

Ruminant Nutrition: Feed Additives

W361 Effects of different feed additives on performance and carcass traits of feedlot cattle. C. A. Zotti, S. da Luz e Silva,* L. S. Martello, R. L. Meirelles, A. P. dos Santos Silva, P. L. Alvarez, P. H. Cancian, A. C. Ianni, L. E. Zanoni, and P. R. Leme, *Faculdade de Zootecnia e Engenharia de Alimentos, Universidade de São Paulo, Pirassununga, SP, Brazil.*

Feed additives are largely used in beef cattle diets to improve performance and reduce risks of nutritional disorder diseases like acidosis and timpanism. However, some countries tend to ban the use of ionophore in animal nutrition due to the risk of resistance of some microorganisms. Therefore, alternatives to non-antibiotic additives must be studied. The objective of this work was to investigate the effects of different feed additives on performance and carcass traits of beef cattle. Twenty 2 Nellore bulls (initial BW 353 ± 20.9 kg) and 22 steers (initial BW 320 ± 1.5 kg; 21mo old) were fed high concentrate diet (85% of concentrate and 15% of sugarcane bagasse as roughage source). The animals were allotted in 4 pens equipped with calan gate system and randomly assigned within sex (block) to one of the 4 treatments: Monensin at 30 mg/kg DM (MON); Virginiamycin at 30 mg/kg DM (VIR); combination to MON (15 mg/kg DM) plus VIR (15 mg/kg DM) (MPV); Blend of Castor Oil Acid and Cashew Oil fed at 500 mg/kg DM (COC). Feed and orts were computed daily for each animal for DMI and feed efficiency calculations. Animals were weighed (BW) and ultrasound scanned at the first day of the feedlot and each 28 d. Ultrasound measurements taken were Longissimus muscle area (ULMA) and backfat thickness (UBFT) between 12th and 13th ribs. After 112 d on feed, cattle were slaughtered and hot carcass weight (HCW), kidney, pelvic and inguinal fat (KPIF) and dressing percentage (DP) calculated. Dry matter intake for treatment MON was smaller (9.7 kg/day; $P = 0.04$) when compared with COC (11.1 kg/day) with no differences of VIRG and MPV (10.1 and 10.4 kg/day, respectively). There was no effect of treatments on ADG (1.60 ± 0.26 kg), feed efficiency (0.156 ± 0.02 kg ADG/kg DMI), final BW (510 ± 47 kg), HCW (307 ± 27.2 kg), dressing percentage ($60.0 \pm 1.78\%$), UBFT (5.9 ± 2.1 mm) and KPIF ($3.12 \pm 1.62\%$) at slaughter. Longissimus muscle area was higher for MPV (80.9 cm²; $P = 0.008$) than COC (74.4 cm²) and VIR (74.4 cm²). The additives studied can replace ionophores to beef cattle fed high concentrate diet, without affecting animal performance and most of carcass traits.

Key Words: additives, feedlot, Nellore

W362 Effect of *Propionibacterium freudenreichii* supplementation in diets containing canola or flaxseed oils on in vitro methanogenesis and lipid biohydrogenation. S. Ding^{1,2}, S. J. Meale^{*1,2}, M. L. He², J. Long³, A. Y. Alazeh², T. A. McAllister², and A. V. Chaves¹, ¹Faculty of Veterinary Science, University of Sydney, Sydney, NSW, Australia, ²Lethbridge Research Center, Agriculture and Agri-Food Canada, Lethbridge, Alberta, Canada, ³Department of Animal Science, Northeast Agricultural University, Haerbin, Heilongjiang, China.

The addition of propionibacterium as a direct fed microbial may provide a competitive pathway to methanogenesis for H₂ disposal in ruminants. Additionally, it has the potential to inhibit the biohydrogenation of polyunsaturated fatty acids (PUFA). Flaxseed and canola oils have been shown to reduce methane formation and rumen biohydrogenation due to the dominant levels of linolenic acid (C18:3) and linoleic acid (C18:2), respectively. However, to-date, no studies have been undertaken regarding the ability of propionibacterium to mitigate methane from

diets supplemented with plant oils or its effects on PUFA biohydrogenation. Thus, a 48 h, in vitro batch culture was conducted to determine the effects of *Propionibacterium freudenreichii* (PFB) on (1) in vitro VFA production and methane and (2) fatty acid biohydrogenation, in a backgrounding diet supplemented with 2 plant oils. Treatments were: backgrounding diet (CON); CON + PFB (CPB) (4×10^9 cfu propionibacteria/40 mL inoculum); CON + canola oil (6% DM; CCO); CCO + PFB (CCPB), CON + flaxseed oil (6% DM; FLO) and FLO + PFB (CFPB). Methane and gas production was measured at 3, 6, 12, 24, 48 h, whereas, total ruminal VFA were determined at 0, 6, 24 and 48 h of incubation. In vitro incubations were repeated twice with 3 replicates per treatment in each run. Data were analyzed using the MIXED procedure of SAS with treatments as fixed effect, and incubation run and interaction incubation run \times treatments as random effects. Cumulative gas production (mL), CH₄ production (mL/g DM) and total ruminal VFA concentration (mM) were not affected by treatment ($P \geq 0.10$). However, there was a tendency ($P = 0.07$) for both oil-based diets, with or without PFB, to reduce CH₄ production (mL/g DM) at 24 and 48 h. Additionally, oil increased ($P \leq 0.002$) the percentage of CLA+VA at 24 and 48 h. Propionibacterium did not affect ($P > 0.05$) biohydrogenation. However, flaxseed and canola oil increased ($P < 0.001$) the proportion of unsaturated fatty acids and decreased ($P < 0.001$) the proportion of saturated fatty acids in the total incubation period, thus inhibiting biohydrogenation based on oil diets. Canola oil based treatments had the highest ($P \leq 0.003$) proportion of C18:1 (cis-9) across all time points, whereas flaxseed oil groups had the greatest ($P \leq 0.04$) percentages of PUFA including C18:3, C18:1 (cis-15), biohydrogenation intermediates from C18:3 at 6, 24, 48h, and CLA (cis-9,trans-11) at 6, 24h, indicating the capability of flaxseed oil to alter biohydrogenation in favor of desirable PUFA production. In conclusion, propionibacterium did not significantly affect methanogenesis and lipid biohydrogenation based on either flaxseed or canola oil diets on current in vitro study.

