

Graduate Student Competition: ADSA-ASAS Northeast Section

145 The effect of an exogenous amylase on performance and total tract digestibility in lactating dairy cows. M. M. McCarthy^{*1}, M. A. Engstrom², E. Azem³, and T. F. Gressley¹, ¹University of Delaware, Newark, ²DSM Nutritional Products Inc., Parsippany, NJ, ³DSM Nutritional Products, Ltd., Basel, Switzerland.

The objective of this trial was to determine performance and digestibility response of lactating dairy cows to a reduced starch diet containing a commercial amylase product. Twenty-six Holstein cows (82 ± 60 DIM) were blocked by parity and DIM and assigned to treatments in a 3 × 3 Latin square design, with 28-d periods. Treatments were normal starch TMR (NS), reduced starch TMR (RS), and reduced starch TMR with exogenous amylase (RSE). The hypothesis was that RS would decrease milk production and diet digestibility compared with NS and that RSE would alleviate some of this decrease. Rations were 41% concentrate and the NS TMR contained 12.8% corn grain, 2.9% soy-hulls, and 2.9% citrus pulp. The RS and RSE TMR contained 6.0% corn grain, 6.9% soyhulls, and 6.9% citrus pulp. Starch concentrations in NS, RS, and RSE TMR were 27.5, 23.2, and 22.4%, respectively. Milk production and DMI were measured daily and milk composition was measured weekly. Fecal grab samples were collected at the end of each period and digestibility of DM and nutrients were determined. Data were analyzed using a mixed model containing the fixed effects of treatment, week, period, and their interactions, and the random effects of cow and block. Contrast statements were used to evaluate effects of dietary starch (NS vs. RS + RSE) and enzyme (RS vs. RSE). There was no effect of starch or enzyme on DMI, milk composition, or starch digestibility ($P > 0.10$). Increased dietary starch increased yields of milk (47.6 vs. 46.2 kg/d, $P = 0.03$), protein (1.38 vs. 1.34 kg/d, $P = 0.03$), and lactose (2.25 vs. 2.18 kg/d, $P = 0.02$) and tended to increase fat yield (1.33 vs. 1.28 kg/d, $P = 0.02$). Addition of amylase did not affect any production parameters. Increased starch reduced NDF digestibility (41.4 vs. 47.0%, $P < 0.01$). Addition of amylase increased digestibility of DM (72.1 vs. 70.1%, $P = 0.02$) and CP (73.4 vs. 71.1%, $P = 0.05$), and tended to increase OM digestibility (73.4 vs. 72.0%, $P = 0.10$). Although addition of exogenous amylase increased nutrient digestibility in a low starch TMR, this was not reflected by improved animal performance.

Key words: amylase, starch, by-product feeds

146 Spoilage yeasts in silage have the potential to directly impact rumen fermentation. M. C. Santos^{*1}, A. L. Lock², G. D. Mechor³, and L. Kung Jr.¹, ¹University of Delaware, Newark, ²Michigan State University, East Lansing, ³Elanco Animal Health, Greenfield, IN.

Yeasts associated with aerobic spoilage of high moisture corn (HMC) and corn silage (CS) were isolated and characterized to determine their potential for direct effects on rumen fermentation. Samples were obtained from 21 US dairy farms; HMC averaged 6.3 and CS averaged 5.4 log₁₀ cfu of yeasts/g of fresh forage. *Candida valida* (CV) was the most predominant species accounting for 35 and 31% of total isolates in HMC and CS, respectively. One isolate of CV was added to in vitro culture tubes containing TMR, buffer and rumen fluid at theoretical concentrations of 0, 4.4, 6.4 and 8.4 log₁₀ cfu/ml; the 6.4 dose was equivalent to a cow consuming 30 kg of fresh CS with 7.0 log₁₀ cfu/g. After 12 and 24 h of incubation at 39°C, samples were analyzed for pH, yeast number, NDF-D, volatile fatty acids (VFA) and fatty acids (FA). Culture pH declined from 6.8 at 0 h to 6.4 and 6.3 after 12 and 24 h, respectively ($P < 0.01$). After 24 h, numbers of viable yeasts for the

control treatment decreased from 2.4 to 0.4 log₁₀ cfu/ml. For the other levels, the measured numbers at time 0 decreased from 4.0, 5.9 and 8.1 to 2.2, 3.9 and 5.3 log₁₀ cfu/ml after 24h, respectively. Inoculation with CV caused a linear decrease in NDF-D at 12 and 24 h ($P < 0.01$). After 12 h, NDF-D for the highest CV addition was 34 vs 44% for control and after 24h NDF-D was 52 vs 58%. At 24 h, the concentration of total VFA, acetate and propionate was 106, 57 and 29 mM for the highest CV dose whereas for control the concentrations were 98, 53 and 25 mM, respectively ($P < 0.05$). FA analysis of CV indicated that it contained ~25% SFA, 60% cis MUFA, and 15% cis PUFA. Overall, the biohydrogenation of unsaturated FA was not altered across treatments and declined over time with an increase in the accumulation of SFA, especially stearic acid; under the conditions tested, CV did not alter the formation of BH intermediates. The results of this study indicate that addition of CV, especially in high levels, can decrease NDF-D and may alter the concentration of propionate and acetate. However, no changes in the production of BH intermediates were detected under the in vitro conditions tested.

