

Undergraduate Student Competition: ADSA-SAD Undergraduate Poster Competition: Original Research

T98 Feeding 5-hydroxy-L-tryptophan (5-HTP) to transition rats increases serum serotonin (5-HT) and calcium and down-regulates mRNA expression of calcium transporters in the gut. C. Cronick*, J. Laporta, S. Weaver, and L. L. Hernandez, *University of Wisconsin, Madison*.

The absorption of calcium (Ca^{2+}) and role of serotonin (5-HT) in the gut are independently well-understood processes. Less clear is the effect of 5-HT on the absorption of Ca^{2+} by the gut during the transition from pregnancy to lactation. In order to better understand 5-HT's effect on Ca^{2+} dynamics in the gut during lactation we fed female rats two different diets: a control diet (CON, $n = 15$) and a diet supplemented with 5-hydroxytryptophan (5-HTP, 0.2% of total diet, $n = 15$), the immediate precursor for 5-HT synthesis, from d13 of pregnancy through d9 of lactation. We collected serum on d1 and 9 of lactation to measure circulating 5-HT and Ca^{2+} concentrations, and also measured Ca^{2+} concentration in the milk. Rats were euthanized on d9 of lactation and gut samples were collected. mRNA expression of tryptophan hydroxylase 1 (TPH1), 5-HT reuptake transporter (SERT), plasma membrane Ca^{2+} ATPase 1 (PMCA1), $\text{Na}^{+}\text{-Ca}^{2+}$ exchanger 1 (NCX1), transient receptor potential cation channel, subfamily V, member 6 (TRPV6), voltage-dependent Ca^{2+} channel ($\text{Ca}_v1.3$), and Ca^{2+} binding protein calbindin- D_{9k} (CalbD9k) were measured by real-time RT-PCR. Serum 5-HT was elevated on d 1 (4.7 ± 0.39 ng/mL vs. 1.83 ± 0.26) and d9 (8.3 ± 0.7 vs. 3.4 ± 0.25 ng/mL) of lactation in the 5-HTP compared to CON dams. On d 1 of lactation 5-HTP dams had increased serum Ca^{2+} concentrations compared to CON ($P = 0.0085$), while no significant changes in milk Ca^{2+} were observed. On d9 of lactation 5-HTP dams had similar serum Ca^{2+} concentrations to the CON dams, but demonstrated increased milk Ca^{2+} concentrations ($P = 0.032$). Gut mRNA expression of SERT and TPH1 was decreased in 5-HTP dams compared to CON ($P < 0.041$). Gut mRNA expression of all Ca^{2+} transporters evaluated were decreased in 5-HTP dams compared to CON ($P < 0.05$). It is probable that during lactation the gut decreases 5-HT synthesis and reuptake as well as expression of Ca^{2+} transporters, because the mammary gland is increasing production of 5-HT and transport of Ca^{2+} into the milk to provide adequate nutrition to the neonate.

Key Words: serotonin, gut, calcium

T99 Mammary clock regulation and function. E. Erickson*, J. Crodian, M. Schutz, T. Casey, and K. Plaut, *Purdue University, West Lafayette, IN*.

Most physiological processes, including lactation, are controlled by the circadian system, which coordinates growth, development, and metabolism to optimal times of day through generation of circadian rhythms. Approximately 10% of genes expressed show circadian rhythms that are generated by interactions of molecular clock genes (e.g., *Clock*, *Bmal1*, *Per1*). BMAL1-CLOCK heterodimers function as a transcription factor that binds E-box sequences in promoters of clock controlled genes. Little is known about how mammary clock is regulated or what genes it controls. We hypothesize mammary clock is regulated by lactogenic hormones and it regulates genes important to mammary development and milk synthesis. Our objectives were to (1) Determine effects of lactogenic hormones on circadian rhythms of molecular clock genes *Bmal1* and *Per1* in mouse mammary epithelial cells, HC-11; and (2) knock down CLOCK to measure effects

on *Per1* and cell cycle gene, cyclin D1 (*Ccnd1*) expression. HC-11 cells were grown to confluence, treated for 2h with RPMI media + 10% calf serum \pm lactogens (5 $\mu\text{g/mL}$ insulin, 2 $\mu\text{g/mL}$ prolactin, 40 ng/mL dexamethasone), after which RNA was collected every 4 h for 48 h to capture circadian gene expression. Q-PCR analysis showed lactogens induced 24-h rhythms of *Bmal1* and *Per1* expression and a 2.5-fold increase in *Bmal1* rhythm amplitude (i.e. peak-trough); 2-way ANOVA revealed significant time and treatment effects ($P < 0.05$). For the second objective, HC-11 cells were transfected with shRNA plasmids with insert mouse *Clock* sequence I [AAACCCACATTCCT-TAGTAAT] or II [ATCAAACCCTGGATTGAATTT]; a negative control was transfected with plasmid insert sequence: ggaatctcattc-gatgcatac. Eighteen clonal cell lines were created through dilution and lines with sequence I or II insert were selected based on greatest *Clock* knockdown. Cells were synchronized by RPMI + 10% calf serum media change and RNA was isolated 4 h later. Q-PCR analysis revealed the 40% knock down of *Clock* by sequence I and the 42% knock down by II was associated with 70% and 35% knock down of *Per1* and a 2.3-fold or 1.8-fold induction of *Ccnd1*, respectively. Data suggest lactogens regulate mammary clock, and mammary clock may regulate *Ccnd1*.