Key Words: biohydrogenation, methane, lipids

W363 Efficacy of an autolysed yeast product (Levabon Rumen) for ruminants versus live yeast and yeast culture in vitro. A. Ganner^{*1}, C. Stoiber¹, I. Dohnal¹, K. Deckardt², F. Klevenhusen², G. Schatzmayr¹, and Q. Zebeli², ¹BiomIn Research Center, Tulln, Lower Austria, Austria, ²University of Veterinary Medicine, Vienna, Austria.

Various yeast products for ruminants have been reported to improve rumen microbial activity, feed utilization and cow performance. Increasingly, literature reports about the benefits of live yeast and yeast culture. However, little has been reported about the effect of autolysed yeasts for ruminants. Moreover, the mode of action of different yeasts, with respect to improving animal performance, is still not completely understood. Target of the present study was to evaluate the efficacy of a novel yeast derivative (Levabon Rumen) on anaerobic bacterial count (batch culture); on crude fiber, dry matter and crude protein digestibility (RUSITEC) versus yeast culture and live yeast, which are used for animal feeding. In a first in vitro batch culture study, rumen simulation reactors were filled with rumen fluid, mixed with saliva and incubated under anaerobic conditions for 24 h. The yeasts (in mg yeast/flask: Levabon Rumen 40–300; yeast culture1 300, 600; yeast culture2 200, 400; live yeast 0.5, 5) were mixed into the ration (50% hay and 50% concentrate). Total viable anaerobic bacteria (batch culture) were counted (cfu/g). In a second experiment with the rumen simulation technique (RUSITEC), different levels of the yeast products (in mg yeast/12 g dry matter: Levabon Rumen 3, 6, 12, 18, 24;

yeast culture¹ 12, 36; yeast culture² 6, 12; live yeast¹ 2.4; live yeast² 0.3) were fed daily for 14 d. The samples were mixed into a ration consisting of 50% hay and 50% concentrate. The parameters measured include nutrient digestibility (Weender analysis), pH, redox potential and volatile fatty acids. Results indicate improved digestibility in RUSITEC and increased amount of anaerobic bacteria in batch culture by the autolysed yeast as well as by the probiotic yeast and the yeast culture. Volatile fatty acids also increased. None of the yeast showed an effect on either pH or redox potential, perhaps due to continuously buffering the system and strictly anaerobic conditions. Based on these findings, Levabon Rumen represents a promising feed additive to improve cattle health and performance.

Key Words: autolysed yeast, probiotic, ruminants

W364 Effect of monensin on methane emissions in dairy cattle can be explained by level of dry matter intake and fat content of the diet. J. A. D. R. N. Appuhamy^{*1}, A. B. Strathe¹, S. Jayasundara², C. Wagner-Riddle², J. Dijkstra³, J. France², and E. Kebreab¹, ¹University of California, Davis, ²University of Guelph, Guelph, ON, Canada, ³Wageningen University, Wageningen, the Netherlands.

Methane (CH₄) is an important greenhouse gas that contributes to climate change. Cattle produce the majority of CH₄ emissions from ruminants accounting for about 30% of the global emissions. Monensin improves feed conversion efficiency and increases propionic acid molar proportion in the rumen. Several studies have investigated the effects of monensin on CH₄ emission from dairy cows and the results vary widely. The objectives of the present study were to conduct a meta-analysis to infer about the overall effect of monensin feeding on CH₄ emission from dairy cows and to quantify factors explaining between-study variability of the monensin effect. Data of mean and SD of CH₄ emission (g/d) in monensin and control groups, sample size (N), treatment duration (T, d), DMI (kg/d), monensin dose (mg/kg of DMI), and basal diet nutrient contents (g/kg of DM) from 11 controlled-trials were included. The Metafor package in R was used to conduct the meta-analysis. Effect size (ES, g/d) was calculated as difference between the means of monensin feeding and control groups. A model accounting for random study effects revealed significant ES heterogeneity between trials (98%, $P < 0.001$) suggesting T, DMI, monensin dose and nutrient contents as potential covariates for explaining between-study variability. The final model including potentially significant covariates were selected using Likelihood-ratio tests. Feeding monensin was shown to have a non-significant effect ($ES = -2.88 \pm 3.28$ g/d, $P = 0.380$) on CH₄ emission from dairy cows. Effect size was positively (covariate coefficient = 1.35 ± 0.56 , $P = 0.016$) and negatively (covariate coefficient = -5.15 ± 1.51 , $P = 0.001$) associated with DMI and crude fat content, respectively. Monensin dose and T did not significantly affect ES. Inclusion of DMI and fat content into the model reduced total heterogeneity of ES (τ^2) by about 70% ($\tau^2 = 322$ vs. 100). Dry matter intake and crude fat content were significant and could explain between-study variability of the monensin effect on CH₄ emission from dairy cows.

Key Words: dairy cows, methane, monensin

W365 Effects of dietary supplementation of a yeast product on performance and morbidity of newly received beef heifers. C. H. Ponce^{*1}, J. S. Schutz¹, C. Elrod², U. Y. Anele¹, and M. L. Galyean¹, ¹Department of Animal and Food Sciences, Texas Tech University, Lubbock, ²Varied Industries Corp. Inc., Mason City, IA.

