



Regulation of cell number in the mammary gland via the control of the exfoliation process in milk in ruminants

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► To cite this version:

Lucile Herve, Vanessa Lollivier, Hélène Quesnel, Marion Boutinaud. Regulation of cell number in the mammary gland via the control of the exfoliation process in milk in ruminants. ADSA-ASAS Joint Annual Meeting, Jul 2015, Orlando, United States. pp.219. hal-01211053

HAL Id: hal-01211053

<https://hal.science/hal-01211053>

Submitted on 3 Jun 2020

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98 New insights in the importance of prolactin in dairy ruminants. Pierre Lacasse^{*1}, Séverine Ollier¹, Vanessa Lollivier², and Marion Boutinaud², ¹*Dairy and Swine R&D Centre, Sherbrooke, QC, Canada*, ²*INRA, Agrocampus Ouest, UMR1348 PEGASE, Saint Gilles, France*.

In most mammals, prolactin (PRL) is essential for maintaining lactation and its suppression inhibits lactation. However, the involvement of PRL in the control of ruminant lactation is less clear because inconsistent effects on milk yield have been observed with short-term suppression of PRL by bromocriptine. Therefore, several experiments were conducted to assess the galactopoietic role of PRL. In an initial experiment, cows in early lactation received daily injections of the dopamine agonist quinalgide (QUIN) for 9 weeks. QUIN reduced milking-induced PRL release and caused a faster decline in milk production. Milk production was correlated with the amount of PRL released at milking. QUIN reduced mammary epithelial cell activity, survival and proliferation. In goats, QUIN did not affect either basal or milking induced PRL release and milk production, whereas injection of cabergoline, another dopamine agonist, caused a decrease of 28% of milk yield the day after the injection. In another experiment, cows were injected for 5 d with QUIN; QUIN + injection of bPRL at milking time; or vehicles. Again, milk, protein and lactose yield were decreased by QUIN. Although PRL injections were not sufficient to restore milk yield, they tended to increase milk protein and lactose yields and increased the viability of milk purified mammary epithelial cells. In late lactation cows, QUIN decreased milk production within the first day of treatment and induced a more rapid changes in several markers of mammary gland involution after drying-off. Similarly, injection at drying-off of cabergoline hastened mammary involution and enhanced mammary gland remodeling. Recently, we stimulated PRL secretion with daily injection of the dopamine antagonist domperidone for 5 weeks. Milk production increased gradually and was greater in domperidone-treated cows during the last 4 weeks of the treatment period. Milk production of both groups became similar again 5d after the last injection. In conclusion, these data, combined with those from other studies, provide a good body of evidence that PRL is galactopoietic in dairy ruminants.

Key Words: prolactin, milk production, cows

99 Regulation of cell number in the mammary gland via the control of the exfoliation process in milk in ruminants. Lucile Herve^{1,2}, Vanessa Lollivier^{1,2}, Hélène Quesnel^{1,2}, and Marion Boutinaud^{*1,2}, ¹*INRA UMR1348, Saint Gilles, France*, ²*Agrocampus Ouest UMR1348, Rennes, France*.

Milk yield is partly influenced by the number of mammary epithelial cells (MEC) in the mammary gland. It is well known that MEC number varies due to cell proliferation and apoptosis. The exfoliation of MEC from the mammary epithelium into milk is another process which might influence MEC number in the mammary tissue. Yet, little is known about the control of MEC exfoliation process. The rate of MEC exfoliation can be assessed by measuring the milk MEC content through flow cytometry analysis or through an immuno-magnetic method for MEC purification. Various experimental models were used to affect milk yield and study the rate of MEC exfoliation. Reducing milking frequency from twice to once daily increased MEC loss per day in goat but not in cow milk. An increased daily rate of MEC exfoliation was also observed during short days as compared with long days or in response to an endotoxin-induced mastitis in cows. Other animal models were designed to investigate the endocrine control of the exfoliation process and its link with milk production. Suppression of ovarian steroids by ovariectomy resulted in a greater persistency of lactation and a decrease in MEC exfoliation.

Administering prolactin inhibitors enhanced MEC loss while exogenous prolactin tended to prevent this negative effect of prolactin inhibitors. These findings suggest that prolactin could regulate MEC exfoliation. In most of these studies, variations of MEC exfoliation were associated with variations in milk yield and changes in mammary epithelium integrity. Exfoliation of MEC could be a process that regulates MEC number in the mammary tissue, and thereby could influence milk yield and lactation persistency.

