

# LACTATION RESEARCH IN MAMMALS AND HUMANS:

## THE MAMMARY GLAND IN HEALTH AND DISEASE

WITH PARTICULAR FOCUS ON  
THE MAMMARY GLAND CLOSE POST PARTUM  
including indicators of inflammation  
and pathogens of the mammary gland

PROCEEDINGS FROM A SYMPOSIUM IN UPPSALA, SWEDEN  
DECEMBER 7-8, 2010

Sigrid Agenäs, Gunilla Hallberg, Elisabeth Kylberg, Karin Persson Waller & Bodil Ström Holst (*editors*)

Uppsala, 2010

CRU Report 24

*Supported by:*



WELCOME to the CRU web!

<http://www.slu.se/sv/centrumbildningar-och-projekt/ny-centrum-for-reproduktionsbiologi/>

ISSN 1404-5915

ISBN 978-91-576-9013-5

Layout: Sigrid Agenäs

©2010 CRU Report 24, Uppsala

Tryck: SLU Service/Repro, Uppsala 2010

## CONTENTS

<b>Foreword</b>	4
<b>Programme</b>	5
<b>Oral presentations</b>	
<b>BREASTFEEDING POLICIES AND RECENT FINDINGS ON THE EFFECTS OF EARLY SUCKING BEHAVIOR ON LACTATION SUCCESS IN WOMEN</b> Kylberg, E.	6
<b>POSITIVE EMOTIONS AND SUCCESSFUL LACTATION IN THE NEONATAL UNIT: A QUALITATIVE STUDY</b> Swanson, V. & McInnes, R.	8
<b>A PROCESS-ORIENTED BREASTFEEDING TRAINING PROGRAM FOR HEALTHCARE PROFESSIONALS TO PROMOTE BREASTFEEDING</b> Ekström, A., Kylberg, E. & Nissen, E.	9
<b>LONG CHAIN POLYUNSATURATED N-3 FATTY ACIDS (PUFA) IN COLOSTRUM ARE AFFECTED BY FISH INTAKE DURING PREGNANCY</b> Olang, B., Faraji, M. H., Hellstrand, S., Palesh, M., Azadnyia, E., Kamali, Z., Ali, M., Strandvik, B. & Yngve, A.	10
<b>RECENT HOT TOPICS IN HUMAN AND BOVINE LACTATION INCLUDING HIGHLIGHTS FROM THE ISRHML MEETING</b> McGuire, M. A. & McGuire, M. K.	11
<b>TIMING OF MAMMARY ANGIOGENESIS: A POSSIBLE EXPLANATION FOR <i>POST-PARTUM</i> APOPTOSIS IN THE BOVINE UDDER?</b> Agenäs, S., Nielsen, M. O., Safayi, S. & Knight, C. H.	13
<b>DAY-TO-DAY VARIATION IN MILK COMPOSITION AND MILK YIELD AT UDDER QUARTER LEVEL</b> Forsbäck, L., Lindmark-Månsson, H., Andrén, A., Åkerstedt, M., Andrée, L. & Svennersten-Sjaunja, K.	14
<b>TRUTH AND MYTH ABOUT MILK PRODUCTION IN THE DEHYDRATED CAMEL (<i>Camelus dromedarius</i>)</b> Dahlborn, K., Bekele, T. & Olsson, K.	15
<b>CHEMOTACTIC FACTORS INVOLVED IN INFLAMMATION OF THE MAMMARY GLAND WITH FOCUS ON THE BOVINE - <i>CAN COMMON MILK CONSTITUENTS HAVE A CHEMOTACTIC EFFECT?</i></b> Östensson, K.	16
<b>HYPOTHESIS: INFLAMMATION IS A HEALTHY PART OF SUCCESSFUL STAGE II LACTOGENESIS IN WOMEN</b> McGuire, M. K. & McGuire, M. A.	18
<b>TOWARDS A CLARIFICATION OF THE CONCEPT “MASTITIS” AS USED IN EMPIRICAL STUDIES OF BREAST INFLAMMATION DURING LACTATION.</b> 2010, Journal of Human Lactation, Feb;26(1):53-9. Review Kvist, L. J.	20
<b>NATURAL VARIATION IN BIOMARKERS INDICATING MASTITIS IN HEALTHY COWS</b> Åkerstedt, M., Forsbäck, F., Larsen, T. & Svennersten-Sjaunja, K.	22
<b>CULTURE INDEPENDENT ANALYSIS OF THE BACTERIAL COMMUNITIES IN MILK FROM WOMEN</b> McGuire, M. A., Hunt, K. M. & McGuire, M. K.	23
<b>ETIOLOGY AND ANTIMICROBIAL SUSCEPTIBILITY OF UDDER PATHOGENS FROM CASES OF SUBCLINICAL MASTITIS IN DAIRY COWS IN SWEDEN</b> Persson, Y., Nyman, A. & Grönlund Andersson, U.	25
<b>LONG-TIME PERSISTENCE OF <i>Staphylococcus aureus</i> IN A HERD WITH MASTITIS PROBLEMS</b> Artursson, K. & Persson Waller, K.	26

## Poster presentations

<b>SURVIVAL AND GROWTH PERFORMANCE OF <i>Staphylococcus aureus</i> IN BEDDING MATERIAL</b> Artursson, K. & Thelander, M.	27
<b>PREDICTION OF MASTITIS BASED ON MILK ELECTRO-CONDUCTIVITY, PH, AND ACUTE PHASE PROTEINS IN DIFFERENT DAIRY ANIMALS IN SUB-TROPICAL CLIMATE</b> Behera, K., Layek, S. S., Prasad, S., Kumaresan, A. & Mohanty, T. K.	28
<b>SEASONAL EFFECT ON MILK SOMATIC CELL COUNT (SCC) AND DIFFERENTIAL LEUKOCYTE COUNT (DLC) IN COWS AND BUFFALOES</b> De, K., Mukherjee, J., Prasad, S. & Dang, A. K.	29
<b>COLOSTRUM QUALITY IN COWS MILKED PREPARTUM</b> Ferneborg, S. & Kronqvist, C.	30
<b>EFFECT OF AN IMMUNOMODULATOR AGENT WITH ANTIBIOTIC THERAPY FOR THE TREATMENT OF COWS DURING THE DRY PERIOD – PRELIMINARY RESULTS</b> Giannechini, R. E., Matto, C. & Gonzalez, M. S.	31
<b>LACTATION RESEARCH IN FINLAND – RESULTS OF A ONE YEAR FOLLOW-UP STUDY</b> Hannula, L.	32
<b>MILK YIELD AND COMPOSITION IN SWEDISH LANDRACE GOATS (<i>Capra hircus</i>) KEPT TOGETHER WITH THEIR KIDS IN TWO DIFFERENT SYSTEMS</b> Högberg, M. & Dahlborn, K.	33
<b>PROTEOME ANALYSIS OF FUNCTIONALLY DIFFERENTIATED MAMMARY EPITHELIAL CELLS ISOLATED FROM COW AND BUFFALO MILK</b> Janjanam, J., Varshney, N., Singh, S., Jamwal, M., Kumar, S., Ajay K., Dang, Jai K., Malakar, K. D., Mishra, B. P., Mukesh, M., Kataria, R., Mukhopadhyay, T., Prakash, B. S., Grover, S., Panigrahi, A. & Mohanty, A. K.	34
<b>INTER-RELATIONSHIP OF PHAGOCYtic ACTIVITY OF BLOOD NEUTROPHILS WITH PLASMA CONCENTRATION OF TNF-<math>\alpha</math>, IL-6 AND MILK SCC IN MASTITIC CROSSBRED COWS</b> Kaswan, S., Mukherjee, J., Prasad, S. & Dang, A.K.	35
<b>THE EFFECT OF A SINGLE PROLONGED MILKING INTERVAL ON INFLAMMATORY PARAMETERS, MILK COMPOSITION AND YIELD IN DAIRY COWS</b> Lakic, B., Svennersten-Sjaunja, K., Norell, L., Bruckmaier, R. M., Dornfalk, J. & Östensson, K.	36
<b>GENETIC VARIATION AND SPREAD OF <i>Staphylococcus aureus</i>, <i>Streptococcus dysgalactiae</i> AND <i>Streptococcus uberis</i> IN MASTITIS IN DAIRY COWS</b> Lundberg, Å., Aspán, A., Nyman, A., Ericsson Unnerstad, H., Östensson, K. & Persson Waller, K.	37
<b>SIGNIFICANCE AND APPLICATION OF THE ONLINE CELL COUNTER IN THE MILKING ROBOT</b> Lusis, I. Antane, V. & Laurs, A.	38
<b>ANALYSIS OF EXPRESSION OF BOVINE INSULIN-LIKE GROWTH FACTOR BINDING PROTEIN-5 (IGFBP-5) DURING LACTATION AND ITS POSSIBLE ROLE IN LACTATION PERSISTENCY</b> Mohapatra, S. K., Singh, S., Varshney, N., Kumar, S., Janjanam, J., Anand, V. J., Kaushik, J. K., Dang, A. K., Datta, T. K., Das, S. K. & Mohanty, A. K.	39
<b>CHRONIC MASTITIS; COULD IT BE A RISK FACTOR FOR BREAST CANCER?</b> Nazemi, S. & Knight, C. H.	40
<b>KANGAROO MOTHER CARE AT TWO SWEDISH NICUS – REPORT FROM AN ONGOING STUDY</b> Thernström Blomqvist, Y., Hedberg Nyqvist, K., Rubertsson, C. & Ewald, U.	41
<b>BEHAVIOUR, HEART RATE AND SALIVA CORTISOL IN GOAT KIDS REARED WITH OR WITHOUT MATERNAL CONTACT</b> Winblad von Walter, L., Hydbring-Sandberg, E. & Dahlborn, K.	42
<b>MASTITIS MONITORING IN THE LOMONOSOVSKI DISTRICT OF THE LENINGRAD REGION</b> Vinokhodova, M. & Sukhinin, A. A.	43

## Foreword

CRU is a multidisciplinary network of almost 100 scientists with different academic backgrounds at SLU, Swedish University of Agricultural Sciences, and UU, Uppsala University. During two days in 2008, Nov 13-14, CRU hosted a symposium on “Lactation research in mammals and humans: comparative aspects with focus on milk composition and mastitis” in Uppsala. The meeting between scientists and research students in human and animal lactation and clinicians from both human and animal medicine proved very successful and it was agreed to make this a biannual tradition.

The second biannual lactation symposium organized by the CRU will take place in Uppsala, December 7-8, 2010. The theme for this symposium is “**Lactation research in mammals and humans: the mammary gland in health and disease** with particular focus on the mammary gland close post partum, including indicators of inflammation and pathogens of the mammary gland”.

Lactation is a fundamental part of reproduction. The composition of milk is important for young both babies and animals, and varies within the lactation period. Optimal nutrition of premature babies is dependent on knowledge of milk composition, and the relationship between milk composition and health later in life is a developing research field. Milk composition in early life of calves is important also for cattle production, and milk composition in dairy cows has a great economical significance for the dairy industry. One way to increase the knowledge about factors influencing milk composition, from synthesis in the mammary gland to handling of milk at the dairy or hospital, is to perform comparative research between human and bovine lactation.

Mastitis is another important field within lactation research. Mastitis in cows has a strong influence both on economy in dairy production and on animal welfare. Treating mastitis is also an important topic, not least because of the increasing problems with microbes that show resistance to antibiotics. In humans, mastitis leads to both economic and health problems for the society. Better methods to prevent, diagnose and treat mastitis are needed. We hope that the bringing together of researchers and clinicians working with different aspects of lactation in animals and humans will lead not only to exchange of information, but also to new ideas and future cooperation.

On behalf of CRU and the organizing committee: Sigrid Agenäs, Gunilla Hallberg, Elisabeth Kylberg, Karin Persson Waller and Bodil Ström Holst wishes you welcome to take part of all aspects of the symposium - oral presentations, panel discussions, posters and informal discussions.