Two truckloads of beef heifers (n = 237; average initial BW = 191 kg \pm 1.17) were used to evaluate the effects of a yeast culture/enzymatically

hydrolyzed yeast product (Celmanax; Varied Industries Corp. Inc., Mason City, IA) included in a 65% concentrate receiving diet on performance and percentage of bovine respiratory disease (BRD) morbidity during a 35-d receiving period. Treatments included: 1) the 65% concentrate receiving diet with the addition of Celmanax; 14 g/heifer/d; mixed in a carrier; or 2) the 65% concentrate receiving diet without Celmanax (Control; 14 g/heifer/d of carrier only). No load \times treatment interactions ($P > 0.157$) were detected for any measures of performance or BRD morbidity. Initial BW did not differ ($P > 0.537$) between treatments nor did d-35 BW ($P = 0.332$). Average daily gain for d 0 to 14 ($P = 0.052$) and for the overall 35-d ($P = 0.078$) period tended to be greater for Celmanax heifers than for controls. Intake of receiving diet DM was greater ($P \leq 0.025$) by the Celmanax heifers than by Control heifers at all the measurement periods during the experiment, but G:F did not differ ($P > 0.175$) between treatments. The proportion of heifers treated once for BRD tended ($P = 0.094$) to be less with the Celmanax treatment (4.1 for Control vs. 10.5% for Celmanax). Results indicate that heifers fed Celmanax ate more receiving diet DM and tended to gain more than Control heifers, with a tendency for decreased BRD morbidity during a 35-d receiving period.

Key Words: newly received beef cattle, feedlot performance, yeast product

W366 Effect of oregano, ginger and thyme oils on in vitro rumen fermentation and methane emission. F. G. Vilela,^{*} I. C. S. Bueno, A. S. Netto, T. S. Canaes, J. E. Freitas Jr., B. C. Venturelli, and F. P. Rennó, University of São Paulo, Pirassununga, SP, Brazil.

In vitro semi automated gas production technique has been used to evaluate the effects of essential oils at different levels (0, 15, 30, 45 and 60 μ /ml) on ruminal fermentation and methane production. The investigated essential oils were *Origanum vulgare* (OV₁₅, OV₃₀, OV₄₅, OV₆₀), *Zingiber officinale* (ZO₁₅, ZO₃₀, ZO₄₅, ZO₆₀) and *Thymus vulgaris* (TV₁₅, TV₃₀, TV₄₅, TV₆₀). The analysis of the essential oils by gas chromatography - mass spectrometry demonstrated that the principal components were carvacrol (70%) and thymol (11%) in OV, zingiberene (33%) and zingerone (17%) in ZO, thymol (40%) and carvacrol (13%) in TV. A total mixed ration (50% roughage: 50% concentrate) was incubated with buffered rumen fluid. Data were analyzed using PROC MIXED of SAS 9.1. The inclusion of OV, TV, ZO₁₅ and ZO₃₀ did not affect significantly on true digestibility of dry matter and organic matter, while ZO₄₅ and ZO₆₀ decreased significantly ($P \leq 0.05$). The efficiency of microbial protein was estimated by partitioning factor (PF, estimated as ratio of truly organic matter degradation and gas volume produced in 24 h of incubation). There was no significant effect of investigated essential oils on PF except OV₄₅, TV₄₅ and TV₆₀ which improved significantly ($P \leq 0.05$) the PF. There was no significant effect of essential oils on the production methane except OV₄₅ and TV₆₀ which decreased significantly ($P \leq 0.05$) the methane production. The NH₃-N concentration was not altered with inclusion of essential oils at different doses. The short chain fatty acids concentration was not altered with inclusion of essential oils except OV₃₀, TV₄₅ and TV₆₀ which increased significantly ($P \leq 0.05$) the propionate concentration. This study suggested that essential oils have the potential to affect rumen fermentation and OV₄₅ and TV₆₀ could be a promising methane mitigating agent.

Key Words: essential oils, gas production, rumen fermentation

W367 Effect of some essential oils on rumen fermentation and methane emission in vitro. F. G. Vilela,* I. C. S. Bueno, A. S. Netto, J. E. Freitas Junior, B. C. Venturelli, T. S. Canaes, and F. P. Rennó, *University of São Paulo, Pirassununga, SP, Brazil.*

The objective of this study was to evaluate in vitro the effects of essential oils at different levels (0, 15, 30, 45 and 60 µL/100 mL buffered rumen fluid) on ruminal fermentation by semi automatic gas production technique. The selected essential oils were *Rosmarinus officinalis* (RO₁₅, RO₃₀, RO₄₅, RO₆₀), *Syzygium aromaticum* (SA₁₅, SA₃₀, SA₄₅, SA₆₀) and *Pimpinella anisum* (PA₁₅, PA₃₀, PA₄₅, PA₆₀). The analysis of the essential oils by gas chromatography-mass spectrometry demonstrated that the principal components were 1.8 cineol (28%) and p-cimeno (13%) in RO, eugenol (40%) in SA and anethol (45%) in PA. The trial was conducted in duplicates making a total of 12 bottles for each treatment. The substrate used was the basal diet consisting of corn silage and concentrate (71.66% corn, 12.12% soybean meal and 10% soybean), in ratio 50:50. Ground samples (1 g) were incubated in 100 mL of diluted rumen fluid (10 mL mixed rumen fluid + 90 mL of buffer solution mineralized) in 160 mL serum bottles. Data were analyzed using PROC MIXED of SAS 9.1. There was no significant effect of essential oils on the production methane except PA₄₅ and PA₆₀ which decreased significantly ($P \leq 0.05$) the methane production. The inclusion of RO₄₅, RO₆₀, SA₁₅, SA₃₀ and PA did not affect on true digestibility of dry matter and organic matter, while RO₁₅, RO₃₀, SA₄₅ and SA₆₀ decreased significantly. The efficiency of microbial protein was estimated by partitioning factor (PF, estimated as ratio of truly organic matter degradation and gas volume produced in 24 h of incubation). There was no effect of investigated essential oils on PF except SA₄₅ and SA₆₀ which improved significantly the PF. The short chain fatty acids concentration was not altered with inclusion of essential oils except PA₃₀, PA₄₅ which increased significantly the propionate concentration. This study suggested that essential oils could modify the rumen fermentation and have a potential in methane mitigation, which may be beneficial for animal growth.

