

Therapeutic Management of Subclinical *Staphylococcus aureus* Mastitis in Association with Iliofemoral Lymphadenopathy in a Cow

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(Received 30 Jun 2011/ Accepted 24 September 2011)

Abstract

A 6.5 year old Holstein-Friesian dairy cow was initially presented with anorexia and reduced milk yield. Subclinical mastitis was diagnosed by California Mastitis Test (CMT). Bacteriological examination of a milk sample of the affected hind quarter revealed the detection of *S. aureus*. Additionally, rectal examination revealed very large and hard iliofemoral lymph nodes. Iliofemoral lymphadenopathy was then confirmed by ultrasound examination. This unusual case suggests that *S. aureus* intramammary infection (IMI) may possess the ability to invade the host organism by lymphatic route without causing visible inflammatory reactions.

Keywords: subclinical mastitis, cow, lymphadenopathy, *Staphylococcus aureus*

Case description

A 6.5 year old Holstein-Friesian cow was presented for clinical examination because of anorexia and milk loss which developed rapidly within two days. At the time of examination, the cow was 93 days in milk and 27 days before the cow found in heat and was inseminated without difficulty by an experienced stockman. Physical examination revealed rectal temperature, respiratory rate and heart rate within reference limits. Rectal examination revealed unchanged genital organs, but very large and hard iliofemoral lymph nodes situated just cranial and lateral to the brim of the pelvis along the left shaft of the ilium. Transrectal ultrasound examination was performed with a 5 MHz linear probe (Aloka SSD-210, Eickemeyer, Tuttlingen, Germany). Enlarged iliofemoral lymph nodes appeared as roundish confluent nodes of moderate echogenicity (Fig. 1). Hyperechoic zones were noted in the periphery as evidence of inflammation (Fig. 1). Additionally, a faint central echo was sometimes observed (data not shown). Further al-

terations were not found within the pelvic cavity. Consequently, mammary gland was carefully examined but there were no grossly visible changes to milk or changes in the mammary quarter. However, mammary lymph nodes appeared to be enlarged moderately on the left side. A California Mastitis Test (CMT) was performed on all quarters and CMT reaction was strongly positive for the left hind quarter. Intramammary infection (IMI) was suggested and a quarter milk sample was taken following German Veterinary Medicine Society guidelines for isolating and identifying mastitis pathogens (DVG, 2000). Milk sample was immediately processed at the laboratory of the Institute of Animal Hygiene of the University of Hohenheim. Briefly, a 0.1 ml aliquot from the sample was cultured on blood agar (Columbia Agar with 5% sheep blood) at 37°C for 24 h. *Staphylococcus aureus* (*S. aureus*) was diagnosed, based on colony morphology, gram staining (positive), catalase reaction (positive), and detection of coagulase using staphylase test (Oxoid Ltd., Hampshire, UK).

Monthly test day somatic cell count (SCC) of total milk of the cow was retrospectively evaluated from a milk recording service (LKV Baden-Württemberg, Stuttgart, Germany). As shown in figure 2, distribution of SCC of total milk during late (last) and early lactation suggests that the cow de-

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veloped a new infection during the dry period or soon after calving.

