bacteria. The increase in SAA suggests that SARA also activated an inflammatory response.

**Table 1.**

	Diet Control	SARA	SE	P value
DMI (TMR + grain), kg/d	18.0	18.3	1.64	0.74
Milk yield, kg/d	27.5	28.6	1.83	0.27
Milk fat, kg/d	0.68	0.63	0.04	0.43
Milk protein, kg/d	1.01	0.98	0.08	0.77
Avg. rumen pH	6.24 <sup>a</sup>	6.01 <sup>b</sup>	0.07	<0.01
Time < pH 5.6, hr/d	187 <sup>b</sup>	308 <sup>a</sup>	32.1	<0.01
LPS, Log 10 EU/mL	4.39 <sup>b</sup>	5.11 <sup>a</sup>	0.15	<0.01
SAA, ug/mL	286.8 <sup>b</sup>	498.8 <sup>a</sup>	85.6	0.03
Hp, mg/mL	0.244	0.265	0.03	0.59

a, b Means differ ( $P < 0.05$ )

**Key Words:** Subacute ruminal acidosis, Lipopolysaccharide endotoxin, Acute phase proteins

**494 The effects of rumen-degradable protein level on fermentation of corn-based diets in continuous culture.** C. J. Fu\*, J. A. Pugh, J. H. Porter, and M. S. Kerley, *University of Missouri, Columbia.*

A single-phase continuous culture system, operated at a 0.04/h dilution rate, was used to determine the effects of rumen-degradable protein (RDP) level on ruminal fermentation when cultures were fed all-concentrate diets. The microbial efficiency (MOEFF), VFA (including lactic acid: LA), pH, and digestibility were measured. The study included seven treatments with four RDP levels. The CORN diet served as the 0.5X RDP treatment, and SBM was used to adjust RDP level to 6.0 (0.75X), 8.0 (1.0X), and 16.0% (2.0X). The other three diets were UREA (urea as RDP source and equal to 1.0X), CORNPH, and UREAPH (CORN and UREA diets with pH controlled at 5.7). Data were analyzed using GLM procedure of SAS with linear and quadratic responses tested by CONTRAST statement. The results indicated that total VFA production increased and LA production decreased linearly ( $P < 0.01$ ) as RDP level increased from 0.5X to 2.0X. The 0.5X and 0.75X diets showed higher ( $P < 0.01$ ) LA production than that of 1.0X and 2.0X diets, resulting in lower pH ( $P < 0.01$ ). As RDP level increased, the MOEFF and the acetic to propionic ratio (AC:PRO) increased and decreased linearly ( $P < 0.01$ ). The ammonia level was higher ( $P < 0.01$ ) for 2.0X treatment compared to other three treatments. When the fermentor pH was controlled, the UREAPH showed lower LA production compared to UREA ( $P < 0.01$ ), however there were no differences observed for the total VFA production or AC:PRO. The MOEFF increased by controlling the pH ( $P < 0.05$ ). In contrast, the CORNPH diet did not show decreased LA production compared to the CORN diet, but showed increased total VFA production ( $P < 0.01$ ). The AC:PRO and MOEFF was not changed by controlling pH. We hypothesized that RDP level altered the dominant species of bacteria present, either by reducing the LA producing species or increasing the LA fermenting species, resulting in an increased pH and improved MOEFF. Alternatively, controlling pH could alter LA producing bacteria when N was not limiting. Concluded from this experiment was that the optimal RDP level was important in

controlling ruminal pH, and optimizing fermentation and efficiency of N use.

**Key Words:** Acidosis, All-concentrate diets, RDP

**495 Effect of roughage level and calcium magnesium carbonate on ruminal metabolism and extent of digestion in steers.** G. I. Crawford\*<sup>1</sup>, M. K. Luebbe<sup>1</sup>, G. E. Erickson<sup>1</sup>, T. J. Klopfenstein<sup>1</sup>, C. R. Krehbiel<sup>2</sup>, and G. A. Nunnery<sup>3</sup>, <sup>1</sup>University of Nebraska, Lincoln, <sup>2</sup>Oklahoma State University, Stillwater, <sup>3</sup>MIN-AD, Inc., Amarillo, TX.