Key Words: essential oils, methane, degradation

W368 Effect of abomasal inorganic phosphorus infusion on phosphorus absorption in lactating dairy cows. X. Feng,* J. P. Jarrett, P. P. Ray, L. Karpinski, B. F. Willing, and K. F. Knowlton, *Virginia Polytechnic Institute and State University, Blacksburg.*

The objective of the study was to evaluate the effect of inorganic P (Pi) supply on P disappearance from the intestinal tract of lactating dairy cows. Four ruminally and ileally cannulated crossbred cows were used in a 4 × 4 Latin square with 21-d periods. Cows were fed a total mixed ration containing 0.21% P, providing 50% of the cows' P requirement. Ytterbium (Yb) labeled corn silage and Co-EDTA were used as particulate and liquid phase markers, respectively, to measure digesta flow. Omasal samples were collected every 9 h from d 10 to 13 to measure omasal P flow from the basal diet. On d 13 to 21 of each period, each cow was infused daily with 0, 20.1, 40.2, or 60.3 g Pi into the abomasum and total collection was conducted on d 18 to 21. Ileal samples were collected every 9 h on d 18 to d 21. Feed, digesta, and fecal samples were analyzed for total P and Pi using the molybdovanadate yellow method and blue method, respectively. All data were analyzed using PROC GLIMMIX in SAS 9.2 using contrasts to evaluate linear and quadratic effects of Pi infusion dose. Dry matter (DM) intake, apparent DM digestibility, milk yield and milk total P were unaffected by Pi infusion ($P > 0.05$). Ileal flow and fecal excretion of total P and Pi increased linearly ($P < 0.05$) with increasing infused Pi. In the small intestine, net disappearance of total P and absorption of Pi decreased ($P < 0.05$; 67.7 to 37.4% and 83.2 to 52.8%, respectively) with increased Pi infusion. Similarly,

total tract disappearance of total P and Pi decreased with increasing Pi infusion ($P < 0.05$; 74.3 to 54.3% and 81.1 to 66.6%, respectively). In the large intestine, net Pi absorption was unaffected but increasing Pi infusion increased disappearance of total P ($P < 0.05$). Results of this study suggest limits to Pi absorption in the small intestine.

Key Words: dairy cows, phosphorus absorption, phosphorus excretion

W369 Effects of essential oils on in vitro ruminal fermentation and methane production of a mixed diet. I. Mateos¹, A. Díez¹, C. Saro¹, D. Yáñez-Ruiz³, M. D. Carro^{1,2}, and M. J. Ranilla^{*1,2}, ¹Dpto. Producción Animal, Universidad de León, Campus de Vegazana, León, Spain, ²Instituto de Ganadería de montaña (CSIC-ULE), Finca Marzanas, Grulleros, León, Spain, ³Estación Experimental del Zaidín (CSIC), Camino del Jueves, Armilla, Granada, Spain.

The effects of 6 doses (0 (control), 40, 80, 160, 320 and 640 mg/L incubation medium) of carvacrol (CAR), thymol (TYM), eugenol (EUG) and garlic oil (GO) on in vitro fermentation of a mixed diet (65:35 alfalfa hay:concentrate) were evaluated in batch cultures of mixed rumen microorganisms from the rumen of sheep fed the same diet. Previous in vitro studies have shown the ability of these compounds to modify ruminal fermentation, but the effects could be dose and diet dependent. After 24 h of incubation, the main fermentation variables were determined. Differences among treatments were declared at $P < 0.05$. Dose 640 mg/L of all additives tested inhibited almost completely rumen microbial fermentation, confirming their antimicrobial activity at high concentrations, and therefore the values were withdrawn from the statistical analysis. GO and EUG did not modify ($P > 0.05$) rumen fermentation at any other dose. TYM at 320 mg/L decreased ($P < 0.05$) total VFA, acetate, propionate and methane production, and increased ($P < 0.05$) acetate:propionate ratio compared with control. CAR has similar effects on VFA and propionate production at dose 160 mg/L, and tended ($P = 0.08$) to reduce methane production. At 320 mg/L, CAR also increased ($P < 0.05$) acetate:propionate ratio and decreased ($P < 0.05$) ammonia-N concentrations, compared with the control. These results confirm that essential oils supplementation shows different effects on ruminal fermentation depending on the administered dose and the nature of the compound. Under the conditions of this study the doses of essential oil that modified VFA profile and reduced methane production also resulted in a reduction in VFA production, which would indicate a decrease in feed digestibility and have negative consequences on animal productivity.