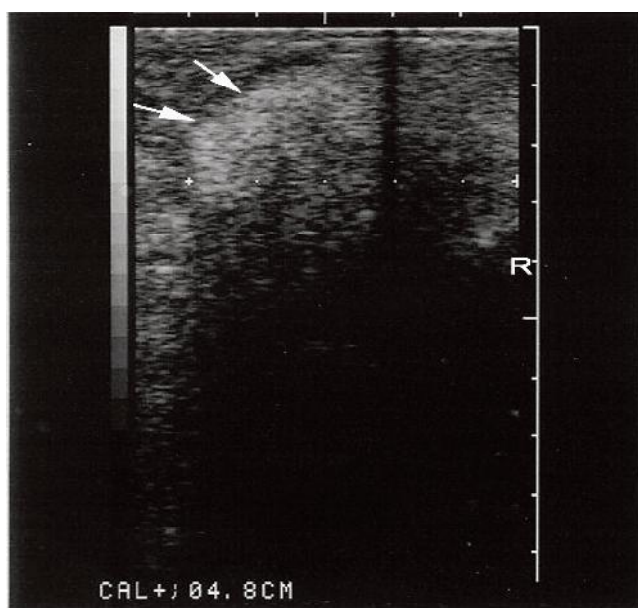


Fig. 1. Ultrasonographic appearance of enlarged iliofemoral lymph nodes (callipers) just cranial and lateral to the brim of the pelvis along the left shaft of the ilium. Note that the whole length could not be imaged in a single scan field. Hyperechoic zones are present in the periphery (white arrows). R = caudodorsal blind sac of the rumen.

Therapy

After the clinical examination, the cow was infused in the affected quarter with a commercial preparation of 200 mg cephalexin (Rilexine® 200 LC, Virbac, Bad Oldesloe, Germany) intramammary twice daily for three days and then injected subcutaneously with marbofloxacin (Marbocyl® 10%, Vêtoquinol, Ravensburg, Germany) 2 mg/kg bw once daily for three days.

Clinical course:

Controls were performed on days 4, 9, 16, and 25 after first examination. From day 4 onwards, increased appetite and milk production was noticed. Mammary lymph nodes disappeared but iliofemoral lymph nodes were still enlarged, albeit gradually reduced in echogenicity (data not shown). Additionally, antibiotic treatment of the cow resulted in immediate decline of monthly test day SCC recorded at day 96 of lactation (Fig. 2). The affected quarter of the cow was sampled at 4, 9, 16, and 25 day after onset of treatment. Apparent bacteriological cure of *S. aureus* was achieved because the same pathogen was not identified in any

posttreatment sample. However, apparent new persistent IMI was identified because coagulase-negative staphylococci (CNS) were isolated from all samples collected between day 4 and day 25.

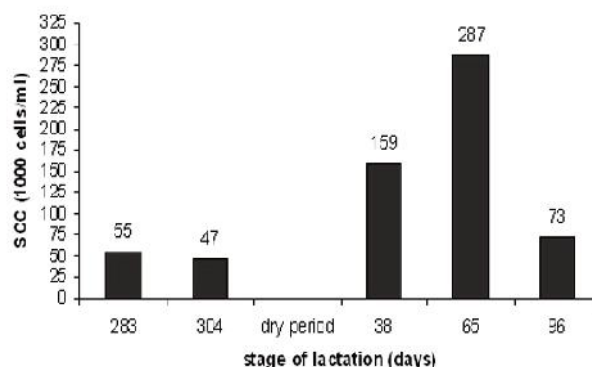


Fig. 2. Distribution of monthly test day somatic cell count (SCC) of total milk of the cow during late (last) and early lactation obtained from milk database records.

Discussion and Conclusions:

The exact reasons why a cow with subclinical mastitis may develop iliofemoral lymphadenopathy are not yet described, but it is likely to be influenced by the pathogen involved as well as the immune status of the cow (Sordillo *et al.*, 1997). *Staphylococcus aureus* pathogenicity is usually due to the production of a number of secreted and cell surface-associated proteins under the transcriptional control of specific virulence genes. For example, *S. aureus* accessory gene regulator (*agr*) genotypes may persist inside macrophages for several days without affecting the viability of these mobile cells (Kubica *et al.*, 2008). Therefore, we suggest that enlarged mammary and iliofemoral lymph nodes in this cow may represent a reactive lymphadenopathy due to *S. aureus* antigen disseminated by mobile macrophages. Our suggestion is supported by the evidence that in subclinical mastitis caused by *Listeria monocytogenes*, bacterial strains were already isolated from supramammary and iliac lymph nodes (Bourry *et al.*, 1995). Intense therapy of *S. aureus* IMI during the lactation should be performed when secondary clinical signs and/or lymphadenopathy are evident during clinical examination. In this case, therapy outcome confirmed the apparently good antimicrobial susceptibility of mastitis *S. aureus* isolates *in vitro* to marbofloxacin (Meunier *et al.*, 2004) and cephalexin (Ganière and Denuault, 2009). However, recovery of *S. aureus*

IMI did not confer immunity to infection with other bacterial species.

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