Uppsala, December 2010

*Sigrid Agenäs, Gunilla Hallberg, Elisabeth Kylberg, Karin Persson Waller, Bodil Ström Holst*  
(editors)

CRU SYMPOSIUM ON  
**LACTATION RESEARCH IN MAMMALS AND HUMANS:  
THE MAMMARY GLAND IN HEALTH AND DISEASE**

WITH PARTICULAR FOCUS ON THE MAMMARY GLAND CLOSE POST PARTUM  
including indicators of inflammation and pathogens of the mammary gland

Uppsala, Ultuna, Main building, Lecture room L

**7 December**

09.00	Registration, coffee	
09.30	Welcome, information	<i>Sigrid Agenäs</i>
<b>SESSION 1</b> (Moderator Gunilla Hallberg)		
09.45	Breastfeeding policies and recent findings on the effects of early sucking behavior on lactation success in women	<i>Elisabeth Kylberg</i>
10.35	Positive emotions and successful lactation in the neonate unit: A qualitative study	<i>Vivien Swanson</i>
10.50	A process-oriented breastfeeding training program for healthcare professionals to promote breastfeeding	<i>Anette Ekström</i>
11.05	Coffee, poster walk	
11.30	Long chain polyunsaturated n-3 fatty acids differ significantly in colostrum in relation to fish intake during pregnancy in women	<i>Behesteh Olang</i>
11.45	Panel discussion	
12.15	LUNCH	
<b>SESSION 2</b> (Moderator Sigrid Agenäs)		
13.30	Recent hot topics in human and bovine lactation including highlights from the ISRHML meeting	<i>Mark McGuire</i>
14.20	Timing of mammary angiogenesis: a possible explanation for post-partum apoptosis in the bovine udder?	<i>Chris H Knight</i>
14.35	Day-to-day variation in milk composition and milk yield at udder quarter level	<i>Linda Forsbäck</i>
14.50	Coffee, poster walk	
15.20	Truth and myth about milk production in the dehydrated camel ( <i>Camelus dromedarius</i> )	<i>Kristina Dahlborn</i>
15.35	Panel discussion	
16.15	Conclusions from session 1 and 2	<i>Sigrid Agenäs</i>
16.30	End of programme day 1	
17.30	DINNER (Veterinary Student Union building, Ultuna)	

**8 December**

<b>SESSION 3</b> (Moderator Elisabeth Kylberg)		
09.00	Chemotactic factors involved in inflammation of the mammary gland with focus on the bovine - <i>Can common milk constituents have a chemotactic effect?</i>	<i>Karin Östensson</i>
09.50	Hypothesis: Inflammation is a healthy part of successful stage II lactogenesis in women	<i>Michelle McGuire</i>
10.35	Coffee, poster walk	
11.00	Towards a clarification of the concept "mastitis" as used in empirical studies of breast inflammation during lactation	<i>Linda J Kvist</i>
11.15	Natural variation in biomarkers indicating mastitis in healthy cows	<i>Maria Åkerstedt</i>
11.30	Panel discussion	
12.15	LUNCH	
<b>SESSION 4</b> (Moderator Karin Persson Waller)		
13.30	Culture independent analysis of the bacterial communities in milk from women	<i>Mark McGuire</i>
14.20	Etiology and antimicrobial susceptibility of udder pathogens from cases of subclinical mastitis in dairy cows in Sweden	<i>Ylva Persson</i>
14.35	Long-time persistence of <i>Staphylococcus aureus</i> in a dairy herd with mastitis problems	<i>Karin Artursson</i>
14.50	Coffee, poster walk	
15.10	Panel discussion	
15.45	Conclusions from session 3 and 4	<i>Sigrid Agenäs</i>
16.00	CLOSING (end of programme 16.15)	<i>Sigrid Agenäs</i>

## **BREASTFEEDING POLICIES AND RECENT FINDINGS ON THE EFFECTS OF EARLY SUCKING BEHAVIOR ON LACTATION SUCCESS IN WOMEN**

Elisabeth Kylberg✉

School of Life Sciences, University of Skövde, Box 408, 541 28 Skövde, Sweden

✉Elisabeth.Kylberg@his.se

There is a conception of breastfeeding as a very natural phenomenon – and indeed it is. Many things have though contributed to turn breastfeeding into a complicated act, which cannot just be taken for granted.

Lactation is biological and breastfeeding is cultural.

Our western culture has managed to make breastfeeding a medical, unnatural phenomenon without status. Our culture succeeded in giving bottle feeding status and spreading it all over the world with the result that today UNICEF estimate that 1.3 million infants die every year by absence of breastfeeding.

Since the 70's WHO has worked to protect, support and promote breastfeeding in the scope of breastfeeding as the optimal way for our infants to be fed. Absence of breastfeeding implies for the infant an increased risk for getting otitis, gastroenteritis, upper respiratory infections, atopic dermatitis, asthma, obesity, type 1 and 2 diabetes, leukaemia, SIDS, necrotising enterocolitis. Absence of breastfeeding implies for the mother an increased risk for getting early post partum depression, type 2 diabetes, breastcancer and ovarian cancer (Ip *et al.*, 2009).

In 1981 the International Code for Marketing of Breastmilk Substitute was launched in order to protect breastfeeding by preventing unethical marketing of breastmilk substitute.

In 1991 the global initiative on Breastfeeding Friendly Hospital Initiative (BFHI) was introduced by UNICEF and WHO. This initiative aims to put breastfeeding friendly routines at the maternity wards into practice all over the world. The initiative is based on Ten Steps for successful breastfeeding. The wards should follow these ten steps in order to be assessed as a baby friendly hospital. In Sweden this initiative was launched in 1992 by the Ministry of Social Health and Welfare and in 1996 all hospitals were evaluated and assessed as baby friendly. In 1997 the National Institute of Public Health led the work in reevaluating the hospitals and to expand the initiative to include the primary health care. 33 out of 52 hospitals were reevaluated and assessed as baby friendly and primary health care (antenatal care and child health care) in 8 regions in Sweden were assessed as baby friendly. Since 2004 no national body has the responsibility of the BFHI – it is up to each region to work with this. In practice knowledge of good breastfeeding support is disappearing and low status is given to breastfeeding.

There is evidence that early initiation of breastfeeding is of importance for success in the continuing breastfeeding. One of the ten steps is formulated in order to facilitate early initiation of breastfeeding: “Encourage and support mothers to initiate breastfeeding during the first awakening period, which usually lasts for 2 hours.” The formulation of this step is based on findings from the research made by a research team from Karolinska Institutet lead by Ann-Marie Widström. The research made by Ann-Marie Widström and her team has focused on the effects of having the new born baby skin-to-skin with the mother. The most recent findings of her research have given an

insight of the behavior of the newborn infant when not interfering with the mother-infant dyad – just to let them be and videotaping has caught what is happening (Widström *et al.*, 2010).

The newborn term infant goes through nine behavioural phases: birth cry, relaxation, awakening, activity, crawling, resting, familiarization, suckling, and sleeping when being skin-to-skin with its mother. It is hypothesized that when the infant has got the possibility to go through all these nine phases peacefully this will result in early optimal self-regulation and be able to communicate its needs.

Unfortunately the routines at many delivery wards in Sweden does not give the newborn baby this opportunity – the first skin-to-skin contact will be interrupted by different routines which includes a separation of the mother and the newborn infant for weighing the infant, mother taking a shower and so on.

Moore *et al.* have done a Cochrane review (last updated 2007) on ‘Early skin-to-skin contact for mothers and their healthy newborn infants’. The review is based on 30 studies and they concluded that if the baby and mother had early skin-to-skin contact, babies interacted more with their mothers, stayed warmer, and cried less. Babies were more likely to be breastfed, and to breastfeed for longer, if they had early skin-to-skin contact. Babies were also, possibly, more likely to have a good early relationship with their mothers, even if this was difficult to measure.

Puig and Sguassero (WHO Reproductive Health Library, last revised 2007) conclude in their report on ‘Early skin-to-skin contact for mothers and their healthy newborn infants’ that 16% of neonatal deaths could be avoided if all infants were breastfed from day 1 and 22 % if breastfeeding started within the first hour.

In conclusion there is evidence for promoting early skin-to-skin contact between mother and her newborn infant in facilitating initiation of breastfeeding and thus promoting health of infants and mothers. More research is needed, preferably well-conducted randomized controlled studies RCTs to demonstrate the real impact of early skin-to-skin contact on maternal and infant health, including preterm babies and mothers who deliver by caesarean section in different settings.

## References

- Bystrova K, Ivanova V, Edhborg M, Matthiesen AS, Ransjö-Arvidson AB, Mukhamedrakhimov R, Uvnäs-Moberg K, Widström AM. Early contact versus separation: effects on mother-infant interaction one year later. *Birth*. 2009 Jun;36(2):97-109.
- Bystrova K, Matthiesen AS, Vorontsov I, Widström AM, Ransjö-Arvidson AB, Uvnäs-Moberg K. Maternal axillar and breast temperature after giving birth: effects of delivery ward practices and relation to infant temperature. *Birth*. 2007 Dec;34(4):291-300.
- Ip S, Chung M, Raman G, Chew P, Magula N, DeVine D, Trikalinos T, Lau J. Breastfeeding and maternal and infant health outcomes in developed countries. *Evid Rep Technol Assess (Full Rep)*. 2007 Apr;(153):1-186.
- Moore ER, Anderson GC, Bergman N. Early skin-to-skin contact for mothers and their healthy newborn infants. *Cochrane Database of Systematic Reviews* 2007, Issue 3.
- Puig G, Sguassero Y. Early skin-to-skin contact for mothers and their healthy newborn infants: RHL practical aspects, last revised: 9 November 2007 The WHO Reproductive Health Library; Geneva: World Health Organization.
- Widström AM, Lilja G, Aaltomaa-Michalias P, Dahllöf A, Lintula M, Nissen E. Newborn behaviour to locate the breast when skin-to-skin: a possible method for enabling early self-regulation. *Acta Paediatr*. 2010 Aug 16.



# POSITIVE EMOTIONS AND SUCCESSFUL LACTATION IN THE NEONATAL UNIT: A QUALITATIVE STUDY

Vivien Swanson<sup>1</sup>✉ & Rhona McInnes<sup>2</sup>

<sup>1</sup>*Department of Psychology, University of Stirling, Stirling FK9 4LA;*

<sup>2</sup>*Department of Nursing and Midwifery, University of Stirling, Stirling FK9 4LA*

✉[vivien.swanson@stir.ac.uk](mailto:vivien.swanson@stir.ac.uk)

## Introduction

In Scotland, most mothers of pre-term babies express breastmilk, but few successfully establish breastfeeding in the neonatal unit (NNU). Negative emotions make establishing lactation difficult, but how positive emotions facilitate lactation in a stressful situation is unclear. This qualitative study investigated reasons for negative and positive emotions related to expressing breastmilk and breastfeeding and maternal self-confidence in a Scottish NNU.

## Material and Methods

Nineteen primiparous women with babies in NNU special (intermediate) care were interviewed, exploring behavioural experiences and perspectives regarding expressing breastmilk and breastfeeding. Responses were transcribed and coded according to self-efficacy theory and emergent themes.

## Results

All participants experienced negative emotions related to premature birth, and worries about their baby. Difficulty expressing breastmilk and perceived poor milk supply were related to anxiety, embarrassment or guilt. Women were less likely to persevere in expressing breastmilk and breastfeeding where they experienced negative emotions and negative feedback. Successfully expressing breastmilk and satisfaction with milk supply were linked with positive mood and mastery, more pleasurable feeding, improved maternal self-concept and perseverance in the face of difficulty. Perceived positive feedback from their baby, partner and nursing staff was related to increased confidence, wellbeing and lactation success.

## Discussion and Conclusions

This qualitative study is novel in considering reasons for negative and positive emotions in mothers of children in NNU care. It supports previous findings regarding strong links between positive emotions and successful lactation, and suggests that providing more positive feedback to mothers around early lactation attempts might counteract negative emotions around premature birth, increasing lactation success.

# A PROCESS-ORIENTED BREASTFEEDING TRAINING PROGRAM FOR HEALTHCARE PROFESSIONALS TO PROMOTE BREASTFEEDING

Anette Ekström<sup>1</sup>✉, Elisabeth Kylberg<sup>1</sup> & Eva Nissen<sup>1,2</sup>

<sup>1</sup> School of Life Sciences, University of Skövde, Skövde, Sweden

<sup>2</sup> Department of Woman and Child Health, Division of Reproductive and Perinatal Health Care, Karolinska Institutet, Stockholm, Sweden

✉Anette.Ekstrom@his.se

## Introduction

The impact of giving an infant food other than breast milk depends on several factors. Evidence to date supports the recommendation for exclusive breastfeeding for six months.

## Aim

The Aim of the this study was to evaluate the effect of a process-oriented training in support during childbirth and breastfeeding for midwives and postnatal nurses in relation to the time of initial breastfeeding session, introduction of breast milk substitute and solids effects on the duration of breastfeeding.

## Materials and methods

Ten municipalities in Sweden were randomized to either intervention (IG) or control groups (CGA and CGB). The intervention included a process-oriented training program for midwives and postnatal nurses in the intervention municipalities. Primiparas (n=540) who were living at either site were asked to respond to questionnaires at three days, three and nine months postpartum. Data collection for mothers in CGA (n=162) started before effects of the intervention could be studied, CGB (n=172) was collected simultaneously with the IG (n=206).

## Results

Preliminary results showed that fewer infants in the IG received breast milk substitute (the first week of life) without medical reasons (p=0.01) and were older (3.8 months) when breast milk substitute was introduced after discharge compared with the infants in the control groups (CGA 2.3 months p= 0.01 and CGB 2.5 months p= 0.03).

## Discussion and Conclusion

A process-oriented training program for midwives and postnatal nurses (by changed attitudes among health staff and changing mothers self imaging) reduced the number of infants who got breastmilk substitute during the first week without medical reasons and delayed the introduction of breast milk substitute after the first week.

**\*Definition of processoriented training program:** In order to change health professionals attitudes the training program was based on literature reviews and collegial discussions containing professional stance, reflective processes, problem-solving processes, and practical skills in relation to support during childbirth and breastfeeding. The health professionals were trained in problem-solving, reflection, decision-making in terms of competence and personal qualifications to ensure that they should be ready to meet the demands of their future profession.

Jerlock M, Falk K, Severinsson E. Academic nursing education guidelines: tool for bridging the gap between theory, research and practice. *Nurs Health Sci* 2003;5:219-228.

# LONG CHAIN POLYUNSATURATED N-3 FATTY ACIDS (PUFA) IN COLOSTRUM ARE AFFECTED BY FISH INTAKE DURING PREGNANCY

Beheshteh Olang<sup>1, 2, 3, 4</sup>✉, Majid Haji Faraji<sup>2</sup>, Sophie Hellstrand<sup>1</sup>, Mohammad Palesh<sup>1, 4</sup>, Ebrahim Azadnya<sup>2</sup>, Zinat Kamali<sup>2</sup>, Mohamed Ali<sup>1</sup>, Birgitta Strandvik<sup>1</sup> & Agneta Yngve<sup>1, 3</sup>

<sup>1</sup>Unit for Public Health Nutrition, Department of Biosciences and Nutrition, Karolinska Institutet, Stockholm, Sweden

<sup>2</sup>National Nutrition and Food Technology Research Institute, Tehran, Iran

<sup>3</sup>Akershus University College, Lillestrøm, Norway

<sup>4</sup>Kermanshah University of Medical Sciences, Kermanshah, Iran

✉Beheshteh.Olang@ki.se

## Introduction

During pregnancy and lactation, the intake of n-3 PUFA in fish and seafood may improve maternal and infants' health outcomes.