Key Words: essential oils, in vitro ruminal fermentation, methane

W370 Effect of feeding *Bacillus subtilis* and *Bacillus licheniformis* on dry matter and nutrient intake and digestibility by lambs fed a low quality roughage diet. E. Martínez-Loarte,* A. A. Rodríguez, and L. C. Solórzano, *University of Puerto Rico, Mayaguez, PR.*

The objective of this experiment was to evaluate the addition of *Bacillus subtilis* and *Bacillus licheniformis* (BSL) to the diet on DM, CP, and NDF intake and digestibility in growing lambs fed a diet consisting of 50% commercial tropical grass hay (mixtures of *Panicum maximum*, *Sorghum halapense*, and *Digitaria decumbens*, 71.7% NDF, 4.9% CP;) and 50% hay of the invasive weed *Hyparrhenia rufa* (78.8% NDF, 3.5% CP). Ten crossbred lambs (11.2 kg) were randomly assigned to one of 2 dietary treatments, with or without daily addition of BSL in the diet. The daily offering of hay was at the rate of 4% of body weight on a dry basis and water was available ad libitum. The bacterial strains were mixed with calcium carbonate carrier, and were included in the additive to provide

1.33×10^9 cfu/g supplying 1.33×10^8 cfu/head/d. The additive was mixed with 225 g of a commercial concentrate (15.67% CP) and fed daily to the lambs during a 49 d period. Nutrient intake and digestibility (DM, CP, and NDF) were determined from d 42 to 49. Data was analyzed using the PROC MIXED procedure of SAS using the initial lamb body weight as covariance and Tukey's test was used for mean separation. Total DM intake and tropical grass hay intake was higher ($P < 0.05$) in lambs consuming the additive containing BSL than control animals (484.26 vs 445.23 g/d and 236 vs 218 g/d, respectively), but intake of *Hyparrhenia rufa* hay was similar (32.18 vs 52.43 g/d). Total forage intake as percentage of total DM intake, and DM intake as percentage of lamb body weight was also higher ($P < 0.05$) in lambs consuming BSL than control (59.08 vs 55.09% and 2.36 vs 2.03%, respectively). Feeding the *Bacillus* strains to lambs also improved NDF digestibility (62.48 vs 58.71%), however, the experimental treatment did not affect DM and CP digestibility. In summary, addition of BSL improved total intake by lambs fed a diet containing commercial concentrate and low quality roughage as well as the individual tropical grass hay component. Addition of BSL to the diet also improved cell-wall digestibility of low quality forage by lambs.

Key Words: *Bacillus* spp., Tropical grass hay, *Hyparrhenia rufa*

W371 Efficacy of live yeast *Saccharomyces cerevisiae* (strain Sc 47) and/or yeast cell wall on rumen fermentation and digestive utilization of corn silage-based diet in mid-lactating dairy cows. C. Bayourthe^{*1,2}, C. Julien^{1,2}, E. Auclair³, and J. P. Marden³, ¹INRA, UMR1289 Tissus Animaux Nutrition Digestion Ecosystème et Métabolisme TANDEM, Castanet Tolosan Cedex, France, ²Université de Toulouse, INPT-ENSAT, INP-ENVT, UMR1289 TANDEM, Castanet Tolosan Cedex, France, ³Lesaffre Feed Additives, Marquette Lez Lille, France.

Four ruminally cannulated Holstein cows in mid-lactating (DIM = 185 d) were assigned in a 4×4 Latin square design. They were fed a TMR composed of 78.8% corn silage, 9.8% soybean meal, 10.9% ground corn and 0.5% minerals at 23 kg DM daily during 10 d either supplemented with 5 g of live yeast (LY) at 10^{10} cfu/g (L1), or 5 g LY + 5 g yeast cell wall (YCW; L2), or 5 g LY + 10 g YCW (L3), or not supplemented (CTRL). Ruminal fluid was withdrawn from donor cows and mixed with 100 mL artificial saliva (1:1) and 3 g TMR for 8 h-anaerobic batch incubations to determine pH, E_h , rH index, total and individual VFA, and ammonia. The same donor cows were given chromic oxide (10 g before each daily meal) as indigestible flow marker to assess the fecal excretion of the cows and thus DM, OM, NDF and ADF digestibility. In vitro results showed that pH, E_h and total VFA were significantly modified (respectively +0.12 pH unit, -24 mV and +0.97 mM/L/h on average when compared with CTRL) with L1, L2 and L3. Ruminal reducing power (rH) was further improved with L2. Acetate contents of L2 and L3 differed significantly with CTRL (in average +0.52 mM/L/h). A 36% mean decrease in rumen ammonia was observed with L3 in comparison to other treatments. Propionate and butyrate contents tended to increase ($P < 0.1$) for L2 and L1 respectively. In vivo NDF and ADF digestibility tended to increase (+8 and +9 points respectively) with L3 compared with other treatments, while DM, OM and starch digestibility showed no significant difference. Combining LY and YCW resulted in improving ruminal conditions followed by better fiber utilization in mid-lactating dairy cows.

Key Words: live yeast, mid lactation cow, yeast cell wall

W372 Effect of live yeast *Saccharomyces cerevisiae* (strain Sc 47) on nutrient digestion and ruminal fermentation in relation with rumen degradable protein content of the diet. C. Julien^{*1,2}, J. P. Marden³, E. Auclair³, R. Moncoulon^{1,2}, and C. Bayourthe^{1,2}, ¹INRA, UMR1289, Tissus Animaux Nutrition Digestion Ecosystème et Métabolisme (TANDEM), Castanet Tolosan Cedex, France, ²Université de Toulouse, INPT-ENSAT, INP-ENVT, UMR1289 TANDEM, Castanet Tolosan Cedex, France, ³Lesaffre Feed Additives, Marquette Lez Lille, France.