Key Words: lactation, mammary, circadian

T100 Potential for compost bedded pack barns in sustainable organic dairy farming systems. H. A. Mussell*¹, J. L. Taraba², and J. M. Bewley¹, ¹University of Kentucky, Department of Food and Animal Sciences, Lexington, ²University of Kentucky, Agricultural Engineering, Lexington.

When managed properly, compost bedded pack barns (CBP) are an excellent housing alternative for dairy cows. Although most CPB barns have been constructed for conventional dairies, these barns fit within the spirit of organic dairy farming by relying on a natural process (composting) to produce a comfortable environment for lactating dairy animals that maximizes cow comfort and longevity and minimizes mastitis and other health problems. These barns provide more freedom for cows to move around or lie down as compared to stall based dairy facilities. The composting process increases the bedding temperature to control disease-causing microorganisms and decreases the bedding moisture by increasing the drying rate. Additionally, CBP barns serve as an area for manure storage and reduce the volume and frequency for manure application to fields. The objective of this study was to examine cow and pack performance metrics within three Kentucky organic dairy farms housing cows in CBP barns. Temperatures (0.1, 0.2, and 0.3 m below pack surface) and bedding samples were collected three times from nine different locations within each barn. Samples were collected at each farm for moisture content and nutrient analysis. Forty cows per farm were scored for hygiene and locomotion at each visit. Mean pack temperature at 0.30 below the pack surface was $27.79^\circ\text{C} \pm 12.03$. These temperatures were lower than the recommended range of 43 to 65°C . Two of the three farms were not consistently tilling their packs, which may have contributed to the lower temperatures observed during these visits. Mean moisture was $57 \pm 7\%$, within the recommended range of 50 to 60%. Mean C:N ratio was 32:1, higher than the ideal range of 25:1 to 30:1. Mean locomotion and hygiene scores were 1.54 ± 0.28 and 1.72 ± 0.55 , respectively, demonstrating that cows managed in organic

dairy compost bedded pack barns can be sound and clean. Compost bedded pack dairy barns could meet the farmer, land and cow needs of organic dairy operations.

Key Words: compost bedded pack barns, cow comfort, organic

T101 Effects of microbial fermentation products on milk production in dairy cows during heat stress. R. M. Wagner*¹, S. I. Kehoe¹, and D. DuBourdieu², ¹*University of Wisconsin-River Falls, River Falls,* ²*R&D Lifesciences, Menomonie, WI.*

Heat stress is a problem that reduces milk production for dairy producers. The objective of this research was to determine the production benefits of feeding a combination microbial product containing fermentation products from *Bacillus subtilis*, *Trichoderma viride*, and *Aspergillus oryzae* (Lactomace; R & D Lifesciences, LLC, Menomonie, WI) during a summer heat wave. Two pens of lactating dairy cattle were matched in DIM and assigned to either a Control treatment with no microbial product (C) or a Microbial treatment (M) fed 5 gm/hd/day for the first 30 days and 10 gm/hd/d for the second 30 days. Daily milk yield and DMI and calculated 3.5% fat-corrected milk (FCM) and FCM:DMI ratio were recorded. Milk fat and protein % were collected from bulk

tank samples after the first 30 d and 60 d. During data collection, heat index (HI) rose to an average of 32°C thereby causing significant heat stress. Data were analyzed using a paired TTest. During periods of high HI, from 29°C to 45°C, least squares means of FCM were significantly higher for the M group compared with C (42.41 ± 1.71 kg and 38.45 ± 1.71 kg, respectively). During this heat time frame, milk yield from C cows was reduced by 15% and M cows by 5% although least squares means were significantly higher for the C group (41.54 ± 1.85 kg and 39.25 ± 1.58 kg for C and M, respectively). The M group had significantly higher FCM:DMI ratios (0.78 ± 0.05 and 0.67 ± 0.05 kg FCM/kg DMI for M and C, respectively). Dry matter intake was not different between treatments (24.60 ± 1.62 kg and 25.26 ± 4.56 kg for M and C, respectively). During the high HI time frame, milk fat % increased in the M treatment compared with a decrease in C cows (+0.56% compared with -0.46% for M and C, respectively). Results of this study conducted on a commercial dairy farm in WI during summer months indicate that FCM is significantly increased with an increased FCM efficiency. These results indicate that supplementation of these fermentation byproducts may improve digestion and utilization of feedstuffs in the diet thus abating the effects of heat stress and supporting production.

Key Words: microbial product, heat stress, milk fat