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Variation in udder health indicators at different stages of lactation in goats with no udder infection

Ylva Persson[a,*], Torben Larsen[b], Ann-Kristin Nyman[a]

[a] Department of Animal Health and Antimicrobial Strategies, National Veterinary Institute, 751 89 Uppsala, Sweden
[b] Department of Animal Health and Bioscience, Aarhus University, Denmark

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ABSTRACT

Mastitis is an important disease in dairy goat production. Subclinical mastitis is common in goats and is mainly caused by contagious bacteria. Several methods to diagnose mastitis in goats are available but have not all been investigated in healthy udders and at different stages of lactation. The purpose of the study was to investigate the variation in some udder health indicators at different stages of lactation in goats without intramammary infection (IMI). The udder health indicators were: somatic cell counts (SCC) measured by DeLaval Cell Counter (DCC) and estimated by California Mastitis Test (CMT), lactate dehydrogenase (LDH) activity, N-acetyl-β-D-glucoseaminidase (NAGase) activity and alkaline phosphatase (AP) activity.

Milk samples from twenty-four clinically healthy dairy goats were collected on two consecutive days in early, mid and late lactation. At milking, each goat's udder half was given a CMT score before udder half milk samples were collected. The milk samples were then analyzed for SCC, LDH, NAGase and AP, and investigated for bacterial growth. Variation in udder health indicators between udder half within goat, samples between sampling days and samples between stages of lactation were investigated using multivariable mixed-effect linear regression and multivariable ordinal logistic regression models.

Of the 24 goats, 18 were considered IMI negative at all samplings, 3 goats had inconclusive results for one udder half in late lactation and 3 (12.5%) had IMI positive udder halves in one or more lactation periods. Period of lactation was significantly associated with all udder health indicators with an increase in all indicators at late lactation compared to mid and early lactation. For NAGase and AP, period of lactation was significant as an interaction term with sampling day. NAGase was significantly higher on day 2 compared to day 1 in mid lactation and significantly lower on day 2 than day 1 in late lactation. AP was significantly higher on day 2 compared to day 1 in early lactation and significantly lower on day 2 than day 1 in late lactation. Moreover, for CMT there was a significant association with udder half with a higher general (over period and day) probability of higher CMT scores in the right udder half compared to the left.

This study shows that SCC, LDH, NAGase and AP were all affected by period of lactation but also to some extent by sampling day and udder half. This must be considered when interpreting udder health indicators sampled at different stages of lactation.

* Corresponding author. Tel.: +46 18 674162.
E-mail addresses: ylva.persson@sva.se (Y. Persson), torben.larsen@agrsci.dk (T. Larsen), ann.nyman@sva.se (A.-K. Nyman).

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1. Introduction

Subclinical mastitis in goats is common (Contreras et al., 2007) and considered an important disease since it can lead to decreased milk yield, impaired milk quality (Leitner et al., 2004a) and poor milk hygiene. Good milk hygiene is especially important when unpasteurized milk is used for cheese production (Oliver et al., 2005). Subclinical mastitis in goats is mainly caused by bacteria; coagulase-negative staphylococci (CNS) and *Staphylococcus aureus* (*S. aureus*) being the most common pathogens (Bergonier et al., 2003; Persson and Olofsson, 2011). It is important to detect goats with IMI at an early stage in order to prevent further spread of bacteria in the herd and to reduce the negative effect on milk production and milk composition. Presence of IMI may be diagnosed by bacterial culturing, but also indirectly by measuring inflammatory indicators in milk; e.g. somatic cell count (SCC), electrical conductivity, acute phase proteins and different enzyme activities. Bacterial culturing takes time and is costly. As many indirect measurements are faster and cheaper to perform, they have the potential to be effective diagnostic tools.

Somatic cell count is the most widely used indicator of udder health in cow, sheep and goat milk, but can be difficult to interpret in goats. Compared to sheep and cows, SCC in goat milk is relatively high also in the healthy udder and it increases throughout the lactation as well as with parity and during oestrus (Paape and Capuco, 1997; McDougall and Voermans, 2002; Christodoulopoulos et al., 2008). There is also a great variation in SCC among farms and among individuals (Schaeren and Maurer, 2006). Nevertheless, some authors claim that SCC is a good predictor of IMI in dairy goats (Poutrel et al., 1997; Persson and Olofsson, 2011). In two longitudinal field studies by Koop et al. (2013) investigating risk factors for subclinical IMI in goat, it was shown that stage of lactation, parity and milk yield influenced the sensitivity and specificity of using SCC at a cut-off of 2000×10^3 cell/mL for finding goats with an IMI caused by a major pathogen.