Four ruminally cannulated early lactating Holstein were involved in a cross over design. Cows were fed for a 33-d period a TMR containing an adequate level of RDP (HL) or was 30% below HL (LL), by using soybean meal or tanned soybean meal, respectively: for 21 d with no live yeast (LY) addition (14 d of adaptation and 7 d of sampling) followed by 12 d when LY (5 g at 10^{10} cfu/g /cow/d) was added to the diet (5 d adaptation and 7 d sampling). The pH and redox potential (E_h) were recorded over 3 consecutive days (d 15 to 17 and d 27 to 29) using the ex vivo method (Julien et al., 2010). Ruminal fluid samples were collected on the same days, before the morning meal then 1, 2, 4, 6 and 8 h post-feeding. Feces were collected individually and quantitatively over 48h (d 20 to d22 and d 32 to d 34) and individual DMI were measured for determining apparent nutrient digestibility. Nutrients daily intake did not differ between diets. Fecal excretion of OM was greater in LL (6.7 kg/d) compared with HL (5.6 kg/d). For LL diet, LY decreased ($P < 0.05$) fecal flows of OM (-19%), ADF (-17%), and N (-20%). Fecal N amount was lower ($P < 0.0001$) for HL than for LL: 128.5 vs. 232.9 g/d. Digestibility of CP was lower ($P < 0.0001$) for LL than for HL while that of ADF was higher ($P = 0.033$). With LY, digestibility of CP and ADF were increased ($P < 0.05$): respectively +0.8 and +4.8 points for HL, and +5.9 and +6.4 points for LL. Ruminal pH and E_h were lower ($P < 0.0001$) with HL compared with LL: 5.95 and -167 mV vs. 6.13 and -144 mV. Live yeast increased ruminal total VFA (+8.6%; $P < 0.05$), C2 (+10%; $P < 0.05$), and C4 (+35%; $P < 0.05$) contents for LL and decreased that of C3 (-9.8%; $P < 0.05$) for HL. Those results suggested a specific interest in using LY in RDP deficient diets for early lactating cows.

Key Words: digestion, live yeast, rumen degradable protein

W373 Effects of difructose anhydride III supplementation on serum calcium, dry matter intake and energy status in periparturient dairy cows. M. Teramura^{*1}, S. Wynn², M. Abe², S. Hisasue², T. Sato¹, M. Ohtani¹, C. Kawashima², and M. Hanada², ¹Nippon Beet Sugar Manufacturing Co. Ltd., Obihiro, Hokkaido, Japan, ²Obihiro University of Agriculture and Veterinary Medicine, Obihiro, Hokkaido, Japan.

Difructose anhydride (DFA) III is an indigestible oligosaccharide. It has been shown that DFA III promotes intestinal Ca absorption in rats and reaches the duodenum without being degraded by rumen bacteria when ingested by dairy cows. The purpose of this study was to investigate the effects of DFA III supplementation on serum Ca, dry matter intake (DMI) and energy status in periparturient dairy cows. Twenty-two multiparous Holstein cows were divided into DFA and control group, which were balanced for parity and BW. DFA group was fed 40 g/d of DFA III from 2 wk before expected calving to 1 wk postpartum (pp) and control group was received no DFA III. DMI (-2 to 3 wk pp) and milk yield (0 to 3 wk pp) were recorded daily. BW was measured weekly during the study period (-3 to 6 wk pp). Blood samples were taken weekly throughout the study period as well as on the day before calving and at 0, 6, 12, 24, 48 and 72 h pp for serum Ca, glucose, NEFA and BHBA. Rumen contractions were also counted at blood sampling (5 cows in each group). All data was compared by repeated measures ANOVA. Changes in serum Ca were compared by Tukey-Kramer method for each group. The incidence of subclinical ketosis (SCK; BHBA ≥ 1400 $\mu\text{mol/L}$) was

compared by Fisher's exact test. Pretreatment blood parameter levels were similar between the groups. Hypocalcemia (≤ 7.4 mg Ca/dL) were observed at pp period in 2 DFA and 4 control cows. Serum Ca recovered to the prepartum level in 12 h pp in DFA cows and 48 h pp in control cows. The number of rumen contractions and DMI were higher in DFA cows than control cows during the first 3 wk pp ($P < 0.05$). Milk yield and BW were not affected by the treatment. DFA cows had lower NEFA at 1 ($P < 0.05$) and 7 ($P < 0.10$) d pp with a lower incidence of SCK during the 6 wk pp (9% vs. 45%; $P = 0.07$). These results suggest that periparturient DFA III supplementation encourages DMI by assisting recovery from hypocalcemia thereby maintaining gastrointestinal motility. Increased DMI by supplement of DFA III might reduce negative energy balance and prevent SCK in early lactation.

Key Words: DFA III, DMI, hypocalcemia

W374 Effects of monensin and extracts of hops and *Yucca schidigera* applied alone or in combination on rumen fermentation in vitro. N. Narvaez, Y. Wang,* and T. A. McAllister, AAFC, Lethbridge, AB, Canada.

There is increasing interest in using plant bioactive as alternative to in-feed antibiotics due to the public concerns over the possible contribution to the emergence of antibiotic resistant bacteria. One possible strategy to increase the efficacy of in-feed antibiotics and possibly circumvent the development of antibiotic resistance is to feed them in combination with plant bioactives. The objective of this study was to assess the effects of monensin (MON), and plant bioactives alone and in combination with MON on in vitro ruminal fermentation. Ethanol extracts of hops (HE; α - and β -acids) and *Y. schidigera* (YE; steroidal saponin) were used as sources of plant bioactives. Treatments were Control (no additive), MON alone (2.5, 5 and 10 $\mu\text{g/ml}$) and HE (600 $\mu\text{g/ml}$) or YE (650 $\mu\text{g/ml}$) alone or combined with the first 2 concentrations of MON. Total gas and CH_4 production were measured at 4, 8, 12, 24 and 48 h of incubation, whereas VFA, $\text{NH}_3\text{-N}$ concentration, microbial protein (MP), true DM disappearance and ruminal bacteria were determined after 48h. All treatments decreased ($P < 0.05$) gas and CH_4 production, MP, and $\text{NH}_3\text{-N}$ accumulation. VFA production decreased ($P < 0.05$) with addition of MON alone (10 $\mu\text{g/ml}$) or in combination with YE. Molar proportion of butyrate were reduced ($P < 0.01$) and that of propionate increased ($P < 0.01$) by all treatments. MON reduced ($P < 0.001$) the relative proportions of 16S rRNA copies associated with *Ruminococcus flavefaciens*, but increased those of *Selenomonas ruminantium* ($P < 0.01$) and *Ruminobacter amylophilus* ($P < 0.05$). All treatments reduced ($P < 0.01$) the relative abundance of methanogens. HE alone or in combination with MON reduced ($P < 0.01$) proportions of *R. flavefaciens*, but increased ($P < 0.05$) those of *S. ruminantium* when combined with MON. YE alone and in combination with MON increased ($P < 0.01$) *R. flavefaciens* and *S. ruminantium*. HE and YE altered rumen microbes and fermentation in a manner similar to MON with many responses being additive when applied in combination.