Key words: *Candida valida*, NDF-D, biohydrogenation

147 The effects of PPAR-gamma agonist and conjugated linoleic acid on mammary and hepatic lipid metabolism in lactating mice. D. Vyas^{*1}, B. B. Teter¹, P. Delmonte², and R. A. Erdman¹, ¹Department of Animal and Avian Sciences, University of Maryland, College Park, ²U.S. Food and Drug Administration, Center for Food Safety and Applied Nutrition, College Park, MD.

Previous studies have demonstrated the antagonizing effects of PPAR-gamma agonists on conjugated linoleic acid (CLA) induced hepatic steatosis and adipose tissue lipodystrophy. We hypothesized that the PPAR-gamma agonist, Rosiglitazone (ROSI), might also antagonize the CLA induced reduction in milk fat synthesis in lactating mice. Our objective was to investigate the interaction of ROSI and CLA on mammary and hepatic lipid metabolism in lactating C57Bl/6J mice. Nineteen lactating mice were randomly assigned to one of 4 treatments (n = 4–5 per treatment) applied from Day 6 to 10 postpartum. Treatments included: 1) Control (C) diet; 2) Control plus 1.5% dietary CLA (CLA); 3) Control plus intra-peritoneal (IP) ROSI injections (10 mg/kg BW) (ROSI); and 4) CLA plus ROSI (ROSI-CLA). Mice on the C and CLA diets received IP phosphate buffered saline (PBS). Day 6 values were used as covariates for milk fat and pup body weight in the 2 × 2 factorial analysis of covariance. Food intakes were similar among treatments although there was a trend ($P = 0.09$) for increased intake with ROSI. Milk fat was depressed 42% by CLA ($P < 0.001$). ROSI significantly reduced milk fat ($P = 0.05$) and the depression was greater with ROSI-CLA. Liver weights were increased by CLA ($P < 0.001$) and reduced ($P = 0.005$) by ROSI. Pup weight gain was reduced 44% by CLA ($P < 0.001$) but not affected by ROSI. Rosiglitazone corrected the apparent steatosis effect of CLA but had no effect on CLA induced milk fat depression. As ROSI is an insulin sensitizing agent, reduced liver lipid accumulation and increased fat oxidation, along with increased glucose uptake by adipose tissue might explain reduced liver weights with ROSI treatment. Since glucose is used as a precursor for de novo fatty acid synthesis in rodents, the reduction in milk fat with ROSI-CLA could have been due to increased glucose utilization in peripheral tissues thereby reducing glucose availability for triglyceride synthesis in the mammary gland.

Table 1.

Item	Control	ROSI	CLA	ROSI-CLA	SEM
Dam food intake, g/d	6.76	7.73	5.61	6.69	0.62
Milk fat (%)	35.47 ^a	34.42 ^a	21.94 ^b	18.48 ^c	1.02
Liver weight, % of BW	8.54 ^b	7.46 ^c	10.54 ^a	8.72 ^b	0.51
Pup weight gain, g/d	0.44 ^a	0.38 ^a	0.23 ^b	0.23 ^b	0.03

Key words: CLA, PPAR- γ , milk fat

148 Expression of T-box (Tbx) 3 in bovine mammary epithelial cells. M. L. Procopio*, A. C. Lopez, K. M. McFadden, T. A. Hoagland, G. W. Kazmer, and K. E. Govoni, *Department of Animal Science, University of Connecticut, Storrs.*

Development of the bovine mammary gland is a complex process that is regulated by several hormones, growth factors and transcription factors. Tbx3 is a transcription factor that is required for mammary gland development in humans, regulates cell cycle, and is overexpressed in many breast cancer cell lines. We recently demonstrated that growth hormone (GH) treatment increases the mRNA expression of Tbx3 in osteoblast cells independent of insulin-like growth factor (IGF)-I. Based on the critical role of Tbx3 in mammary gland development and

its response to GH, we hypothesize that GH treatment will increase Tbx3 expression in bovine mammary epithelial cells (MEC). Primary bovine MEC were isolated from lactating cows at slaughter and cultured in Dulbecco's Modified Eagle Medium (DMEM) supplemented with fetal bovine serum. In addition, MAC-T cells, a well established MEC line, were cultured under similar conditions. Prior to treatment, cells were serum starved in phenol red free media for 24 h. Cells were treated with control (CON) media (DMEM + 0.2% BSA), and GH (500 ng/ μ L) or IGF-I (200 ng/ μ L) for 24 h and RNA was extracted. mRNA expression was determined by real-time RT-PCR and data were analyzed using students *t*-test. Expression of Tbx3 was similar between MEC and MAC-T cells ($P = 0.7363$), therefore the primary MEC were used for additional experiments. Surprisingly, we did not observe a significant change in Tbx3 expression in cells treated with GH ($P = 0.10$). However, IGF-I treatment reduced Tbx3 expression 1.5-fold compared with CON ($P < 0.05$). In summary, we did not observe any effect of GH on Tbx3 expression in MEC, however a slight decrease was observed with IGF-I treatment. These findings are in contrast to the role of Tbx3 in mediating GH in the osteoblast, thus demonstrating that Tbx3 action may be cell and/or tissue specific.

Key words: mammary epithelial cells, growth hormone, transcription factors