Key Words: cow, lactation, mammary epithelial cell

100 Mammary response to infection: A critical balance between pathogen elimination and collateral damage. David E. Kerr^{*}, *University of Vermont, Burlington, VT*.

Mastitis is an inflammatory disease of the mammary gland. The disease is generally classified as sub-clinical (no obvious signs) or 3 levels of clinical disease including: mild (abnormal milk); moderate (abnormal milk with swelling or redness of the gland); severe (abnormal milk, gland inflammation, with systemic signs of illness). Recent large-scale studies indicate that mild, moderate, and severe forms make up approximately 60%, 30%, and 10% of clinical cases of mastitis, with severe mastitis predominately caused by gram-negative bacteria. Efforts to reduce severe mastitis are of utmost importance to dairy animal welfare, and these cases are associated with greatest milk production losses. Experimental challenge studies under controlled conditions reveal animal-to-animal variation in the severity of the resulting mastitis. This suggests a genetic basis to disease severity and the potential for finding genetic markers for use in breeding programs to produce animals with a reduced tendency to develop severe mastitis. However, the evolving field of epigenetics suggests that in utero and early life environments can modify gene expression and thus modify an animal's phenotype. Our approach is to develop a cell culture challenge model predictive of an animal's innate response phenotype. Such a model could potentially be used with cells from young animals to determine their response phenotype and thus facilitate selection of herd replacements. In our dermal fibroblast model, the cells are cultured under controlled conditions and then challenged with LPS to determine innate response magnitude. This model has revealed breed differences (Angus vs. Holstein) and epigenetic differences in samples from the same animals (i.e., same genotype) collected at 5 and 16 mo of age. Further, animals with low vs. high fibroblast response phenotype produce less BSA in milk following experimentally induced mastitis. Future studies employing this and other model systems, combined with well-controlled disease challenges of extreme phenotypes will lead to a greater understanding of factors contributing to animal variation in the severity of response to mammary infection.

101 Blood-derived proteins in milk during the colostral period: Active or passive transfer? Samantha K. Wall^{*1}, Josef J. Gross¹, Evelyne C. Kessler¹, Kris Villez², and Rupert M. Bruckmaier¹, ¹*Veterinary Physiology, Vetsuisse Faculty University of Bern, Bern, Switzerland*, ²*Swiss Federal Institute of Aquatic Science and Technology, Dübendorf, Switzerland*.

Colostrum has a different composition than milk in established lactation. This difference is in part due to the partially open blood-milk barrier, which prevents the interdiffusion of blood and milk components. In the first days of lactation, α -lactalbumin (LALBA), a milk protein, is typically present in blood and several blood-derived proteins are present in milk such as IgG₁ (very high concentration), IgG₂, serum albumin (ALB), and lactate dehydrogenase (LDH). With the exception of IgG₁, which is transferred by active transcellular transport, other proteins are

thought to pass paracellularly through the temporarily open barrier. This study aimed to examine the decline patterns of each protein relative to IgG₁, to distinguish between paracellular and transcellular transport through the blood-milk barrier during the first days of lactation. Ten Holstein cows were milked at 4 h after parturition, the next 5 consecutive milkings, and the afternoon milking on d 5, 8, 10, and 14 of lactation for a total of 10 milking time points and blood samples were taken in parallel. Blood and milk samples were analyzed for the concentrations of LDH, ALB, IgG₁, IgG₂ and LALBA. Protein concentration curves were generated from all 10 time points and were evaluated using the tau time constant model to determine the rate of decline of the slope of each protein. When examining blood-derived proteins in milk, the

concentration of IgG₁ declined significantly faster than the proteins IgG₂ and LDH. Interestingly, the decline of ALB was not statistically different from IgG₁ nor IgG₂ and LDH. IgG₁ concentration in milk far exceeded levels in plasma, and this protein exhibited a recovery increase in plasma during the experimental period. IgG₂, ALB, and LDH concentrations in milk did not reach plasma levels. Plasma LALBA followed a different pattern, declining significantly slower than all blood-derived proteins in milk. These results indicate that there is active transport of only IgG₁, with a sharp decline at parturition, compared with IgG₂, ALB, LDH, and LALBA which are following the closure of the blood-milk barrier.

Key Words: colostrum, blood-milk barrier, blood-derived protein