Key Words: monensin, plant bioactives, rumen fermentation

W375 Concentrate level and combined use of ionophore and virginiamycin on feeding behavior of Nelore steers fed high grain diets. A. J. C. Nuñez¹, V. V. Almeida², J. P. Schoonmaker³, F. T. Mercado¹, F. Pinese¹, I. E. Borges¹, R. R. Casagrande¹, P. R. Leme¹, and J. C. M. Nogueira Filho¹, ¹USP/FZEA, Pirassumunga, SP, Brazil, ²USP/ESALQ, Piracicaba, SP, Brazil, ³Purdue University, West Lafayette, IN.

The objective in this study was to evaluate feeding behavior of Zebu cattle fed 2 concentrate levels and 2 virginiamycin (VM) levels in diets

containing salinomycin (SL). Eight ruminally cannulated Nelore steers (434 \pm 35 kg initial BW) were randomly allocated to a 4x4 replicated Latin square design (21-d periods). Two concentrate levels (70C and 90C diets had 70 and 90% concentrate on DM basis, respectively) and 2 VM levels (0 and 15 ppm) were tested in a 2x2 factorial arrangement of treatments. Animals were housed in individual pens and fed once daily at 0800 h. The forage source was sugarcane silage (72.4% NDF on DM basis), and all diets included the ionophore SL (13 ppm). During a 24-h period, eating and ruminating activities were visually monitored every 5 min and assumed to persist for the entire 5-min interval. Total chewing time (TCT; min/d) was calculated as the sum of total time spent eating (TTSE; min/d) and ruminating (TTSR; min/d). Rate of intake (RI; g/min) was determined by dividing DM intake in the evaluation day by TTSE, and rate of rumination (RR; min/kg) was calculated by dividing TTSR by DM intake. Statistical analyses were conducted using the GLM procedure of SAS. There was an interaction ($P = 0.02$) between concentrate and VM levels only for TTSE. Within the VM treated group, steers receiving 70C spent 71.9 min/d more eating compared with 90C (234.4 \pm 7.9 vs. 162.5 \pm 11.7 min/d, respectively; $P < 0.01$). Due to higher NDF content of the diet, TTSR was higher for the 70C compared with the 90C treatment (427.8 \pm 23.1 vs. 301.6 \pm 18.9 min/d, respectively; $P < 0.01$). As a consequence, TCT increased as concentrate level decreased (643.7 \pm 29.6 vs. 481.2 \pm 21.3 min/d for 70C and 90C, respectively; $P < 0.01$). Because of the higher NDF content of the diet, the 70C group also showed lower RI (37.9 \pm 1.7 vs. 49.7 \pm 3.8 g/min for 70C and 90C, respectively; $P < 0.01$) and higher RR (53.6 \pm 2.6 vs. 35.9 \pm 1.7 min/kg for 70C and 90C, respectively; $P < 0.01$). In summary, feeding behavior of Nelore steers is more influenced by concentrate level than by VM supplementation in diets containing SL.

Key Words: antibiotics, beef cattle, salinomycin

W376 The effect of different doses of exogenous enzymes preparation on in vitro gas production and ruminal fermentation activities of some fibrous feeds in cows. M. M. Y. Elghandour¹, C. G. Peñuelas-Rivas¹, M. Ronquillo¹, A. Z. M. Salem¹, H. Gado², and N. E. Odongo³, ¹Facultad de Medicina Veterinaria y Zootecnia, Universidad Autónoma del Estado de México, Toluca, Estado de Mexico, Mexico, ²Faculty of Agriculture, Ain Shams University, Cairo, Egypt, ³Animal Production and Health Section, Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture, International Atomic Energy Agency, Vienna, Austria.

This study was conducted to investigate effects of different doses of exogenous enzyme preparation (ENZ- 0, 0.06, 0.12 and 0.24 g/g DM) on in vitro gas production and some ruminal fermentation patterns such as truly degraded substrate (TDS), short chain fatty acids (SCFA), and microbial protein production (MP) of 4 fibrous feeds (FF - Sugarcane leaves, taiwan grass, sorghum straw and llanero grass) in cows. Rumen liquor was collected before the morning feeding from 2 cows (450 \pm 6 kg BW) fitted with permanent rumen cannula. In vitro gas production (GP) was recorded at 2, 4, 6, 8, 10, 12, 24, 48 and 72 h of incubation. After 72 h, the incubation was stopped and the inoculants pH was determined and filtered to determine TDS. Ruminal fermentation parameters such as 24 h partitioning factor (PF24), gas yield (GY24), in vitro organic matter digestibility (IVOMD), metabolizable energy (ME), SCFA, and MP were also estimated. Data were analyzed as 4 (4 doses of ENZ) x 4 (4 FF) factorial experimental design. Interaction between FF x ENZ ($P < 0.05$) only occurred for gas production at 12 h (GP12) and 24 h (GP24) of incubation, but there were no interactions after 24 h of incubation. Relative to control, addition of ENZ increased ($P < 0.05$) accumulated gas volume of all FF during the 72 h of incubation