Six ruminally and duodenally fistulated Holstein steers were used in a metabolism experiment to determine effects of adding a ruminal buffer to high-concentrate diets with increasing levels of roughage. A 6 x 6 Latin square design with a 3 x 2 factorial treatment arrangement was used with factors consisting of alfalfa hay level (4.5, 9.0, or 13.5%) and calcium magnesium carbonate (CaMg(CO<sub>3</sub>)<sub>2</sub>; MIN-AD<sup>®</sup>, MIN-AD, Inc., Amarillo, TX) inclusion (0 or 1.0%) to replace MgO and partially replace limestone. Ruminal pH and DMI were continuously monitored throughout each collection period. An alfalfa x CaMg(CO<sub>3</sub>)<sub>2</sub> interaction ( $P < 0.05$ ) occurred as steers consuming 13.5% alfalfa, 1.0% CaMg(CO<sub>3</sub>)<sub>2</sub> had greater DMI/meal than those consuming 4.5% alfalfa, no CaMg(CO<sub>3</sub>)<sub>2</sub> and 9.0% alfalfa, 1.0% CaMg(CO<sub>3</sub>)<sub>2</sub>. Steers consuming 13.5% alfalfa, 1.0% CaMg(CO<sub>3</sub>)<sub>2</sub> and 9.0% alfalfa, no CaMg(CO<sub>3</sub>)<sub>2</sub> spent more time eating/meal ( $P < 0.05$ ) than steers consuming 4.5% alfalfa, no CaMg(CO<sub>3</sub>)<sub>2</sub>. No other differences ( $P > 0.10$ ) were observed in DMI, ruminal metabolism, or nutrient digestibility due to CaMg(CO<sub>3</sub>)<sub>2</sub> or an alfalfa x CaMg(CO<sub>3</sub>)<sub>2</sub> interaction. Ruminal pH increased linearly ( $P < 0.05$ ) with alfalfa level, averaging 5.41, 5.52, and 5.58 for steers fed 4.5, 9.0, and 13.5% alfalfa, respectively. Total tract OM digestibility decreased linearly ( $P < 0.05$ ) with increasing alfalfa level, measuring 85.7, 84.9, and 81.8% for steers fed 4.5, 9.0, and 13.5% alfalfa, respectively. A quadratic response ( $P < 0.05$ ) due to alfalfa level occurred for propionate and acetate:propionate ratio (A:P), with propionate measuring 34.3, 40.3, and 30.3 mM and A:P measuring 1.38, 1.23, and 1.72 for steers fed 4.5, 9.0, and 13.5% alfalfa, respectively. Based on Ca and Mg digestibilities, CaMg(CO<sub>3</sub>)<sub>2</sub> is similar to limestone and MgO in terms of availability, but did not produce ruminal pH responses similar to those observed by increasing alfalfa in high-concentrate diets.

**Key Words:** Acidosis, Feedlot steers, Ruminal buffer

**496 Effect of method of adding a fibrolytic enzyme to a dairy cow diet on ruminal fermentation and TMR degradation.** D. B. Dean\*<sup>1,2</sup>, A. T. Adesogan<sup>1</sup>, C. R. Staples<sup>1</sup>, S. C. Kim<sup>1</sup>, and R. Littell<sup>1</sup>, <sup>1</sup>University of Florida, Gainesville, <sup>2</sup>Universidad del Zulia, Maracaibo, ZU, Venezuela.

This study evaluated the effect of applying a fibrolytic enzyme (Promote<sup>®</sup>) to different portions of a bermudagrass-based diet on ruminal fluid pH, VFA and ammonia-N concentrations and on in situ TMR degradation (ISD) in dairy cows. Dietary treatments were: no enzyme (Control), or enzyme applied (4 g/cow/d) at feeding to the concentrate (EC), TMR (ETMR), or forage (EF), and enzyme applied (1.3 g/kg of DM) at ensiling (TS) to bermudagrass. Diets contained Tifton 85 bermudagrass (*Cynodon* spp.) silage, corn silage and a concentrate mixed at 35, 10 and 55% of dietary DM, respectively. Five ruminally-fistulated cows were fed for three consecutive 15-d periods, each consisting of 12 d of diet adaptation, 2 days of ISD measurements, and 1 d of ruminal fluid collection at 0, 2, 4, 6, 8 and 10

h after feeding. During periods 2 and 3, ISD kinetics were measured by incubating TMR samples in nylon bags for 0, 2, 4, 6, 8, 24, and 48 h and fitting an exponential model to the data. Mean ruminal pH was lower in cows fed EC (pH=6.02) than cows fed the Control diet (pH=6.29). Cows fed ETMR had lower ruminal NH<sub>3</sub>-N and total VFA concentrations, lower acetic acid concentration and a lower acetate:propionate ratio than cows fed the Control diet. Cows fed EF also had lower (P<0.01) acetic acid concentration and higher (P<0.01) isovaleric concentration acid than those fed the control diet. Cows fed TS had higher (P<0.05) isovaleric acid than those fed the Control diet. Kinetics of ISD were unaffected by enzyme addition, except that the TS TMR DM tended to be degraded at a faster (P=0.107) rate than the Control diet. Enzyme addition to the TMR was the most effective method of improving ruminal fermentation