## Aim

To estimate essential fatty acid concentrations in breast milk in relation to high and low fish diet in two provinces of Iran.

## Materials and Methods

Breast milk was collected directly after delivery from 120 randomly selected mothers, 60 in each province. The mothers were interviewed to complete a FFQ. The fatty acid composition in the breast milk was measured with gas-liquid chromatography (GLC).

## Results

Mothers in coastland (Guilan) had significantly higher intake of fish or seafood during pregnancy than the mothers in inland (Kermanshah). High fish and seafood intake were associated with significantly higher docosahexaenoic acid (DHA) concentration and lower (arachidonic acid) AA/DHA ratio in early breast milk.

There were no significant differences in the total PUFA status and in linoleic and  $\alpha$ -linolenic acids concentrations between the two provinces. Total n-3 fatty acids and DHA in breast milk were significantly higher in the Guilan compared to the Kermanshah province, but total n-6 fatty acids and AA did not differ. The ratios of total n-6/n-3 fatty acids and AA/DHA in breast milk of the mothers from Guilan were significantly lower than those found in mother's milk from Kermanshah. Consumption of saturated fat was higher in Kermanshah and olive and olive oil intake was significantly higher in Guilan.

## Conclusion

In order to satisfy the infant's need for a balanced intake of n-6/n-3 fatty acids, the present study points at the importance for pregnant and lactating mothers to eat fish and seafood at least once a week.

## RECENT HOT TOPICS IN HUMAN AND BOVINE LACTATION INCLUDING HIGHLIGHTS FROM THE ISRHML<sup>1</sup> MEETING

Mark A. McGuire<sup>1</sup>✉ & Michelle K. McGuire<sup>2</sup>

<sup>1</sup>*Department of Animal & Veterinary Sciences, University of Idaho, Moscow, USA*

<sup>2</sup>*School of Biological Sciences, Washington State University, Pullman, USA*

✉ [mmcguire@uidaho.edu](mailto:mmcguire@uidaho.edu)

Research presented at recent human and animal lactation meetings has offered some interesting thoughts regarding the biology and strategies for successful lactation.

### **Serotonin Regulation of Lactation**

Work by Dr. Nelson Horseman at the University of Cincinnati has demonstrated that serotonin is important in the process of initiation and cessation of lactation through regulation of tight junctions. An important regulatory component of the serotonin mechanism is the presence of a recycling mechanism through the serotonin transporter (SERT). This transporter is a target of pharmaceuticals, known as selective serotonin reuptake inhibitors (SSRI). This class of drugs is commonly used to treat depression, including that occurring in the postpartum period. Horseman and his collaborators (*J Clin Endo Metab*, 2010, 95:837-846) demonstrated that women taking SSRI were more likely to experience delayed secretory activation (stage II lactogenesis). The serotonergic signaling system is also present in dairy cows (see work from Dr. R.J. Collier, University of Arizona, e.g., *J Endo*, 2009, 203:123-131) but significant differences exist compared with human and mouse mammary epithelial cells. It appears that the serotonergic system is directly involved in milk protein gene expression in the bovine. The potential serotonergic regulation of mammary gland function may be expanded beyond tight junctions to include cell turnover, the milk ejection reflex and mammary blood flow.

### **Continuous Lactation**

For the dairy industry in the United States, the 305 d lactation and 60 d dry period were adopted during World War II to provide maximum production and accelerate genetic progress during a time of food shortage. Since that time, the dairy industry has improved milk production by 5,000 kg per lactation on the average dairy, placed a larger emphasis on profit, and enhanced genetic progress by artificial insemination and embryo transfer. These changes have led to reevaluation of dry period length and impact of delaying breeding, but evaluation of the optimal dry period has received little attention. Previous work evaluating continuous lactation or no dry period was done in the mid-1960s on cows reaching peak milk yields of 18-27 kg/d, compared to today's genetically superior, high-producing cow that reaches peak milk yields greater than 45 kg/d. Work in the 1960s demonstrated production losses of 18-29% in the lactation following an omitted dry period (Swanson, 1965, Smith et al., 1967, Swanson et al., 1967). Furthermore, they concluded that nutrition, hormonal regulation of lactation, and mammary cell numbers were not the causative factors of the reduced milk production. Reduced functionality of mammary cells was the favored hypothesis following this research. Analysis of dairy records determined that a dry period of 60 d is optimal and less than 40 d results in production losses (Remond, 1997). Many of these studies analyzed records from farms managing for a 60 d dry period, so why were cows getting a shortened or omitted dry period? Missed calving dates, twinning, and mismanagement are a few reasons why

---

<sup>1</sup> International Society for Research in Human Milk and Lactation ([www.isrhml.org](http://www.isrhml.org))

these cows did not have an optimal dry period, all of which unfavorably bias the results of a dry period less than 40 d. Recently, Bachman (2002) evaluated a shortened dry period in a study conducted in Florida and concluded a 30 d dry period was equivalent to a 60 d dry period in subsequent milk production.

In addition to improved genetics, there have been many management technologies that were introduced since World War II that improve milk yield, some of which address the issue of reduced functionality in mammary cells without a rest period. Among these technologies are total mixed ration, increased milking frequency, bovine somatotropin (bST), and photoperiod management. Bovine somatotropin (bST) is of particular interest because it increases milk yield by 10-15% and has been hypothesized to impact mammary cells by all or a combination of the following mechanisms: improved synthetic activity on a per cell basis, more cells in a secretory state and fewer resting, and reduced cell loss (Bauman and Vernon, 1993). Could the use of bST eliminate the need for a dry period by enhancing mammary synthetic activity and reducing mammary cell loss?

All of the aforementioned genetic and management improvements and the resulting production increases provide new opportunities for an omitted dry period in today's dairy cow. The first opportunity is improved milk yield at the time of dry off. Many cows are producing more than 22 kg/d at dry off and have the potential to milk through the last 60 d of gestation at a profitable production level. Another opportunity exists at the other end of the lactation curve in the fresh period. Continuously milking cows and eliminating dramatic dietary and physiological changes in the transition from pregnant and dry to lactating should improve feed intake and result in fewer metabolic problems at calving. Studies have demonstrated the potential for continuous lactation in multiparous cows but potential negative impacts in primiparous cows. Shortened or omitted dry periods may impede mammary growth in primiparous cows resulting in reduced milk yield in the subsequent lactation. In contrast, a shortened or omitted dry period with either bST continuously or reinitiation the 9<sup>th</sup> week after calving did not alter production in multiparous cows treated with bST.

In work from Peru by Dr. Grace Marquis (Pediatrics, 2002, 109:e56; J Nutr, 2003, 133:2585-2591), an overlap of lactation and late pregnancy in women had negative consequences on infant growth. Infants had reduced milk intake and milk IgA concentrations were reduced in women who nursed continually. Mothers that did nurse continually, though, did not report any mastitis while it was rare in the group that stopped nursing during late pregnancy.

# TIMING OF MAMMARY ANGIOGENESIS: A POSSIBLE EXPLANATION FOR *POST-PARTUM* APOPTOSIS IN THE BOVINE UDDER?

Sigrid Agenäs<sup>1</sup>, Mette Olaf Nielsen<sup>2</sup>, Sina Safayi<sup>2</sup> and Christopher H Knight<sup>2</sup>✉

<sup>1</sup>SLU Animal Nutrition and Management, 753 23 Uppsala, Sweden

<sup>2</sup>University of Copenhagen Faculty of Life Sciences, DK1870 Frb C Denmark

✉chkn@life.ku.dk

## Introduction

Observations of mammary secretory cell apoptosis occurring early *post partum* have been reported by several groups. It is somewhat counter-intuitive that secretory cells should proliferate extensively during pregnancy but then die without secreting milk. We sought an explanation for this phenomenon.

## Aims

The aim of the project was to compare the timing of mammary development (secretory cell proliferation and differentiation) and angiogenesis in bovine udder during pregnancy and lactation.

## Methods

Mammary biopsies were obtained from 44 cows managed either for 15 month lactation cycles or continuous lactation (no intentional dry period). Biopsies were analysed for expression of 31 genes involved in mammary development and angiogenesis using real-time reverse transcription polymerase-chain reaction (rtPCR).

## Results

Genes involved in mammary cell proliferation were up-regulated during pregnancy, whereas genes involved in secretory processes were up-regulated *post partum*. Genes involved in angiogenesis were up-regulated at the same time as the secretory genes, ie after the main phase of mammary growth.

## Conclusions

On the basis of these data we can propose that mammary growth and angiogenesis are not well coordinated in time. We observed apoptosis-associated changes during early lactation. A possible explanation is that some of the secretory cells that proliferate during late pregnancy do not receive adequate blood supply *post partum* as a consequence of the relatively late occurrence of angiogenesis. Further study of the factors controlling angiogenesis in the bovine udder is warranted, as it may enable early-lactation apoptosis to be avoided. This would be expected to increase milk production potential.

# DAY-TO-DAY VARIATION IN MILK COMPOSITION AND MILK YIELD AT UDDER QUARTER LEVEL

Linda Forsbäck<sup>1</sup>✉, Helena Lindmark-Månsson<sup>3</sup>, Anders Andrén<sup>2</sup>, Maria Åkerstedt<sup>1,2</sup>, Lisa Andréé<sup>1</sup> & Kerstin Svennersten-Sjaunja<sup>1</sup>

<sup>1</sup>Swedish University of Agricultural Sciences, Department of Animal Nutrition and Management, Uppsala, Sweden

<sup>2</sup>Swedish Dairy Association, Lund, Sweden

<sup>3</sup>Swedish University of Agricultural Sciences, Department of Food Science, Uppsala, Sweden

✉linda.forsback@huv.slu.se

## Introduction

The growing herd size in dairy production requires reliable tools for management of the cows. Automatic in-line sampling and measurement of milk composition is one alternative for monitoring milk quality and health disturbances. Data on milk components and milk yield provide information on milk quality alterations and cow health status, but are also useful in planning feeding and breeding. In automatic milking systems udder quarters are milked individually, enabling analysis and recording at udder quarter level. Earlier studies have described the day-to-day variations in milk components for cow composite milk, but there is a lack of studies performed at udder quarter level.

## Aim

The aim of this investigation was to study day-to-day variations in milk composition and milk yield at udder quarter and cow composite level in clinically healthy cows over a three week period.

## Material and Methods

Udder quarter and cow composite milk samples were collected from 42 consecutive milkings of 10 cows during 21 days. Milk yield was recorded and the milk was analyzed for total protein, whey protein, casein, fat, lactose and somatic cell count (SCC).

## Results and Conclusion

The results showed similar mean values and day-to-day variation for milk composition between healthy udder quarters within cow. Different milk components had different day-to-day variation, the lowest and highest variation being found in lactose (0.9%) and fat (7.7%), respectively. This suggests that repeated milk sampling and analysis at udder quarter level can be used to detect alterations in composition and cow health, and would thus be helpful in the management of the dairy herd.

# TRUTH AND MYTH ABOUT MILK PRODUCTION IN THE DEHYDRATED CAMEL (*Camelus dromedarius*)

Kristina Dahlborn✉, Tafesse Bekele & Kerstin Olsson

*Department of Anatomy, Physiology and Biochemistry, Swedish University of Agricultural Sciences, SE-750 07 Uppsala, Sweden*

✉[kristina.dahlborn@afb.slu.se](mailto:kristina.dahlborn@afb.slu.se)

## Introduction

Camels are known to endure long periods without drinking water and yet continue to produce milk. In addition, camels are reported to dilute their milk during water deprivation, which would be of great importance during drought because calf and people share the milk. Dilution of milk in dehydrated camels implies that their mammary gland has a unique morphology and physiology. It would contradict the generally accepted view that milk osmolality is kept iso-osmotic to blood plasma osmolality.

## Aim

To investigate if water deprived camels dilute the milk

## Materials and Methods

Seven camels were hand milked twice daily and subjected to four watering regimes in a Latin square design. Each regime lasted 16 days with five days of daily watering between periods: Water was offered at 1315 h once daily (W1), every 4<sup>th</sup> day (W4), on d 8 and 16 (W8), and on d 16 (W16). Camels were offered 6 kg of concentrate daily and hay *ad libitum*. A blood sample was taken at 1300 h d 1, 4, 8, 12, and 16.

## Results

A small reduction of milk volume occurred during the 2<sup>nd</sup> week of W16. Milk osmolality increased during water deprivation and dropped immediately after watering. During W16, osmolality increased from  $318 \pm 3$  mosm/kg to  $348 \pm 3$  mosm/kg, which was linearly correlated to the rise in plasma osmolality.

## Discussion

The water exchange mechanisms between milk and blood plasma in the mammary gland are the same in camels as in lactating ruminants.

## Conclusion

Camels can not dilute the milk, this wide spread idea is a myth.



# CHEMOTACTIC FACTORS INVOLVED IN INFLAMMATION OF THE MAMMARY GLAND WITH FOCUS ON THE BOVINE

## *CAN COMMON MILK CONSTITUENTS HAVE A CHEMOTACTIC EFFECT?*

Karin Östensson

*Department of Clinical Sciences, Division of Reproduction, Swedish University of Agricultural Sciences, Uppsala, Sweden.*

✉[karin.ostensson@kv.slu.se](mailto:karin.ostensson@kv.slu.se)

### **Introduction**

Mastitis (clinical and subclinical) in cows constitutes a significant animal health problem associated with animal welfare aspects and substantial economic losses for the producer. Mastitis is the most common disease in dairy cows and represents a major part of the antibiotic used for treatment of cows, additionally, often with a low efficiency in terms of complete recovery rate. In the light of the antibiotic resistance development there is an urgent need for a strict and sensible use of antibiotics and it has become increasingly important to prevent mastitis and to find therapeutic alternatives or complements to antibiotic treatment of clinical mastitis cases. Much research has for several decades focused on the details of initiation, mediation and shutting down the inflammatory reaction in the udder with the aim to find immunological factors that could be used in mastitis therapy to support the cows own defence. There is a great amount of research performed on possible immunomodulatory effects of common nutrient components of cow milk, with focus on the health of the human consumers but this field has been poorly investigated in relation to mastitis.

