

Dietary anion-cation difference and day length differently affect milk calcium secretion pathways

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LACTATION BIOLOGY I

0404 Temporary alterations to milking frequency, immediately postpartum, modifies expression of milk synthesis and apoptosis genes in the mammary glands of grazing dairy cows. T. M. Grala¹, J. K. Kay², J. R. Roche², A. G. Rius³,

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Temporary changes to postpartum milking frequency can elicit lactation-long changes in milk production. The hypothesis tested in this experiment was that the immediate and long-term milk yield response to altered milking frequency would correspond to gene expression changes associated with mammary cell secretory activity and/or number. Multiparous, grazing Holstein-Friesian cows (n = 150) were randomly assigned to 1 of 5 groups at calving: milked once daily $(1\times)$ for 3 or 6 wk and twice daily $(2\times)$ thereafter; milked $2\times$ for the entire lactation (control); or milked thrice daily $(3\times)$ for 3 or 6 wk and $2\times$ thereafter. Milk yields were recorded daily and milk composition weekly. Mammary tissue was collected at 3, 6, and 9 wk postpartum (n = 12 cows/treatment), and gene expression measured using quantitative reverse transcription PCR. Data were analyzed using mixed models fitted with REML (Restricted Maximum Likelihood) in GenStat, including: treatment and contrasts to test milking frequency $(1 \times, 2 \times, 3 \times)$, duration (3) wk, 6 wk), and their interaction as fixed effects, and cow as a random effect. Immediate (P < 0.001) and lactation-long (P< 0.01) decreases in milk and energy-corrected (ECM) production were recorded in cows milked 1× postpartum relative to 2×; however, cows milked 3× only produced greater (P <0.05) milk volumes during the treatment period and did not differ (P > 0.05) in ECM production. Transcript levels from genes involved in milk fat (ACACA, FASN), protein (CSN1S1, CSN2), and lactose (LALBA, B4GALT1) synthesis were not altered in cows milked $3\times$, but were downregulated (P < 0.05) at 3 and 6 wk postpartum in cows milked 1×. Decreased (P <0.05) expression of these genes was maintained after $1 \times cows$ were switched to 2× milking. Furthermore, at 9 wk postpartum, cows milked $1 \times$ for 3 wk had lower (P < 0.05) expression of genes involved in fat and lactose synthesis than cows milked $1 \times$ for 6 wk. In contrast, apoptotic genes (*PYCARD*, *FAS*) were up-regulated (P < 0.05) in cows milked 1×. This effect was still apparent at 9 wk (P < 0.01), indicating that greater mammary cell death was maintained post-treatment. In conclusion, greater milk volumes during 3× milking were not associated with altered expression of genes involved in milk synthesis or mammary cell death. However, changes in expression of genes involved in these processes may underpin the long-term reduction in milk and ECM yields in cows milked 1× postpartum.

Key Words: milking frequency, mammary apoptosis, gene expression

0405 Dietary anion-cation difference and daylength differently affect milk calcium secretion pathways. M. Boutinaud*¹, A. Bondon¹, A. Narcy², C. Hurtaud¹, M. Johan¹, J. Couedon¹, and P. Lamberton³, ¹INRA, Saint Gilles, France, ²INRA, Nouzilly, France, ³INRA, Le Rheu, France.

Milk is an important source of Ca for growth and development of children. However, it has been recently shown that dairy milk Ca content decreases with long-day photoperiod and varies according to the type of diet. Many proteins are involved in the secretion of Ca into milk by the mammary epithelial cell (MEC). The aim of this study was to identify the role of gene expression of these proteins in the regulation of milk Ca content. A trial was performed according to a Latin square design using 8 dairy cows averaging 103 ± 44 d in milk, with 2 treatments in a factorial arrangement with 4 periods of 14 d. The cows received 2 levels of dietary anion-cation differences (DCAD; 0 mEq/kg DM for d 0 and 400 mEq/kg for d 400) and 2 daylengths (8 h of light/d for short days and 16 h/d for long days). The DCAD treatments were conceived to mimic diets based either on corn silage or on herbage. The cows were exposed to solarium lights providing UVA and UVB. Once per period, MEC were prepared after milk centrifugation by purification using an anti-cytoketatin antibody bound to magnetic beads to study by real-time RT-PCR the mRNA level of genes involved in Ca secretion expressed related to RPLP0 housekeeping gene. Data were analyzed using Mixed procedure. There was no significant interaction between daylength and DCAD level. Milk and Ca yields did not vary with any treatments, averaging 32.7 kg/d and 41.1 g/d, respectively. With d 400 compared with d 0, milk Ca content increased (P < 0.01) with no link with casein content. No significant variation was observed on gene expression with DCAD treatment and on kappa casein and a-lactalbumin mRNA levels with any treatments. Milk Ca and casein content were lower with long days compared with short days (P < 0.05). The lower Ca secretion was associated with lower mRNA levels for SPCA1, ITPR1, and PMCA1, 3 Ca transporters in milk purified MEC (P <0.05). This work suggests that Ca secretion pathways may be downregulated with long photoperiod, and that could explain a part of the seasonal decrease of milk Ca content during summer. In contrast, no significant variation of gene expression could explain the increase in milk Ca content with d 400.

Key Words: milk calcium, mammary epithelial cell, photoperid, feeding