compared with control (0 g ENZ/g DM). Among the FF species, gas production was higher ($P < 0.001$) in sorghum straw and lower ($P < 0.001$) in sugarcane leaves. There was no significant effect of ENZ on gas production parameters (i.e., b; asymptotic gas production, c; rate of gas production and L; discrete lag time before gas production) of all FF. Addition of ENZ doses increased ($P < 0.05$) gradually gas production and this increase was higher ($P < 0.05$) for 0.24 g ENZ/g DM. Addition of ENZ was increased ($P < 0.05$) some fermentation parameters (i.e., IVOMD, ME, GY24 and SCFA) were higher ($P < 0.05$) of all FF, while the highest ($P < 0.05$) increased were in sorghum straw with a lower values in sugarcane leaves. Results indicated that exogenous enzymes preparation probably improve the ruminal fermentation activities of fibrous feeds in cows. The authors wish to acknowledge the financial support from IAEA/FAO project MEX 16307.

Key Words: exogenous enzymes, fibrous feeds, gas production

W377 Nutrient intake and ruminal parameters in response to *Bacillus subtilis* included on beef steer diet. B. Vieira*, R. Telles, V. Naves, I. Carvalho, and R. Reis, *Universidade Estadual Paulista*.

The objective of this experiment was to evaluate the inclusion of *Bacillus subtilis* on the diet of 4 Nellore steers, with rumen cannulas and allocated on a Latin square design, with inclusion of Biotop (a probiotic product containing 10^9 colony-forming unit (cfu) *Bacillus subtilis*/g, as main plots and sampling time as subplots. The period last 20 d each and it was evaluate the dry matter (DM), organic matter (OM), crude protein (CP), ether extract (EE), neutral detergent fiber (NDF), non fiber carbohydrates (NFC) total carbohydrates (TC) and total digestible nutrients (TDN) intakes and, was measured the ruminal pH and N-ammonia. Treatments were defined as control (no addition of) and 3 doses (1, 2 and 4g.day⁻¹). The sampling times for pH and ammonia were at the time (0), at 3, 6, 9, 12 and 24 h after the morning the diet. The forage utilized was corn silage, in a 40:60 forage and concentrate ratio. The DM, OM, CP, EE, NDF and TC intakes were not different (Table 1). However, TDN intake increased with the addition of the probiotic. The pH and the N-ammonia were not affected by treatment, but a quadratic equation was adjusted as a function of sampling time, with a minimum point at 6.2 and 6.2 h after feeding for pH and N-ammonia with values of 5.88 and 24.17mg.dL⁻¹, respectively. It can be concluded that the addition of Biotop on beef cattle diet on the doses between 1 and 4g.day⁻¹ do not affect ruminal pH and N-ammonia but can improve the TDN intake.

Table 1. Nutrient intake (%BW) of animals fed with *Bacillus subtilis* (Bs) in the diet

Item	C	Bs			CV (%)
		1g	2g	4g	
DM	1.91	1.99	1.99	1.86	8.76
OM	1.80	1.87	1.88	1.79	9.47
CP	0.24	0.25	0.25	0.23	8.88
EE	0.05	0.05	0.05	0.05	6.95
NDF	0.54	0.56	0.56	0.54	9.54
NFC	0.98	1.02	1.01	0.96	8.95
TC	1.52	1.57	1.58	1.50	9.25
TDN	1.36 ^b	1.41 ^a	1.42 ^a	1.40 ^a	7.32

^{a,b}Means followed by different letter in row differ at 5% probability by Tukey test.

Key Words: ammonia, pH, probiotic

W378 Effect of exogenous fibrolytic enzymes on *in vitro* rumen fermentation of corn silage. X. Chen, J. K. Wang, H. L. Mao, C. H. Wu, Y. M. Wu, and J. X. Liu,* *Institute of Dairy Science, MOE Key Laboratory of Molecular Animal Nutrition, Zhejiang University, Hangzhou, China*.

The aim of this study was to investigate the effect of exogenous fibrolytic enzymes (EFE) as potential additives to improve the utilization of corn silage. A total of 11 commercial EFE, including 5 cellulase and 6 xylanase products, were used. Endoglucanase and xylanase were considered as key enzymatic activities for cellulase and xylanase products, respectively. One unit of enzyme activity was expressed as the amount of enzymes needed to release 1 μmol xylose or glucose per min per gram of enzyme products at 39°C and pH 6.6. Corn silage as substrate was added without (control) or with cellulase (30 units/g DM), xylanase (40 units/g DM). Rumen content was collected from 3 donor animals before feeding in the morning. Gas pressure measurements were recorded at 3, 6, 9, 12, 24 and 48 h of incubation and then converted to gas volume. At the end of the incubation, the fluid was sampled to determine the pH and volatile fatty acids (VFA). The contents in the bottle were completely rinsed out to determine the degradation of DM and neutral detergent fiber. Methane production was determined at 6, 12 and 24 h of incubation. Addition of cellulase resulted in a decreased potential gas production ($P < 0.05$) but increased rate of gas production ($P < 0.05$) with the highest rate being 83.7% compared with the control. Total VFA concentrations were increased ($P < 0.05$) for some EFes, but degradation of DM and neutral detergent fiber was enhanced ($P < 0.05$) for all enzyme additives with highest value at 65.4%. The enzymes showed various effects on methane production. From the results of this study, it is indicated that addition of EFes has potential to inhibit methane production while increasing fiber digestibility of corn silage.

Key Words: corn silage, exogenous fibrolytic enzymes, rumen fermentation