Subclinical, silent, mastitis constitutes the most severe problem in the mastitis complex. The increased milk somatic cell count (SCC) during mastitis is largely attributable to polymorphonuclear leukocytes (PMN) recruited to the mammary gland to combat the insult. Episodes of elevated SCC may also occur without any detectable reason or without any obvious inflammatory challenge (e.g. related to different milking interval length). They are usually of short duration and it has been questioned if they may be attributable to normal milk constituents with a chemotactic effect. Along with more detailed and frequent on farm automatic recording of milk characteristics and an increasing awareness that also a modest increase in SCC is associated with reduced milk yield, deteriorated milk quality and additionally might indicate the start of a more prominent mastitic reaction – it has become more relevant for the farmer to know how to handle short-term and small changes in SCC.

The aim of this paper is to highlight common milk constituents, as possible factors, directly or indirectly influencing the migration of PMN in the udder and thereby contributing to increased milk SCC of cows.

### **Chemotactic proinflammatory cytokines in bovine mastitis**

Main proinflammatory cytokines known to be involved in the mastitis reaction of the cow are the interleukins (IL) IL-1, IL-2, IL-6, IL-8 and IL-12; tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ), interferon- $\gamma$  and granulocyte-monocyte-colony stimulating factor (GM-CSF). The response of different cytokines may vary depending on the challenging microorganism. Based on extensive research, there is a common agreement that PMN and their phagocytosis represent a key factor in the cows' defense against intramammary infection. A fast and prominent recruitment of PMN to the udder is significant for the outcome of the mastitis. The complex inflammatory reaction involves interactions between leukocytes, tissue cells and cytokines with redundant immunological

functions. Also cytokines without direct chemotactic effect on PMN might indirectly influence their migration through activating cells to produce chemokines. IL-1, IL-8 and TNF- $\alpha$  are considered to be most important in the innate immune response in the udder with significant effect on PMN recruitment. However, experimental intramammary infusion of several of the other cytokines identified in mastitis, have been shown to result in a recruitment of PMN to milk.

### **Milk components with a chemotactic effect**

Cow milk contains a variety of components that can regulate immune functions, in non-ruminant as well as ruminant species. This has attracted interest from different scientific angles. The importance of milk to provide humoral and cellular immunological factors for development of the immune system of the newborn calf has for long been obvious. These factors have also been extensively studied in mastitis research. What is considered as common milk nutrient components, especially proteins, and hormones that are naturally present in milk have also been shown to exhibit immunomodulatory effects in a broad sense. Interestingly, the main part of the research in this area has concerned milk as a functional food and its effects on the human consumers' immune system and health. There is now a growing interest, addressing a possible effect of various milk constituents locally in the udder, on the immune system of the milk producing animal itself. In this context, a particular area of interest is, naturally, the apparently more "physiological" non-infectious episodes of temporarily increased SCC that occur in the cow udder and e.g. the involution process – and a possible interaction with e.g. the susceptibility of infections. Factors influencing chemotaxis of PMN are especially relevant considering a possible background to the "physiological" SCC reactions and because phagocytosis is a key factor in the defence against mastitis.

Milk constituents that have been investigated regarding their effect on PMN migration and/or other functions, and/or production of cytokines, in humans and a few animal species, are e.g. casein,  $\alpha$ -lactalbumin,  $\beta$ -lactoglobulin, lactoferrin, lactoperoxidase, whole whey protein fraction and prolactin. It should be noted, that there are examples that the immunomodulatory effect of purified whey fractions (like  $\alpha$ -lactalbumin,  $\beta$ -lactoglobulin, lactoferrin etc) appears to be reduced or eliminated by their combination in whey or when mixed with minor whey components. It has also been shown that when a single complex protein is fractionated (e.g. whole casein vs  $\beta$ -casein) often complementary biological effects become apparent, changing the immunological effect.

Bovine prolactin has been observed to stimulate PMN migration. In contrast,  $\alpha$ -lactalbumin appears to rather suppress chemotaxis which means that *decreased* concentration of  $\alpha$ -lactalbumin may result in enhanced PMN migration, due to a reduced inhibitory effect. These findings are interesting since increased concentration of prolactin and a drop in  $\alpha$ -lactalbumin has been observed in milk during episodes of elevated SCC and proportion of PMN, after a prolonged milking interval.

### **Conclusions**

Milk constituents, particularly in the protein fraction, can evidently influence chemotaxis of PMN and other immunological functions in e.g. humans. Immunomodulatory effects of such milk components have still not been proven in the bovine species but this field of research might be relevant to further explore related to mastitis and SCC reactions in cows.

# **HYPOTHESIS: INFLAMMATION IS A HEALTHY PART OF SUCCESSFUL STAGE II LACTOGENESIS IN WOMEN**

Michelle K. McGuire<sup>1</sup>✉ & Mark A. McGuire<sup>2</sup>

<sup>1</sup>*School of Biological Sciences, Washington State University, Pullman, USA*

<sup>2</sup>*Department of Animal & Veterinary Sciences, University of Idaho, Moscow, USA*

✉ [smcguire@wsu.edu](mailto:smcguire@wsu.edu)

## **Introduction and Dogma**

When used to describe events occurring during lactation, the term “inflammation” invariably refers to negative consequences such as pain, engorgement, and mastitis. Indeed, redness, elevated concentrations of inflammatory markers, and presence of immune cells in the milk are generally considered indicative of breast disease. Nonetheless, the similarities between the classic signs and symptoms of acute inflammation (heat, redness, swelling, and pain) and the situation experienced by healthy women at the initiation of copious milk production are striking.

## **Hypothesis**

It is possible that the processes by which lactation are initiated and sustained are driven, at least in part, by the same mechanisms intimately involved in acute inflammation, and we hypothesize that secretory activation (stage II lactogenesis) in women is caused to some extent by a healthy, homeorhetic shift in mammary inflammation which supports lactation.

## **Supporting Evidence**

In humans, there is very little mammary development prior to puberty. There is, however, some monthly menstrual-related development and more profound growth and development during pregnancy and early lactation. This cyclical growth and development appears to be related to inflammatory events. For instance, a relatively large literature provides evidence of systemic inflammation during the period immediately preceding ovulation (Baser et al., 2009; Becerik et al., 2010; Brenner et al., 2005) associated with elevated or decreasing progesterone levels (Mandane et al., 2009). This is also the period of time when women report transient soreness in their breasts – again suggesting local inflammation. Studies in cyclic, lactating goats (Peaker & Linzell, 1974) and ovulatory, lactating women (Hartmann & Prosser, 1982) suggest cyclical mammary inflammation, as evidenced by acute changes in milk composition during the preovulatory period. For instance, milk increases in sodium concentration and decreases in potassium levels during this time. This increased sodium:potassium ratio (a measure often used as a biomarker for subclinical human mastitis) is thought to be caused by opening of “leaky” tight junctions in the vascular epithelium as part of the acute inflammatory cascade. It is possible that this represents a homeorhetic shift in the inflammatory state which helps prepare the tissue for milk production should pregnancy ensue. Perhaps more importantly is what must occur for milk production after a neonate is delivered. Although the onset of stage II lactogenesis in women is known to require a drop in progesterone and adequate levels of prolactin, the physiologic trigger has yet to be identified (Neville and Morton, 2001). However, changes in milk composition during the first few days postpartum, including a profound influx of immune cells and their products, clearly indicate a state of local inflammation even in the healthiest of women. Limited evidence from laboratory and production species as well as women suggests that the local release of nitric oxide (NO) *in response to the physical shearing stress of suckling on the breast* might be this trigger (Kim & Wu, 2009; Pero et

al., 2006; Popeski & Woodside, 1994; Prosser et al., 1996). Indeed, milk NO content is very low in colostrum, high in transition milk, and then intermediate in mature milk (Hord et al., 2010). In addition, early milk NO content has been shown to be positively correlated to milk production during the first 10 d of life (Iizuka et al., 1997).

### **Possible Implications**

If acute release of NO by the mammary vasculature in response to suckling is involved in the onset and maintenance of lactation, then dysregulation of the NO system (and/or other local inflammatory events) may help explain many of the risk factors common to both delayed stage II lactogenesis and mastitis. For instance, it is possible that the chronic inflammation associated with obesity dysregulates acute inflammatory events in the breast. This might help explain increased risk for delayed stage II lactogenesis reported in overweight and obese women (Nohr et al., 2009; Rasmussen, 2007). Other risk factors common to delayed stage II lactogenesis and mastitis include nipple irregularities, inadequate suckling frequencies, and stress – all of which might be explained by a dysregulation of NO release and/or responsiveness (Dewey et al., 2003; Kvist, 2010). Further understanding of this putative mechanism might lead to development of diagnostic tools that could be used to identify women at especially high risk for lactation difficulties. Treatments (e.g., NO donors or degradation inhibitors) could also be developed to prevent and treat these problems.

### **Conclusion**

Researchers studying lactation might want to consider a paradigm shift in terms of how they think about the interplay between inflammation and mammary function. It is possible that increased, tightly orchestrated local inflammation is needed for successful lactation initiation and maintenance, but that too much or too little can lead to lactation problems such as delayed stage II lactogenesis, inadequate milk production, and/or mastitis. Rigorous documentation of the relationships among local inflammatory events (e.g., NO release), suckling characteristics, timing of stage II lactogenesis, milk production, and breast health is warranted.

# **TOWARDS A CLARIFICATION OF THE CONCEPT “MASTITIS” AS USED IN EMPIRICAL STUDIES OF BREAST INFLAMMATION DURING LACTATION.**

2010, *Journal of Human Lactation*, Feb;26(1):53-9. Review

Linda J. Kvist

*Department of Obstetrics & Gynaecology, Helsingborg Hospital, Helsingborg, 251 87 Sweden and Division of Nursing, Department of Health Sciences, Lund University, Lund, Sweden.*

✉ Linda.Kvist@skane.se

## **Introduction**

The objective was to clarify the use of the term mastitis in contemporary empirical studies of lactational mastitis in humans. Concepts are the building blocks of theory and development of theory is reliant on an understanding of the concepts that are related to the phenomena of interest. Scandinavian researchers suggested in the 1980s that mastitis should be classified into three classes: milk stasis, non-infectious inflammation and infectious inflammation of the breast. A recent important award-winning paper from Australia questioned our understanding of mastitis and posed a tentative hypothesis that physiological responses in the breast during mastitis may cause symptoms similar to those caused by pathogens and recent research from Sweden, comparing bacterial content of breast milk in women with and without lactational mastitis has questioned the feasibility of dividing lactational mastitis into infectious and non-infectious groups. The reader of scientific articles is often required to have previous knowledge of the subject in question in order to understand the rationale for the research. Contained within this previous knowledge is often a subliminal supposition that the reader and the researcher share a common understanding of the concepts used in the article. Such assumptions may lead to misunderstanding and inability to consolidate research findings. New research may be changing how lactational mastitis is understood and therefore there is a need to clarify the current scientific use of the term mastitis.

## **Method**

A review of empirical studies on lactational mastitis was carried out. Studies were identified by means of literature searches in PubMed and the Cochrane Library. Limitations applied were that the articles should be written in English and published between 1998 and 2008 in peer-reviewed journals. Some 165 articles were retrieved. Many were concerned with animal mastitis, were in other languages than English, had no abstract, had HIV as the primary focus, had bacterial diversity of breast milk as primary focus or were reviews or case studies. Twenty-two empirical studies were primarily selected as suitable for critical appraisal for use of the term mastitis. When it was seen that more than one article was written by the same authors, checks were made as to whether the same study population was reported more than once. This was the case with four articles and these were then excluded since there was a clear probability that the concept of mastitis would be used in the same way in studies using the same populations. Eighteen articles published between 1998 and 2008 were critically analysed and a concept clarification of the term mastitis was carried out. In order to compare and contrast how the authors considered the term mastitis, questions were posed to the texts. These were as follows:

- In the study, who made the diagnosis of mastitis and was an operational definition used?
- Was lactational mastitis defined exclusively as inflammation?
- Was lactational mastitis defined exclusively as infection?
- Was it suggested that lactational mastitis may be either inflammation or infection?
- Were distinctions made between milk-stasis, non-infectious mastitis, and infectious mastitis?
- Was bacterial cultivation used?
- Was bacterial cultivation suggested as appropriate?
- Were leukocyte counts and/or CRP used?
- Were leukocyte counts suggested as appropriate?
- Which causes of lactational mastitis were suggested?
- Was the use of antibiotics suggested as appropriate treatment?

## **Results**

Mothers, doctors and healthcare providers made the diagnosis of mastitis in the included studies. Four articles suggested that mastitis was an infection, four made no mention of the aetiology of symptoms and ten articles described mastitis as an inflammation or an infection. Eight studies made no use of distinctions between milk-stasis, non-infectious mastitis, and infectious mastitis. Bacterial cultivation was used in five articles and one article measured leukocytes and CRP. The authors reported that CRP was not a useful test. Several different causes of mastitis were suggested. Antibiotics were suggested as appropriate treatment in four of the articles. A further four stated that antibiotics could be needed in some cases, one of which stated that the presence of methicillin resistant staphylococcus aureus (MRSA) was an indication for antibiotic therapy. The remaining ten articles made no suggestion for treatment by antibiotics.

## **Discussion**

A state of the art systematic review of studies on the use of antibiotics for lactational mastitis is available from the Cochrane Library and the authors clearly state that there is insufficient evidence to confirm or refute the effectiveness of antibiotic therapy for the treatment of mastitis during breastfeeding. Until evidence is available recommendation for antibiotic therapy as the first line treatment for mastitis should be considered carefully since problems related to unnecessary use of antibiotics are well known to all health care providers.