Earlier investigations have revealed that enzyme activities in the udder epithelium change markedly (Bogin et al., 1976; Banga et al., 1989) due to mastitic inflammation. Similarly, enzyme activities in blood serum/plasma or fractions of blood cells have proven to be indicative of experimentally induced mastitis (Symons and Wright, 1974; Banga et al., 1989; Heyneman and Burvenich, 1992). More practical attention has been given to detection of enzyme activity in milk, and numerous enzymes have been proposed and listed as reliable markers of bovine mastitis (Kitchen, 1981; Korhonen and Kaartinen, 1995). Among milk enzymes, NAGase (N-acetyl-β-D-glucoseaminidase; EC 3.2.1.30) has obtained the greatest attention due to the relative simplicity of analysis and its high correlation with SCC of milk (Kitchen et al., 1978, 1980). NAGase has been known as an indicator for mastitis detection in the last 2 to 3 decades, but other enzymes have also been suggested, i.e. alkaline phosphatase (AP; EC 3.1.3.1) (Rasmussen et al., 2008; Larsen et al., 2010) and β-glucoronidase (EC 3.2.1.31) (Larsen and Aulrich, 2011). Recently lactate dehydrogenase (LDH; EC 1.1.1.27) proved comparable qualities to NAGase for use for mastitis detection (Chagunda et al., 2006). Other studies also imply that LDH activity is one of the most reliable enzymes evaluated for the detection of IMI (Katsoulos et al., 2010; Stuhr et al., 2013). Moreover, Leitner et al. (2004a,b) as well as Stuhr et al. (2013) found that NAGase activity was significantly higher in infected udder halves compared to non-infected. Furthermore, Katsoulos et al. (2010) showed that AP activity was higher in infected udder halves compared to non-infected udder halves. However, as Stuhr et al. (2013) could show that both SCC and LDH was significantly influenced by week of lactation and parity, stage of lactation and parity must be taken into consideration when interpreting these udder health markers. To our knowledge, no public study has investigated the variation of LDH, NAGase or AP in healthy goats over an entire lactation.

The aim of the study was to investigate the variation in some udder health indicators at different stages of lactation in clinically healthy goats without IMI.

2. Methods

2.1. Animals

Every other dairy goat ($n = 24$), of the Jämtlandic breed (a typical Scandinavian dairy goat), in one farm in central Sweden was sampled in 2010. The farm was chosen because of known very good udder health and also since it was one of few farms close to the National Veterinary Institute. Of the sampled goats 6 were primiparous and the median parity was 4th parity (range 1st–10th parity). From earlier studies in this herd, we knew that the only isolated species were Coagulase-negative staphylococci (CNS) To investigate the effect of stage of lactation, the goats were sampled at three occasions during one lactation; early, mid and late lactation. The mean days in milk (DIM) in the period called early lactation was 40.2 (SD: 5.9), in mid lactation the mean DIM was 117.2 (SD: 5.9) and in late lactation the mean DIM was 223.2 (SD: 5.9). Samples in early and mid lactation were collected during morning milking and samples in late lactation were collected during afternoon milking. In early and mid lactation, does where milk twice a day with 12 h milking interval and in late lactation once a day with a milking interval of 24 h. The same person collected all samples. Only clinically healthy animals without any changes in udder consistency or milk appearance were included in the study (IDF, 1999). The herd was free from Caprine Arthritis Encephalitis Virus (CAEV).

2.2. Milk sampling and measurement of SCC

At each occasion two consecutive samples were collected 24 h apart in order to get a more reliable status for IMI (Sears et al., 1990), and to investigate if there is a day-to-day variation in the analyzed indicators. All milk samples were collected just before machine milking. The first milk was discharged. Milk samples were tested using CMT and graded from 1 to 5. The scores are ranked according to an increase in viscosity, where the highest viscosity (CMT 5) is more or less correlated to the highest SCC (modified from Schalm et al., 1971). After cleaning the teat ends with alcohol (70%), an aseptic milk sample was collected (IDF, 1999) from each udder half in sterile test tubes and sent to the National Veterinary Institute, Uppsala, Sweden, for bacteriological analysis. Milk from each udder half was also collected in additional test tubes, with bronopol as a preservative, for cell counting and measurement of enzymes. Milk aliquots from each bronopol test tube were analyzed individually at the laboratory the same day with the DeLaval Cell Counter (DCC) (DeLaval International AB, Tumba, Sweden (Berry and Broughan, 2007)).

2.3. Enzymes

Test tubes with bronopol preserved milk were deep frozen at −20 °C and sent to the faculty of Agricultural Science, Aarhus University, Foulum, Denmark for analysis of LDH, NAGase and AP. Enzyme activities were determined by kinetic fluorometric measurements. Lactate dehydrogenase activity was analyzed according to Larsen (Larsen, 2005);