**Key Words:** Fibrolytic enzymes, Dairy cows, Rumens fermentation

#### **497 Effects of feeding oxidized fat supplemented with antioxidant AGRADO on rumen nutrient digestibility and protein synthesis.**

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A dual effluent continuous culture system was used to investigate in a 2 x 2 factorial design the effect of feeding fresh (FF) or oxidized

fat (OF) when supplemented with or without antioxidant on nutrient digestibility and bacterial protein synthesis. Twelve fermenters were fed twice a day for 9 days a diet that consisted of 48 % grain mixture and 52 % forage that contained 3% (DM basis) FF or OF blend of unsaturated fats (33% fish, 33 % corn, and 26 % soybean oil and 7 % inedible tallow) and supplemented with 0 or 100 ppm of AGRADO<sup>®</sup>. OF contained higher concentration of peroxides (215 vs 3.5 meq/kg), and lower EPA (C20:5; 3.6 vs 5.1%) and DHA (C22:6; 1.7 vs 2.4 %) than FF. Feeding OF reduced CP digestibility (P = 0.009), microbial N yield (P < 0.03) and efficiency (P =0.03) when compared to FF. Adding Agrado improved total CHO (P=0.05), NDF (P=0.02) and ADF (P=0.04) digestibility, non-ammonia-N (P =0.008), microbial N yield (P=0.08), and the amount of digested feed N converted to microbial N (P=0.01), and reduced ruminal ammonia-N (P=0.008). From this study it can be concluded that feeding OF reduced CP digestibility and microbial protein synthesis and efficiency. The negative effect of feeding OF on rumen fermentation was corrected in the presence of Agrado. In addition, Agrado improved fiber digestibility in the presence of both types of fat.

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**Key Words:** Oxidized fat, Antioxidants, AGRADO

## **Teaching/Undergraduate and Graduate Education**

#### **498 Comparing vocational agriculture and non-vocational agriculture student success on high stakes testing.** D. Ritenour\* and D. Nash, *Ferrum College, Ferrum, Virginia.*

The purpose of this study was to determine if participation in vocational education courses has an effect on high-stakes test scores, namely the Virginia Standards of Learning (SOL) exams. Previous research indicates that students with vocational concentrations did not score significantly higher or lower on high-stakes exams. For this study, data for 400 students from a semi-rural high school in southwestern Virginia was collected. Variables included gender, SOL scores, agriculture course participation (ACP) and cumulative grade point average (GPA). The ACP data represents the number of agriculture courses, including animal science, the student participated in grades 9-12. The SOL scores are on a scale of 400 to 600 and include the content areas of algebra, geometry, world and US history, earth science, biology, chemistry, reading, and writing. Students are required to take SOL tests until they achieve a passing score of 400. However, no more than five students were required to re-test for any SOL test. The GPA data was given on a 4.0 scale, though 69 cases exhibited a GPA above 4.0 due to weighted credits on advanced placement courses. The SOL scores were regressed on GPA, gender, and ACP. Standardized coefficients for ACP for math (average of algebra and geometry), history (average of world and US history), science (average of earth science, biology and chemistry), reading, and writing scores were .231, .205, .263, .225 and .217, respectfully, and were each significant at the .0001 level. The positive coefficients indicate that as students' participation in agriculture courses increases their SOL scores increase as well. These results show the positive effect agriculture course participation appears

to have on students' success on high-stakes testing, but also makes a valid argument for the continuation of vocational agricultural education in the public school system. On the undergraduate and graduate level this research should be important to teacher educators due to the fact that the role of the agricultural educator, as well as educators in general, is swiftly changing to meet the needs of high-stakes testing.

**Key Words:** Vocational, High-stakes, Education

#### **499 Collegiate LifeKnowledge: A student-centered leadership development program.** C. M. Wood\*, *Virginia Polytechnic Institute and State University, Blacksburg.*

Collegiate LifeKnowledge (CLK), a leadership curriculum developed for members of collegiate agricultural student organizations, is an initiative of the National FFA Organization in partnership with the John Deere Corporation and CSREES. Collegiate LifeKnowledge was initiated and developed by stakeholder groups that included students, educators, student organization representatives, and industry representatives. A taskforce met in January 2005 to identify the direction of the project, followed by a writers' meeting in February. Lessons that focused on four facets of leadership—Personal, Organizational, Career, and Community—were completed in June. The lessons were developed to be taught by college students to college students—no lectures allowed. Each lesson can be taught in 15, 30 or 45 minutes and is divided into four parts. Part 1 gives the student leader all the logistical information related to the lesson. Part 2 has all the background information necessary to become an instant expert for