In 2001, an Australian researcher considered the fact that mothers with apparently non-infective mastitis (considered as such because of spontaneous resolution of symptoms without antibiotic therapy) present with symptoms suggesting systemic response, such as pyrexia, rigors and flu-like symptoms. These observations have recently been confirmed in a large study from Sweden which showed that 85 % of women with symptoms of mastitis recovered without antibiotics and with few cases of residual symptoms that required antibiotics.

## **Conclusions**

Use of the concept of mastitis in empirical studies remains inconsistent and this state of vagueness may in part be due to the disparate views of the causes of the symptoms of mastitis. The global health care community is in need of consensus on the definition of the term and the diagnosis of the condition because of the ever present threat of development of multi-resistant bacteriological strains. We need also to understand why some women who present with symptoms suggesting septic reaction recover without antibiotic therapy.

# NATURAL VARIATION IN BIOMARKERS INDICATING MASTITIS IN HEALTHY COWS

Maria Åkerstedt<sup>1,2</sup>✉, Linda Forsbäck<sup>1</sup>, Torben Larsen<sup>3</sup> & Kerstin Svennersten-Sjaunja<sup>1</sup>

<sup>1</sup> Department of Animal Nutrition and Management, Kungsängen Research Centre, Swedish University of Agricultural Sciences, SE-753 23 Uppsala, Sweden

<sup>2</sup> Department of Food Science, Swedish University of Agricultural Sciences, SE-750 07 Uppsala, Sweden

<sup>3</sup> Department of Animal Health and Bioscience, Faculty of Agricultural Sciences, Aarhus University, DK-8830 Tjele, Denmark

✉maria.akerstedt@lmv.slu.se

## Introduction

Dairy herds are expanding and with increasing numbers of animals in each herd there is a need for automatic detection of mastitis, for example by monitoring indicators in milk. A number of biomarkers for mastitis have been suggested over the years. In cows, mastitis usually occurs in one of the four udder quarters and since it is now possible to milk each udder quarter separately in automated milking systems, it is important to evaluate the normal variation in the biomarkers at udder quarter level.

## Aim

The aim of this study was to evaluate the normal variations between milkings for some biomarkers in clinically healthy cows in quarter as well as cow composite milk samples.

## Materials and Methods

The biomarkers studied were serum amyloid A (SAA), haptoglobin (Hp), lactate dehydrogenase (LDH), N-acetyl- $\beta$ -D-glucosaminidase (NAGase), and alkaline phosphatase (AP). Samples of udder quarter milk and representative composite milk samples were obtained from ten cows in 42 consecutive milking occasions, giving a total of 2100 individual milk samples.

## Results

Each cow had its individual profile for the concentrations and variations in the parameters analysed. Although there was relatively large variation between cows, ranging from 7-57% for the biomarkers analysed, the variation between milkings in clinically healthy quarters within cows was often below 10%. The biomarker with the lowest variation in this study was LDH.

## Discussion and Conclusion

The results suggest that by comparing quarters within an individual cow it may be possible to identify deviations from the natural variations between milkings. This could be a valuable tool instead of, or in combination with, a cut-off value for each parameter in order to detect changes in the milk indicating mastitis.

# CULTURE INDEPENDENT ANALYSIS OF THE BACTERIAL COMMUNITIES IN MILK FROM WOMEN

Mark A. McGuire<sup>1</sup>✉, Katherine M. Hunt<sup>1</sup> & Michelle K. McGuire<sup>2</sup>

<sup>1</sup>*Department of Animal & Veterinary Sciences, University of Idaho, Moscow, USA*

<sup>2</sup>*School of Biological Sciences, Washington State University, Pullman, USA*

✉ [mmcguire@uidaho.edu](mailto:mmcguire@uidaho.edu)

## Introduction

Human milk is considered universally to be the optimal source of nutrition for most healthy, full-term infants due to the considerable health benefits it confers upon the breastfeeding child. Importantly, lactation also positively impacts maternal health. For instance, breastfeeding reduces risk for some forms of breast cancer in women; protects against diarrheal and respiratory diseases in infants, especially in developing countries; and is associated with reduced long-term risk of obesity and diabetes in infants. Past research has extensively investigated the presence and health implications of the traditional nutrients in milk, such as lipids, carbohydrates, minerals, and vitamins. However, there are clearly other less well described health-promoting substances (such as pre- and probiotic factors) in human milk. For instance, these factors likely play a major role in determining an infant's gastrointestinal microbial community which is then maintained to a large extent through adulthood. This may be especially important, as research has shown a connection between bacterial communities in the adult gastrointestinal tract and the risk of obesity. As such, commensal bacteria found in human milk may serve as the initial inoculation of the neonatal gastrointestinal tract, thus substantially influencing lifelong health outcomes (such as risk of obesity). However, healthy milk as a potential source for these initial cultures is poorly studied. Culture dependent assessment of bacteria in healthy milk rarely attempts to cultivate any more than a few bacterial species due to the diversity of culture conditions required to grow a wide variety of species. Recent studies utilizing microbial characterization techniques based on the amplification of bacterial 16S rRNA and through PCR-denaturing gradient gel electrophoresis and quantitative real time PCR, has shown that aseptically collected human milk contains several genera of bacteria including *Staphylococcus*, *Streptococcus*, *Lactobacillus*, and *Bifidobacterium*. While these studies provide clear evidence that healthy human milk contains bacteria, very little work has examined milk bacterial community characteristics such as phylotype richness and relative abundance, or the stability of these communities within an individual over time. These types of analyses are critical to more fully characterize the human milk microbiome because they allow for subsequent investigations designed to determine if alterations in these communities play a role in maintaining mammary gland health, bacterial colonization of the infant's gastrointestinal tract, and other indices of short- and long-term maternal and infant health.

## Aim

The present study was designed to probe more deeply into the richness and diversity of human milk bacterial communities with minimal bias by taking advantage of the increased sequencing capacity available with high throughput 454 pyrosequencing methods.

## Materials and Methods

Milk samples were obtained aseptically from 16 lactating women self-described as healthy (free from lactational mastitis) at 3 different time points over a 4-wk interval (with the exception of one participant who donated only 2 samples). Using DNA extracted from each milk sample, the V1-V2



region of the gene encoding 16S ribosomal RNA was amplified using bar coded primers, and the resulting amplicons were sequenced using 454 pyrosequencing techniques.

## Results

After implementing quality control measures to remove reads with potential sequencing error, over 160,000 partial length sequences remained, averaging 3,500 operational taxonomic units (OTU)/ sample. The most commonly observed genera in milk in the 3 samples from each of the 16 subjects included *Streptococcus*, *Staphylococcus*, *Serratia* and *Corynebacteria*. The communities present were reasonably complex with 12 different genera representing an average relative abundance of  $\geq 1\%$ .

## Discussion

One of the original objectives of the National Institutes of Health's Human Microbiome Project was to determine if any "core" sets of microbes were common to particular body sites across healthy individuals. Analysis of our data set to determine which, if any, OTUs were present in every sample from every woman identified 9 such OTUs that were then classified into taxonomy. This small proportion of the overall membership of the milk microbiome represents a reasonably large (50%) proportion of the relative abundance in the total community in the 16 subjects. This small sample set therefore suggests a significant common core milk microbiome among all individuals, although a larger sample size will be required to determine if this core is universally present. It is noteworthy that the relative abundance of the individual core microbiome of each woman greatly exceeded the relative abundance of the overall core, providing more support that the host influences these communities. This implies that some phylotypes persisted in the milk of certain individuals and not in others.

Furthermore, results suggest that the particular host (in this case, each woman) appeared to experience her own dynamics in terms of the bacteria present in her milk. For example, in the samples from one subject, *Staphylococcus* was either the first or second most abundant genera in the milk representing 22-59% of the community. In contrast, *Staphylococcus* was only a minor contributor to the community in each sample (<5%) from another subject. These patterns suggest the potential for one or more characteristic(s) of the host to influence which phylotypes exist in the milk she produces. Also, in some individuals the milk bacterial communities are consistent and relatively unchanging over time, while in others there is little stability as community dynamics appear to shift dramatically over time.

## Conclusion

In summary, we have shown that milk bacterial communities are highly diverse and complex, and a logical next step will be to sample a larger more diverse population to determine which characteristics, if any, of the milk microbiome are associated with healthy outcomes for mothers and infants. Likewise, it is of interest to determine if factors such as maternal race, parity, mode of delivery, or maternal diet influence these community characteristics. Previous studies have shown that the microbiota present in the lower gastrointestinal tract, vagina and oral cavity, and more importantly the differential composition of these communities in healthy versus diseased states, are related to the health of the human host. The application of these principles to human milk has important implications for mammary health, bacterial colonization of the infant's gastrointestinal tract, and short- and long-term infant health. Determining the health consequences associated with variation or alteration in this community for both mother and infant presents a pertinent and urgent area of study.

# ETIOLOGY AND ANTIMICROBIAL SUSCEPTIBILITY OF UDDER PATHOGENS FROM CASES OF SUBCLINICAL MASTITIS IN DAIRY COWS IN SWEDEN

Ylva Persson<sup>✉</sup>, Ann Nyman & Ulrika Grönlund Andersson

*Dept of animal health and antimicrobial strategies, National Veterinary Institute, Uppsala*

✉Corresponding author: ylva.persson@sva.se

## Introduction

A nationwide survey on the microbial etiology of cases of subclinical mastitis in dairy cows was carried out on dairy farms in Sweden.

## Aim

The aim was to investigate the microbial panorama and the occurrence of acquired antimicrobial resistance. Moreover, differences between newly and chronically infected cows were investigated.

## Material and Methods

In total, 583 quarter milk samples were collected from 583 dairy cows (i.e. one quarter/cow) at 226 dairy farms from February 2008 to February 2009. The quarter milk samples were subjected to bacteriological investigation and scored according to the California Mastitis Test. Staphylococci were tested for betalactamase production and presence of acquired resistance was evaluated in all specific udder pathogens. Differences between newly infected cows and chronically infected cows were statistically investigated using logistic regression analysis.

## Results, discussion and conclusions

The most common isolates were *Staphylococcus (S) aureus* (19 %) and coagulase-negative staphylococci (CNS; 16 %) followed by *Streptococcus (Str) dysgalactiae* (9 %), *Streptococcus (Str) uberis* (8 %), *Escherichia coli* (2.9 %), and *Streptococcus* spp. (1.9 %). Samples with no growth or contamination constituted 22 % and 18 % of the diagnoses, respectively. The multivariable analysis showed that there was an increased risk of finding *S. aureus*, *Str. uberis* or *Str. dysgalactiae* in milk samples from chronically infected cows compared with milk samples from newly infected cows. Four percent of the *S. aureus* isolates and 35 % of the CNS isolates were resistant to penicillin by  $\beta$ -lactamase production. Overall, resistance to other antimicrobials than penicillin was uncommon.

# LONG-TIME PERSISTENCE OF *Staphylococcus aureus* IN A HERD WITH MASTITIS PROBLEMS

Karin Artursson<sup>✉</sup> & Karin Persson Waller

National Veterinary Institute, Uppsala, Sweden

✉karin.artursson@sva.se

## Introduction

The most commonly isolated bacteria in bovine mastitis in Sweden is *Staphylococcus aureus*. In a study performed in 2006-07 we showed that there usually is one strain of *S. aureus* predominating in milk at each farm. In 2010 we went back to one of the farms to repeat the milk sampling.

## Aim

The aims were to investigate if the same strain of *S. aureus* was still present in the herd and to study if *S. aureus* could be isolated repeatedly from individual cows and thus more likely correlate to intramammary infection.

## Material and methods

Quarter milk samples from 50 lactating cows, collected at three occasions within a period of 6 weeks in spring 2010, were bacteriologically examined. Officially registered udder disease scores, 0-9, based on somatic cell count were noted. A low score indicates no or low-grade inflammation.

## Results

*S. aureus* was isolated on one occasion or more from 31 cows. More than one third of the *S. aureus* positive cows had an udder disease score 0-2. In six cows *S. aureus* was isolated twice, three of these cows had an udder disease score 0-2. Six isolates from cows with repeated isolation of *S. aureus* were characterized by PFGE and were all shown to be of the same pulsotype as isolated in the herd in December 2006.

## Discussion

Can cows with low somatic cell counts act as reservoirs of *S. aureus*? This has to be further investigated.

## Conclusions

- The same strain of *S. aureus* still exists in this herd, continuously having problems with *S. aureus* mastitis.
- *S. aureus* can be isolated from cows with a low somatic cell count, sometimes repeatedly.

# **SURVIVAL AND GROWTH PERFORMANCE OF *Staphylococcus aureus* IN BEDDING MATERIAL**

Karin Artursson<sup>✉</sup> & Magnus Thelander

National Veterinary Institute, Uppsala, Sweden

<sup>✉</sup>karin.artursson@sva.se

## **Introduction**

*Staphylococcus aureus* is a common cause of bovine mastitis and can be found in infected milk, on body sites and in the environment.

## **Aim**

Gain more knowledge about *S. aureus* in the environment.

## **Material and methods**

1) The occurrence of *S. aureus* on the floor and on hocks were investigated in lactating cows in a herd with free stall housing. Samples were cultivated on Baird Parker agar and 5% bovine blood agar. 2) The survival time of *S. aureus* in an isolated, naturally infected cubicle was studied by sampling at different times. 3) Growth performance of *S. aureus* was studied at the laboratory in chopped wheat straw, peat and wood shavings. Samples were inoculated with *S. aureus* in ultra pasteurized milk, incubated for 12 hours and then diluted in peptone water, titrated and cultivated.

## **Results**

1) *S. aureus* was found in 3 of 20 floor samples and in 7 of 20 hock samples. 2) It was still possible to isolate *S. aureus* after 48h from the investigated isolated cubicle. 3) The in vitro growth of *S. aureus* was of the same magnitude in peat and wood shavings, but 0.5-1.5 log higher in straw.

## **Discussion**

The observed differences between growth performance in different bedding materials might be explained by a lower pH in peat (pH 3.5) and wood shavings (pH 4.9) compared to straw (pH 7.4).

## **Conclusions**

- *S. aureus* could be isolated from the floor and from hocks in a herd with mastitis problems and survived for at least 48 hours in bedding.
- *S. aureus* grew better in straw than in peat or wood shavings.

# **PREDICTION OF MASTITIS BASED ON MILK ELECTRO-CONDUCTIVITY, PH, AND ACUTE PHASE PROTEINS IN DIFFERENT DAIRY ANIMALS IN SUB-TROPICAL CLIMATE**

K. Behera, S. S. Layek, S, Prasad, A. Kumaresan, T. K. Mohanty✉

*Cattle Yard, LPM, National, Dairy Research Institute, Karnal – 132 001, Haryana, India Telephone +91-9215508002,*

✉ *mohanty.tushar@gmail.com*

## **Introduction**

Early mastitis detection is an ardent need to cater the exponentially growing Indian dairy sector.

## **Aim**

To establish milk electro-conductivity (EC) and pH as reliable markers for early mastitis detection in sub-tropical climate.

## **Materials and Methods**

Milk pH and electro-conductivity and their correlation with the plasma Acute Phase Proteins (Serum Amyloid A-SAA and Haptoglobin-Hp) were measured (Life Diagnostics Inc., USA) in 200 cows of each breed of Sahiwal, HF Crossbred and Murrah buffaloes.

## **Results**

The milk EC of healthy, subclinical and clinical mastitis in Sahiwal was  $5.25\pm 0.19$ ,  $7.13\pm 0.53$  and  $7.94\pm 0.91$  mS/cm in crossbred was  $5.88\pm 0.23$ ,  $6.96\pm 0.76$  and  $7.67\pm 1.13$  mS/cm and in buffaloes was  $5.44\pm 0.35$ ,  $6.42\pm 0.72$  and  $7.54\pm 1.02$  mS/cm, respectively. The milk pH of healthy animals was  $6.55\pm 0.19$ ,  $6.68\pm 0.23$  and  $6.64\pm 0.35$ , subclinical mastitis  $6.63\pm 0.27$ ,  $6.79\pm 0.34$  and  $6.78\pm 0.41$  and mastitis was  $6.93\pm 0.37$ ,  $7.17\pm 0.43$  and  $7.09\pm 0.29$ , in Sahiwal, Crossbred and Murrah respectively. The Hp and SAA levels in the subclinical, clinical and healthy cases were  $2.4\pm 0.77$ ,  $1.69\pm 0.94$  and  $0.17\pm 0.02$   $\mu$ g/l and  $1.427\pm 2.04$ ,  $2.58\pm 1.09$  and  $0.03\pm 0.04$   $\mu$ g/l, respectively. The correlation between Hp and EC was 0.87 and 0.94 and SAA and EC was 0.73 and 0.91 in subclinical and clinical cases. The sensitivity of EC in detecting subclinical mastitis in Sahiwal, crossbred and buffaloes was 83.2, 80.6 and 85.3%, whereas specificity was 74.1, 73.2 and 78% respectively.

## **Discussion**

Hp and SAA were very good predictors of clinical mastitis; however, in subclinical mastitis Hp proved to be better and highly correlated with EC. Higher Hp level in subclinical mastitis may be due to the chronic nature of the subclinical cases.

## **Conclusion**

EC with high sensitivity can be easily put into AMS for accurate prediction of subclinical mastitis.

# SEASONAL EFFECT ON MILK SOMATIC CELL COUNT (SCC) AND DIFFERENTIAL LEUKOCYTE COUNT (DLC) IN COWS AND BUFFALOES

Kalyan De<sup>✉</sup>, Joydip Mukherjee, Shiv Prasad, Ajay Kumar Dang

*National Dairy Research Institute, Karnal 132 001, Haryana, India*

✉*kalyande2007@gmail.com*

## Introduction

SCC is an important index for evaluating the incidence and prevalence of subclinical mastitis which is affected by environmental factors mainly temperature and humidity. Milk SCC along with DLC can be used as an effective index for estimating mammary health.

## Materials and methods

Milk was collected from 80 Karan Fries (KF) crossbred (Holstein Friesian x Tharparkar) cows and 35 Murrah buffaloes in the autumn (Max=30.6°C, Min=18.6°C, RH=76.6%) and winter (Max=21.5°C, Min=8.3°C, RH=66.6%). SCC and DLC were counted in a microscop at 40 X and 100 X respectively after staining with leishman stain. Milk SCC was expressed in total count and DLC were expressed in percentages.

## Results

Milk SCC were found to be higher ( $P<0.01$ ) in cows as compared to buffaloes during all the seasons. A higher milk SCC ( $P<0.01$ ) was found in autumn than the winter in both cows and buffaloes. In milk from crossbred cows neutrophil percentage increased ( $P<0.01$ ) in the autumn. Both cows and buffaloes showed a decrease ( $P<0.01$ ) in milk lymphocyte percentage in the winter.

## Discussion and Conclusion

It is likely that the increase in milk SCC and neutrophil percentage in autumn milk reflected the prevalence of pathogenic micro-organism in the environment when the temperature and humidity was high.

# COLOSTRUM QUALITY IN COWS MILKED PREPARTUM

Sabine Ferneborg<sup>✉</sup> & Cecilia Kronqvist

*Department of Animal Nutrition and Management, Swedish University of Agricultural Sciences, Kungsängen Research Centre, 753 23 Uppsala, Sweden*

<sup>✉</sup>[sabine.ferneborg@huv.slu.se](mailto:sabine.ferneborg@huv.slu.se)

## Introduction

Colostrum is essential for calf survival, providing the calf with important immunoglobulins. It is known that milk leakage prior to calving may impair colostrum quality. However, the effect of prepartum milking, which is sometimes used for improvement of calcium homeostasis, is poorly documented.

## Aim

The aim of the study was to investigate the effect of prepartum milking on colostrum quality measured as concentration of immunoglobulin G (IgG).

## Materials and Methods

Nine cows of the Swedish Red breed were milked during the last days prior to calving. Five cows served as controls and were only milked after calving. Both before and after calving cows were milked twice daily and milk samples were collected until the sixth milking postpartum.

## Results

Prepartum milked cows (PPM) were milked for 1 to 7 days before calving and milk yield the day before calving was on average 13 l (range 6-26 l). The concentration of IgG in milk was initially very high in the PPM group, but slowly decreased until calving. At the first milking after calving there was a tendency for lower IgG concentration in the PPM group than in the control group, but the level in the control group decreased rapidly and at the second milking there were no differences between the groups.

## Discussion

Prepartum milking reduced colostrum quality, but within 24 hours after calving, no differences remained between treatments. The colostrum quality deteriorates in pace with the decreasing uptake-window of the calf intestine.

## Conclusions

Prepartum milking reduced the quality (IgG content) of colostrum in the first milking after calving.

# EFFECT OF AN IMMUNOMODULATOR AGENT WITH ANTIBIOTIC THERAPY FOR THE TREATMENT OF COWS DURING THE DRY PERIOD – PRELIMINARY RESULTS

Ruben E Giannechini<sup>1,2</sup>✉, C Matto<sup>1</sup> & MS Gonzalez<sup>1</sup>

<sup>1</sup>Laboratory Veterinary Direction “Miguel C. Rubino”, Northwest Regional Laboratory, Paysandú, Uruguay.

<sup>2</sup>Institute of Microbiological Sciences, Veterinary Faculty, Regional North, UdelaR, Salto, Uruguay.

✉edgarnel@adinet.com.uy

## Introduction

Sub clinical mastitis (SCM) is the most prevalent form of bovine mastitis world-wide, in which no change in the milk is apparent, despite an important elevation of somatic cells count (SCC). Antibiotic cow therapy at drying off is an important tool to control the level of SCM cases.

## Aim

The aim of this assay was to study the effect of different doses of an immunomodulator agent to improve the antibiotic therapy at drying off.

## Material and Methods

This work was performed in a dairy farm located in Uruguay. One hundred cows were selected with SCC >200000 cell/ml almost in one quarter, positive udder pathogens isolation and history of high level of SCC during the last lactation. These were distributed in four groups, the control group received at drying off only antibiotic dry therapy. While, groups 1 to 3 received antibiotic and 5cc; 10cc and 15cc of a biologic immunomodulator, composed by lipopolysaccharide of *Escherichia coli* and cellular body of *Propionibacterium granulosum* (Inmodulen®), respectively. They received the same doses of Inmodulen® 20 days before calving and first day post partum. Milk quarters samples were taken between 7 to 10 days post partum to bacteriology and SCC. The results of SCC were compared by two-sample *t*-test, and the percentage of health quarter and health animal obtained post partum were analysed by test of two-sample proportions. The SCC data were log<sub>10</sub> transformed.

## Results

The SCC levels in the group 3 was significant lower respect to control group, obtained at post partum (5.233±0.735 vs. 4.907±0.652), also was statistically significant the differences in the percentage of health animal, 52% in the group 3, whereas only 27% in the control group.

## Discussion and conclusion

The immunomodulators are substances that augment immune responses, in udder involve activation of the innate system and recruitment of its defence mechanisms. The correct immuno response is necessary to complement the antibiotic effect, we demonstrated in this assay doses of 15 cc of Inmodulen® improve the effect of dry cow therapy.



# LACTATION RESEARCH IN FINLAND – RESULTS OF A ONE YEAR FOLLOW-UP STUDY

**Leena Hannula**

*Helsinki Metropolia University of Applied Sciences, Helsinki, Finland*

✉ [Leena.Hannula@metropolia.fi](mailto:Leena.Hannula@metropolia.fi)

## **Introduction**

Almost all (97 - 99 %) mothers initiate breastfeeding in maternity hospitals in Finland, but only 21 % breastfeed exclusively during the stay (2-3.5 days) in maternity hospitals and 60 % during the first month.

## **Aim**

The aim of this intervention study was to support exclusive breastfeeding and study factors related to the duration of exclusive breastfeeding.

## **Material and methods**

The intervention package included: 1) The “vauvankaa.fi” web pages to strengthen parents’ breastfeeding self-efficacy and offer professional and peer support for parenting and breastfeeding. 2) Midwives in the hospital were educated to promote skin - to - skin contact, parents’ breastfeeding self-efficacy and evaluate breastfeeding situations by using the LATCH - method. 3) Mothers at risk for early cessation of breastfeeding had access to a hospital breastfeeding clinic.

The mothers (n=705) were recruited from maternity hospitals (two intervention and one control hospital) in Southern Finland. The mothers were followed at 6 weeks, 6 months and 12 months postpartum. The data was analysed using multivariate statistical methods.

## **Results**

The mothers in the intervention group breastfed to a significantly larger extent compared to the mothers in the control group. At the follow up at 12 months postpartum the difference in the duration of exclusive breastfeeding between the groups was not statistically significant. Supplementary feeding in the hospital was linked to supplementary feeding at six weeks as well as breastfeeding problems.

## **Discussion and conclusions**

Interventions increased exclusive breastfeeding in the hospital, but there was no significant difference of the duration of exclusive breastfeeding after discharge. There is a need to develop better strategies to support exclusive breastfeeding during infancy.

# **MILK YIELD AND COMPOSITION IN SWEDISH LANDRACE GOATS (*Capra hircus*) KEPT TOGETHER WITH THEIR KIDS IN TWO DIFFERENT SYSTEMS**

Madeleine Högberg<sup>✉</sup> & Kristina Dahlborn

*Dept of anatomy, physiology and biochemistry. Box 7011. 750 07 Uppsala. Sweden.*

<sup>✉</sup>a05mahol@stud.slu.se

## **Introduction**

Swedish goats are mainly held for cheese production and the milk composition is therefore of great importance.

## **Aim**

The aim was to investigate how milk yield and composition were affected when kids suckled their dams for 8 weeks.

## **Materials and methods**

Eleven lactating goats were kept in two different systems and were machine milked twice daily. Group 1 was separated daily from their kid between 7:30 and 15:00 and they were allowed to suckle before each milking. In group 2 the dam and kid were kept together for 24hours but the kid was only allowed to suckle one teat as the other teat was covered with a bra.

## **Results**

The daily milk yield was higher in group 1 ( $P < 0.01$ ) compared with group 2;  $2.5 \pm 0.8$  and  $2.1 \pm 0.6$  kg, respectively. The fat content was higher in group 1 ( $P < 0.001$ )  $4.9 \pm 0.9$  % than in group 2;  $4.4 \pm 0.7$  %. The protein and lactose content were  $3.2 \pm 0.4$  % and  $4.8 \pm 0.4$  % in group 1 and  $3.1 \pm 0.3$  and  $4.7 \pm 0.3$  % in group 2. The daily casein number (% of total protein) was  $72 \pm 5$  % in both groups.

## **Discussion**

The results indicate that it is possible to maintain milk yield with one kid present. Suckling may also increase the fat content if the kid is allowed to suckle before each milking.

## **Conclusion**

This study shows that the milk composition is positively affected by keeping goats and kids together.

# PROTEOME ANALYSIS OF FUNCTIONALLY DIFFERENTIATED MAMMARY EPITHELIAL CELLS ISOLATED FROM COW AND BUFFALO MILK

Jagadeesh Janjanam<sup>1</sup>, Nishant Varshney<sup>1</sup>, Surender Singh<sup>1</sup>, Manu Jamwal<sup>1</sup>, Sudarshan Kumar, Ajay K. Dang<sup>1</sup>, Jai K. Kaushik<sup>1</sup>, D. Malakar, B. P. Mishra<sup>2</sup>, Manish Mukesh<sup>2</sup>, Ranjit Kataria<sup>2</sup>, T. Mukhopadhyay<sup>3</sup>, B.S. Prakash<sup>1</sup>, S. Grover<sup>1</sup>, Aswini Panigrahi<sup>4</sup> & Ashok. K. Mohanty<sup>1</sup>✉

<sup>1</sup>*Animal Biotechnology Centre, National Dairy Research Institute, Karnal, India*

<sup>2</sup>*National Bureau of Animal Genetic Resources, Karnal, India*

<sup>3</sup>*National Centre for Human Genome Studies and Research, Punjab University, Chandigarh, India*

<sup>4</sup>*King Abdullah University of Science and Technology, Thuwal, Jeddah, Kingdom of Saudi Arabia*

✉ashokmohanty1@gmail.com

## Introduction

The mammary gland is made up of a branching network of ducts that end with alveoli. Each alveolus consists of epithelial cells surrounding a lumen. Mammary Epithelial Cells (MEC) reflects the milk producing ability of farm animals under selection pressure. To date, a two dimensional electrophoresis (2-DE) reference map of proteins or shotgun proteome of MECs is not yet available.

## Aim

The aim of our study was to characterize the mammary epithelial cells isolated from milk and to identify proteins of MECs expressed during peak lactation to create a protein database using two dimensional gel electrophoresis and shotgun proteomics approaches.

## Materials & Methods

MECs from milk were purified by using Immunomagnetic separation. The purity of the MECs was confirmed by qRT-PCR and Western blotting. The Cell lysate was subjected to 2-DE followed by identification of proteins spots by MALDI-TOF-TOF. The cell lysate was also subjected to the RP-LC coupled with the ESI-MS for shotgun proteomics and data was analyzed using Sequest and Trans Proteome Pipeline Software.

## Results & Discussion

The homogeneity of isolation of isolated MECs was validated by expression analysis of Cytokeratin 8,  $\alpha$ -LA and EMA in MECs qRT-PCR and the  $\alpha$ -LA expression was 98 times higher in MECs compared to the Somatic cells. Expression of Cytokeratin 8 and casein was further confirmed by western blotting. After image analysis of 2D gel, we observed 215 protein spots in cattle. In shotgun proteome after data analysis, we identified 260 proteins in cattle and 422 proteins in buffalo MECs with >95% confidence. The proteins identified in shotgun approach involved in many biological processes including carbohydrate metabolism, fat metabolism, electron transport, protein metabolism and cell structure.

## Conclusion

This study represents to date the first detailed and reproducible 2D protein map of bovine MECs. The shotgun proteome analysis of both cattle and buffalo represent the maximum number of proteins specific to mammary epithelial cells like cytokeratins, casein, lactalbumin, lactoglobulin, butyrophillin etc. indicating the purity of isolated epithelial cells.

# INTER-RELATIONSHIP OF PHAGOCYtic ACTIVITY OF BLOOD NEUTROPHILS WITH PLASMA CONCENTRATION OF TNF- $\alpha$ , IL-6 AND MILK SCC IN MASTITIC CROSSBRED COWS

Sandeep Kaswan<sup>1</sup>✉, Joydip Mukherjee<sup>2</sup>, Shiv Prasad<sup>1</sup> & A.K. Dang<sup>2</sup>

<sup>1</sup>*Livestock Production and Management Division*

<sup>2</sup>*Dairy Cattle Physiology Division*

*National Dairy Research Institute, Karnal, Haryana, India 132001*

✉deepu02vet@gmail.com, deepu02vet@yahoo.com

## Introduction

Understanding the inflammatory responses elicited by cytokines is fundamental to developing specific preventive and therapeutic strategies for mastitis.

## Aim

The aim was to provide quantitative estimation of cytokines TNF- $\alpha$  and IL-6 along with alterations in phagocytic activity (PA) of blood neutrophils and milk somatic cell count (SCC) during clinical mastitis.

## Material and Methods

Blood and milk samples were collected from mastitic Karan Fries (Holstein Friesian x Tharparkar) cows (n=8) for consecutive 5 days of clinical mastitis. *In vitro* PA of isolated blood neutrophils were estimated by Nitroblue Tetrazolium Chloride (NBT) reductive assay. SCC of milk was estimated microscopically. Quantitative estimation of bovine cytokines (IL-6 and TNF- $\alpha$ ) was done by using ELISA kit. Significance was tested by employing two-way ANOVA using SYSTAT.

## Results

Milk SCC, plasma concentration of TNF- $\alpha$  and IL-6 were higher (P<0.01) on day of onset of mastitis and tended to normalize during remaining days of clinical mastitis. PA of blood neutrophils was lower (P<0.01) on the first day of mastitis and then increased. On comparison between days of mastitis a positive correlation (P<0.05) was found between TNF- $\alpha$ , IL-6 and SCC and these were negatively correlated (P<0.05) with PA of blood neutrophils.

## Discussion

PA of blood neutrophils significantly impaired due to mastitis. These cytokines are found to be associated with recruiting leukocytes to the udder and thereby seen as elevated SCC and inactivation of PA of blood neutrophils and prolongation of their lifespan and recruitment to udder tissue.

## Conclusions

Further investigations are needed to find the exact mechanism for the relationship between cytokines and activity of blood and milk leukocytes, to expand scope for emerging cytokine immunotherapy.

# THE EFFECT OF A SINGLE PROLONGED MILKING INTERVAL ON INFLAMMATORY PARAMETERS, MILK COMPOSITION AND YIELD IN DAIRY COWS

Branislav Lakić<sup>1</sup>✉, Kerstin Svennersten Sjaunja<sup>2</sup>, Lennart Norell<sup>3</sup>, Rupert. M. Bruckmaier<sup>4</sup>, Johanna Dernfalk<sup>5</sup> & Karin Östensson<sup>1</sup>

<sup>1</sup>*Department of Clinical Sciences, Swedish University of Agricultural Sciences, Box 7054, 750 07 Uppsala, Sweden*

<sup>2</sup>*Department of Animal Nutrition and Management, Kungsängen Research Centre, 753 23 Uppsala, Sweden*

<sup>3</sup>*Unit of Applied Statistics and Mathematics, Swedish University of Agricultural Sciences P.O. Box 7013, 750 07 Uppsala, Sweden*

<sup>4</sup>*Veterinary Physiology, Vetsuisse Faculty University of Bern, Bremgartenstr. 109a, 3001 Bern, Switzerland*

<sup>5</sup>*Department of Anatomy and Physiology, Swedish University of Agricultural Sciences, PO Box 7011, SE-750 07 Uppsala, Sweden*

✉lakic.branislav@kv.slu.se

## Introduction

A technical stop in automatic milking systems may result in a severely prolonged milking interval (PMI) with subsequent impact on milk somatic cell count (SCC).

## Aims

To investigate the inflammatory reaction, milk composition and yield after exposing cows to a single PMI of 24 h.

## Materials and methods

Composite milk and blood samples were collected prior and subsequent to PMI from 27 Swedish red cows and analyzed for common inflammatory indicators.

## Results and discussion

At the first milking after the PMI, a sharply increased proportion of milk polymorphonuclear leukocytes (PMN) but marginally increased SCC were observed. The peak in SCC was not observed until morning milking day 2 after the PMI, notably, concomitantly with a decreased PMN proportion. Increased concentrations of blood lactose, milk bovine serum albumin (BSA), serum amyloid A (SAA) and prolactin (PRL), and a drop in milk alpha lactalbumin (ALA) were observed concomitantly with the peak in PMN. In despite of increased SCC after PMI lactate dehydrogenase was decreased in afternoon milk, as was afternoon milk yield. Milk composition was slightly altered, probably due to differences in milk yield. Unaltered concentrations of milk and blood cortisol suggest that a PMI was not associated with stress. Interleukin-1 $\beta$  could not be detected in milk at any time. The changes in SCC, SAA and PRL had the longest duration. SAA, ALA and PRL, respectively, may directly or indirectly induce chemotaxis, thus changed concentrations observed after the PMI might have contributed to the enhanced migration of PMN to milk.

# GENETIC VARIATION AND SPREAD OF *Staphylococcus aureus*, *Streptococcus dysgalactiae* AND *Streptococcus uberis* IN MASTITIS IN DAIRY COWS

Åsa Lundberg<sup>1,2</sup>✉, Anna Aspán<sup>1</sup>, Ann Nyman<sup>1</sup>, Helle Ericsson Unnerstad<sup>1</sup>, Karin Östensson<sup>2</sup> and Karin Persson Waller<sup>1,2</sup>

<sup>1</sup>National Veterinary Institute, Uppsala, Sweden

<sup>2</sup>Swedish University of Agricultural Sciences, Uppsala, Sweden

✉asa.lundberg@sva.se

## Introduction

Mastitis is the most common disease among dairy cows and most clinical cases occur just after calving. The earlier in lactation the cow gets mastitis the larger the consequences on milk yield and udder health. Mastitis is mostly caused by bacterial infections, among which *Staphylococcus aureus* (Sa), *Streptococcus dysgalactiae* (Strd) and *Streptococcus uberis* (Stru) are common in Sweden.

## Aim

The overall aim is to gather knowledge about mastitis caused by Sa, Strd and Stru to prevent such infections. Specific aims are to investigate genetic variation among Sa, Strd and Stru, to investigate when infections of importance for udder health around calving occur in relation to calving, and to identify sources of infection of Sa, Strd and Stru in affected herds.

## Material and methods

Bacterial isolates collected during a Swedish study of clinical mastitis in 2002/2003 will be genotyped by relevant methods such as pulsed-field gel electrophoresis. The effect of different bacterial strains on udder health will be compared using data extracted from the milk recording scheme. In the second part, quarter milk samples will be collected from cows at calving and four days after calving, and from cases of clinical mastitis during the first month after calving in 20 dairy herds. The samples will be analyzed for Sa, Strd and Stru. Based on the results of part two, environmental and animal body samples will be collected in selected herds to identify important sources of Sa, Strd and Strd infections.

## Results

No results have yet been generated. The project started 2010 and will finish 2014/15.

# **SIGNIFICANCE AND APPLICATION OF THE ONLINE CELL COUNTER IN THE MILKING ROBOT**

Ivars Lūsis<sup>✉</sup>, Vita Antane & Armins Laurs

*Latvia University of Agriculture*

✉[ivars.lusis@llu.lv](mailto:ivars.lusis@llu.lv)

## **Introduction**

Online cell counter (OCC, DeLaval) built in the milking robot for assessing somatic cell count (SCC) of an individual cow during milking is offered. Each day several SCC values for all cows can be obtained, and farmers are concerned about practical use of them, especially when SCC fluctuates.

## **Aim**

The aim was to study an individual cow SCC dynamics, characterise changeability over milking sessions, and to elaborate recommendations for practical use of OCC data.

## **Material and Methods**

On a dairy farm, where a milking robot had been used for more than one year, OCC data were compared with a routine SCC count (automatic analyzer Somacount300). A qualitative comparison was performed using Cohen's kappa statistics at mastitis diagnostic SCC threshold 200 000 cells/ml. The SCC dynamics pattern obtained from the herd management data system during one month was graphically analyzed in 35 cows. Dynamics patterns were defined depending on the level and changeability of SCC.

## **Results and Discussion**

The qualitative agreement of results from OCC and the reference method was high ( $\kappa=0.92$ ). According to SCC pattern during 20 milking sessions cows could be assigned to one of four dynamics types (groups): dynamics below threshold (23% of cows), dynamics below threshold with some increases (31%), dynamics near threshold (31%) and dynamics in wide range around threshold (15%).

## **Conclusions**

The milking robot OCC is a reliable tool for monitoring of SCC in milk from individual cows. In order to identify possible mastitic cows we recommend that assessment should be based on SCC dynamics analysis of at least 20 consecutive milking sessions.

# ANALYSIS OF EXPRESSION OF BOVINE INSULIN-LIKE GROWTH FACTOR BINDING PROTEIN-5 (IGFBP-5) DURING LACTATION AND ITS POSSIBLE ROLE IN LACTATION PERSISTENCY

Sushil K. Mohapatra, Surender Singh, Nishant Varshney, Sudarshan Kumar, Jagdeesh Janjanam, Vijay Anand J, Jai K. Kaushik, Ajay K. Dang, T. K. Datta, Subrata K. Das and Ashok K. Mohanty✉

*Proteomics and Structural Biology Lab, Animal Biotechnology Centre, National Dairy Research Institute, Karnal, 132001, Haryana, India*

✉[ashokmohanty1@gmail.com](mailto:ashokmohanty1@gmail.com)

## Introduction

IGFBP-5 is a 29 kDa pro-apoptotic protein associated with apoptosis of secretory mammary epithelial cells during mammary gland involution. Over expression of IGFBP-5 in the mammary gland causes impaired mammary gland development and cessation of milking.

## Aim

Investigation of IGFBP-5 in bovine milk during the lactation from healthy, involuting and mastitic udders and understanding the possible role of IGFBP-5 in lactation persistency.

## Materials and Methods

Expression profiling of bovine IGFBP-5 in milk was analyzed by ELISA and Real-Time PCR (RT-PCR) in 6 lactating Sahiwal cows in different stages including involution, less persistent lactations and mastitis.

## Results

The RT-PCR data on IGFBP-5 showed  $69.1 \pm 14.16$  and  $23.27 \pm 6.21$  fold expression during involution and colostrum respectively than peak lactation. Expression of IGFBP-5 ( $\mu\text{g/ml}$ ) during lactation was observed to be  $420.00 \pm 127.01$  (colostrum);  $5.32 \pm 1.36$  (50 days),  $25.02 \pm 6.21$  (100 days),  $15.17 \pm 3.94$  (150 days),  $173.91 \pm 23.35$  (200 days),  $131.35 \pm 48.8$  (250 days),  $164.03 \pm 25.85$  (300 days),  $2675 \pm 158.77$  (involution),  $58.41 \pm 10.00$  (less persistent lactations),  $417.95 \pm 123.42$  (sub-clinical mastitis) and  $946.43 \pm 55.02$  (clinical mastitis). Expression level of IGFBP-5 during involution was observed to be 7.7, 19.7, 129.5 and 608.9 times higher in comparison to colostrum, late, peak and early lactation respectively. The expression of IGFBP-5 in subclinical and clinical mastitis was observed to be 16.7 and 37.82 fold higher in comparison to peak lactation. It was clearly indicated that there was a gradual increase in the level of IGFBP-5 in mammary gland from early to late lactation and a sharp increase during involution.

## Discussion

Expression of IGFBP-5 during lactation is negatively correlated with milk yield. In all possibility IGFBP-5 has a role to play in dictating lactation persistency.

## Conclusion

IGFBP-5 in milk could be explored to be used as a prognostic biomarker of lactation persistency.



# CHRONIC MASTITIS; COULD IT BE A RISK FACTOR FOR BREAST CANCER?

Sasan Nazemi✉ and Christopher H Knight

*University of Copenhagen Faculty of Life Sciences, Grønnegårdsvej 7, DK1870 Frb C Denmark*

✉*sasan@life.ku.dk*

## Introduction

A causal relationship between chronic inflammation and tumour incidence has been proposed in several tissues, but has not been investigated in the mammary gland. Inflammation of the breast (mastitis) is a relatively common occurrence, affecting around 20% of breastfeeding mothers, often in a chronic way. Breast cancer affects around 10% of women in developed countries. Breast cancer databases do not record mastitis incidence, so any investigation of a possible relationship requires an animal model.

## Aims

The aim of the project which has begun recently and which will be briefly described here is to develop a murine model of chronic mastitis and tumourigenicity.

## Methods

Individual mammary glands of lactating mice will be inoculated through the teat duct with a strain of *Staphylococcus aureus* derived from a subclinical bovine mastitis case at doses designed to create a chronic subclinical mastitis. Spontaneous tumour incidence will be compared in inoculated and control glands of tumour-susceptible and tumour-resistant strains. Tumour induction strategies employing steroid challenge and/or transformed cell seeding may also be employed.

## Results

This is a preliminary report. Development of the murine model is under way and to date the microinjection based inoculation procedure has been successfully established *post mortem*.

## Conclusions

The mouse represents a species where there is good data regarding tumourigenesis and physiological inflammation (involution) but relatively little regarding pathogenic mastitis. We anticipate that a murine mastitis model will be useful not only for the stated aims but also for investigation of the mammary acute phase response and other inflammation-related phenomena.

# **KANGAROO MOTHER CARE AT TWO SWEDISH NICUS – REPORT FROM AN ONGOING STUDY**

Ylva Thernström Blomqvist<sup>✉</sup>, Kerstin Hedberg Nyqvist, Christine Rubertsson & Uwe Ewald

*Department of Women's and Children's Health, Uppsala University, and University Children's Hospital, Uppsala, Sweden*

✉ylva.thernstrom\_bloomqvist@kbh.uu.se

## **Introduction**

Kangaroo Mother Care (KMC) is an intervention for facilitating the transition from intra- to extra-uterine life of low birth-weight infants'. KMC consists of skin-to-skin contact between the low birth-weight infant and the mother or a substitute for the mother, such as the father. KMC has been associated with higher breastfeeding rates at discharge and up to six months

Data on parents' experiences of KMC are required for giving parents adequate guidance in applying KMC and for continuing the development of this model of care.

## **Aim**

To explore mothers' and fathers' experiences of providing KMC to their preterm infants.

## **Materials and Methods**

In study A 23 mother received a questionnaire with questions about the infant's care and KMC. In study B interviews were conducted with 7 fathers to infants born prematurely.

## **Result**

Study A showed that Swedish mothers were willing and able to provide their preterm and/or ill newborns with KMC, provided that they received the assistance and support they desired from nurses.

Study B showed that application of KMC made fathers feel they had control and did something good for their infant, although the infant's care could be demanding and stressful. Some fathers stayed with the infant during the whole hospital stay, others visited the infant every day.

## **Discussion and conclusion**

Provided that these parents received the assistance and support they required, they accepted the KMC method very well. This supports the perception that Swedish parents are able and willing to provide KMC - also during the infant's whole hospital stay, although this sometimes can be demanding.

# BEHAVIOUR, HEART RATE AND SALIVA CORTISOL IN GOAT KIDS REARED WITH OR WITHOUT MATERNAL CONTACT

Louise Winblad von Walter<sup>✉</sup>, Eva Hydbring-Sandberg & Kristina Dahlborn

*Faculty of Veterinary Medicine and Animal Science, Swedish University of Agricultural Sciences*

*Department of Anatomy, Physiology and Biochemistry, Box 7011*

*SE – 750 07 Uppsala, Sweden,*

<sup>✉</sup>*Louise.Winblad@afb.slu.se*

## Introduction

A common practice on dairy farms is to permanently separate mother and young immediately after parturition. In developing countries the milk is used both for suckling and human consumption and does and kids are only temporarily separated.

## Aim

To investigate how different rearing systems affect behaviour and physiology in goat kids.

## Materials and methods

After the colostrum period, 10 kids were reared with other kids but fed goat milk (Group 1). Six kids were separated from mothers at daytime (Group 2). Six kids were not separated from their mothers (Group 3). At 2 weeks of age, behavioural observations were made. In addition, an isolation test with subsequent reunion with mothers (Group 2 and 3) or the other kids (Group 1) was performed. A Polar Sport tester (Polar Ltd, Bromma, Sweden) was used for telemetric recordings of heart rate during isolation and saliva samples were taken before and after.

## Results

There were no differences in weight gain between the groups. Group 3 spent more time lying down alone than Group 1 ( $P \leq 0.05$ ) and Group 2 and 3 slept more alone than Group 1 ( $P \leq 0.05$  and  $P \leq 0.001$  respectively). Heart rate and saliva cortisol concentration did not differ between groups at isolation or reunion. The mean heart rate was higher during reunion than isolation for all groups ( $P < 0.05$ ).

## Discussion

Regardless of rearing system, all kids were fed goat milk which apparently was sufficient for proper weight gain. Kids reared with their mother spent more time alone than kids reared without their mother. At reunion all kids suckled mother/self-feeder immediately.

## Conclusion

Rearing system had no effect on weight gain but influenced the choice of resting alone or with social companion.

# MASTITIS MONITORING IN THE LOMONOSOVSKI DISTRICT OF THE LENINGRAD REGION

Maria Vinokhodova<sup>✉</sup> & A. A. Sukhinin

Saint-Petersburg State Academy of Veterinary Medicine, Russia

<sup>✉</sup>hscmvmv@mail.ru

## Introduction

The Leningrad region has a leading position in North-East of the Russian Federation in the agricultural industry, including black-mixed cattle with high milk production. However problems with mastitis are holding back further intensification of the dairy industry in the area.

## Aim

The aim of our investigation was to reveal the most common mastitis pathogens and describe the causes of mastitis in farms in the Leningrad region.

## Materials and methods

The three largest farms in the Lomonosovski district of the Leningrad region were chosen for monitoring during 11 months in 2010. Samples from cows were taken once per month per cow. Clinical mastitis was diagnosed by clinical examination of cows and organoleptic evaluation of milk. Subclinical mastitis was diagnosed with Mastidin (FGUP Privolzhskaja biofactory) and Keno test (CID Lines N. V.). Bacteria in milk were isolated, identified and tested for antibiotics sensitivity according to standard methods.

## Results

A total of 13600 milk samples were analyzed, 1782 cases of mastitis were revealed, among them 790 subclinical cases, 17 animals were culled due to severity of their mastitis. In 254 animals more than one incidence of mastitis was observed.

**Table 1.** Number of mastitis cases and isolated mastitis agents in three dairy farms in the Leningrad region during 2010.

Farm/number of cows/number of analysis	1/505/4550	2/780/7400	3/155/1650
Number of mastitis cases	736	988	58
- subclinical cases	416	364	12
- clinical cases	320	624	46
Animals with recurrent disease	146	98	10
Number of culled animals	4	10	3
The most common pathogens in milk samples	<i>E.coli</i> , <i>Actinomyces pyogenes</i>	<i>E.coli</i> , <i>Str.epidermidis</i> , <i>St.aureus</i>	<i>E.coli</i> , <i>Citrobacter sp.</i> , <i>Enterococcus faecalis</i>

## Discussion

The mastitis agent species composition allowed us to establish the causes of the pathology. The antibiotics sensitivity analysis of agents allowed us to choose the most effective preparations for therapy of the disease.

## Conclusions

1) The main causes of mastitis in the examined farms were inappropriate management of cows and milking machines. 2) The most effective preparations for treating mastitis in the examined farms were *ampicillinum* and *enroxilum*. 3) Improved management of cows and milking machines is necessary for further intensification of the dairy industry in the Leningrad region.

## **CRU Publication series**

### **Report 1**

Proceedings of the Inauguration of the Centre for Reproductive Biology, 1997, by *Andersson H, Kindahl H, and Neil M (editors)*

### **Report 2 (In Swedish)**

Svinforskning vid SLU - Presenteras för Sveriges grisproducenter vid föreningens årsmöte på Ultuna 25-27 juni, 1998 by *Andersson K (editor)*

### **Report 3**

Advances in Canine Reproduction - Minisymposium at SLU, September 3, 1998, by *Linde-Forsberg C (editor)*

### **Report 4**

Sperm behaviour prior to fertilization in animals - Special symposium at SLU, November 16, 1998, by *Larsson B and Rodriguez-Martinez H (editors)*

### **Report 5**

Aspects of Equine Reproduction- symposium at SLU, April 15, 1999, by *Rodriguez-Martinez H (editor)*

### **Report 6**

Impaired Reproductive Performance in Postpartum Cows- symposium at SLU, May 26, 1999, by *Kindahl H (editor)*

### **Report 7**

Dairy production in Estonia - Today and Tomorrow. Symposium at Estonian Agriculture University, Tartu, June 7, 1999, by *Tiirats T and Magnusson U (editors)*

### **Report 8**

Aspects of Ovine Reproduction - Special symposium at SLU, June 8, 1999, by *Söderquist L, Forsberg M and Rodriguez-Martinez H (editors)*

### **Report 9**

Symposium I: Reproduction in Aquatic Organisms

Symposium II: Bird Reproduction Symposium: Avian Fertility - Mechanisms and Application, by *Madej A, Waldenstedt L and Norrgren L (editors)*

### **Report 10**

Proceedings of the Inauguration of CRU: Reproduction and Our Environment, Uppsala November 4, 1999, by *Ulf Magnusson (editor)*

### **Report 11**

Feeding, metabolism and infections in farm animals with special reference on reproduction.

Proceedings from a symposium at Estonian Agriculture University, Tartu, February 24-25, 2000, by *B Aasmäe, T Tiirats and U Magnusson (editors)*

### **Report 12**

Sexual Biology from Fish to Humans. Proceeding from a symposium in Uppsala, May 18, 2000, by *U Magnusson, M Neil and M Olovsson (editors)*

**Report 13**

Hormones in the Environment and Animal Production - a Public Health Concern? Proceeding from a symposium in Uppsala, March 13, 2001, by *B Brunström and U Magnusson (editors)*

**Report 14**

Reproductive Failure in Farm Animals. Proceedings from a symposium at Estonian Agriculture University, Tartu, June 14-15, 2001, by *T Tiirats (editor)*

**Report 15**

Envirovet Baltic: Workshop on Ecosystem Health. Proceedings from a workshop in Saaremaa, Estonia, August 15-19, 2002, by *Börje K. Gustafsson and Ulf Magnusson (editors)*

**Report 16**

Farm animal reproduction: Reducing infectious diseases. Proceedings from a symposium at the Faculty of Veterinary Medicine, Jelgava, Latvia, January 22-23, 2003, by *Vita Antane and Ulf Magnusson (editors)*

**Report 17**

Farm animal reproduction: Conserving local genetic resources. Proceedings from a mini-symposium at The Faculty of Veterinary Medicine, Kaunas, Lithuania September 13-15, 2003, by *Renée Båge and Aloyzas Januskauskas (editors)*

**Report 18**

Reproductive techniques in conservation biology. Proceedings from a CRU seminar at SLU, March 18, 2004, by *Renée Båge (editor)*.

**Report 19**

Animal farming in transition - the role of animal reproduction: Mastitis symposium. Proceedings from a symposium at the St Petersburg Academy of Veterinary Medicine, Russia, January 10-12, 2007, by *Renée Båge, Nina Fedosova, Kirill Plemyashov and Maria Stakheeva (editors)*.

**Report 20**

Avian reproductive physiology. Report from a workshop at Hiiumaa/Dagö, Estonia, December 10-12, 2006, by *Mats Björklund and Mare Lohmus (editors)*.

**Report 21**

Animal farming in transition – the role of animal reproduction: Modern farming and domestic animal reproduction. Proceedings from a symposium at the Vitebsk State Academy of Veterinary Medicine, the Republic of Belarus, March 5-7, 2008, by *Renée Båge and Ratislau Kuzmich (editors)*.

**Report 22**

Lactation research in mammals and humans: Comparative aspects with focus on milk composition and mastitis. Proceedings from a symposium in Uppsala, Sweden, November 13-14, 2008, by *Elisabeth Kylberg, Bodil Ström Holst and Kerstin Svennersten Sjaunja (editors)*.

**Report 23**

Antimicrobial strategies in human and animal reproduction. Proceedings from a symposium in Uppsala, Sweden, May 7, 2009, by *Renée Båge, Matts Olovsson and Bodil Ström Holst (editors)*