J. Dairy Sci. 96:3039–3043
http://dx.doi.org/10.3168/jds.2012-6397

Short communication: Experimentally induced mastitis reduces weight shifting between the rear legs while standing in dairy cows

N. Chapinal,*†[1] C. E. Fitzpatrick,*[2] K. E. Leslie,* and S. A. Wagner‡
*Department of Population Medicine, University of Guelph, Guelph, Ontario, N1G 2W1, Canada
†Animal Welfare Program, University of British Columbia, Vancouver, V6T 1Z4, Canada
‡Department of Animal Sciences, North Dakota State University, Fargo 58108

ABSTRACT

The objectives of this study were to evaluate changes in weight shifting between legs while standing on a weighing platform in response to endotoxin-induced clinical mastitis, and to evaluate the effect of the nonsteroidal antiinflammatory drug flunixin meglumine on weight distribution between legs while standing in dairy cattle with endotoxin-induced clinical mastitis. Clinical mastitis was induced in 10 primiparous and 9 multiparous lactating dairy cows (days in milk = 55 ± 12; mean ± standard deviation) by intramammary infusion of 100 μg of *Escherichia coli* lipopolysaccharide (LPS) into the right rear quarter. Four hours later, 10 animals were randomly assigned to receive flunixin meglumine intravenously (2.2 mg/kg of body weight; treated group) and 9 received an equivalent volume of sterile isotonic saline solution (control group). Body temperature was monitored rectally 3 d before LPS infusion, immediately before LPS infusion, and 4, 7, 10, 13, 16, and 28 h after LPS infusion. The weight applied to each leg was recorded while cows were standing on a weighing platform on the day before the challenge and 7, 10, 13, 16, and 28 h after LPS infusion. Two measures of weight shifting between the rear legs were calculated for each recording session: the standard deviation of the weight applied to the legs over time and the frequency of steps. The LPS infusion resulted in a consistent case of clinical mastitis approximately 4 h after the LPS infusion, as assessed by the presence of visible swelling and elevated rectal temperature in all cows. However, control animals had a higher temperature 7 h after LPS infusion compared with treated animals (40.8 vs. 39.0°C; standard error of the difference = 0.2). Overall, weight shifting between the rear legs was decreased 7 h after the LPS infusion compared with baseline, and this decrease was not affected by treatment with flunixin meglumine. It is likely that weight shifting increases friction between the swollen udder and the legs, increasing the pain experienced by the cow. Thus, cows with endotoxin-induced mastitis avoided shifting weight, particularly at the times when the most severe signs of inflammation occurred. Further research is needed to assess the efficacy of flunixin meglumine in mitigating udder pain and the accuracy of behavioral measures such as weight shifting in assessing analgesia in cows with mastitis.
Key words: flunixin meglumine, lipopolysaccharide, nonsteroidal antiinflammatory drug, pain

Short Communication

Mastitis is a commonly occurring disease in the dairy industry (Olde Riekerink et al., 2008), with high associated economic costs (Halasa et al., 2007). Evidence exists that even mild to moderate cases of mastitis are painful (Milne et al., 2003; Banting et al., 2008; Fitzpatrick et al., 2013) and, therefore, it is a welfare concern. Traditionally, udder pain was assessed using subjective scales (Banting et al., 2008). Recently, Fitzpatrick et al. (2013) validated a pressure algometer that assesses pain sensitivity by measuring the maximum pressure applied to the udder that an animal is willing to tolerate. However, as pain sensitivity is subject to individual variability, changes in pain sensitivity have to be assessed within each animal. This requires continuous monitoring of the pain response, which can be challenging on farm, particularly as herd size increases. Fortunately, automated methods to continuously monitor pain-related behaviors are available. For example, lying behavior can be easily monitored using data loggers (Ledgerwood et al., 2010). Several studies (Siivonen et al., 2011; Cyples et al., 2012; Yeiser et al., 2012) have described a decrease in lying behavior in the hours following the experimental induction of mastitis. These results suggest that lying down at the time when the most severe signs of local inflammation occur causes pain, and that cows choose to stand to relieve

Received November 20, 2012.
Accepted January 22, 2013.
[1]Corresponding author: nchapinal@yahoo.com
[2]Current address: Department of Large Animal Clinical Sciences, University of Saskatchewan, Saskatoon, Saskatchewan, S7N 5B4, Canada.

the pressure exerted in the udder. However, little research exists on whether cows with mastitis also adapt the way they stand to minimize udder pain. Milne et al. (2003) described a greater distance from one hock to the other hock in cows with mastitis compared with healthy cows. It is known that cows increase weight shifting between legs as well as their stepping behavior (the most obvious form of weight shifting) when they stand for long periods of time on concrete or rubber (Chapinal et al., 2011; Krebs et al., 2011). Increases in weight shifting while standing have also been associated with painful cases of lameness (Rushen et al., 2007; Chapinal et al., 2010). However, it is possible that pain associated with mastitis results in less weight shifting and stepping behavior, whereas standing is a way for the cow to minimize pain in the udder. Anecdotally, Chapinal et al. (2011) reported a decrease in the ratio number of steps to actual weight shifting in a cow with naturally occurring mastitis; the cow was shifting most of the weight without lifting the hooves off the ground. Such changes in weight distribution and stepping behavior can be assessed automatically by using specialized weighing platforms (Chapinal and Tucker, 2012).

The availability of reliable automated methods to assess pain is the first step to evaluating treatments intended to relieve pain. Nonsteroidal antiinflammatory drugs (**NSAID**) are commonly used in cattle to alleviate inflammation, pain, and fever, although the analgesic effect of NSAID in dairy cattle is not well understood. Nonsteroidal antiinflammatory drugs have been reported to decrease udder pain in mastitis (Lohuis et al., 1991; Banting et al., 2008; Fitzpatrick et al., 2013). However, the efficacy of NSAID in counteracting the changes in behavior that are likely associated with mastitis pain or discomfort, such as lying behavior or rumination, is controversial (Zimov et al., 2011; Yeiser et al., 2012; Fitzpatrick et al., 2013). The objectives of this study were (1) to evaluate changes in weight shifting between legs while standing on a weighing platform in response to endotoxin-induced clinical mastitis and (2) to evaluate the effect of the NSAID flunixin meglumine on weight shifting between legs while standing in dairy cattle with endotoxin-induced clinical mastitis. Flunixin meglumine is approved by the US Food and Drug Administration for use in cattle to treat fever associated with mastitis, respiratory disease, and endotoxemia, and for the alleviation of inflammation in endotoxemia. In this study, we also monitored rectal temperature to evaluate the occurrence and duration of the clinical mastitis episode induced by the endotoxin, and the activity of the drug.

Nineteen lactating dairy cows were enrolled in this study from January to September 2011, in 7 groups of 2 to 4 cows at a time. Only animals with no clinical signs of illness and no history of treatment for illness in the last 30 d were selected. All animals were between 38 and 73 DIM (mean ± SD = 55 ± 12 DIM). The study group was blocked into primiparous (n = 10) and multiparous (n = 9; parity range = 2 to 5) groups. All animals were housed in a freestall facility at the North Dakota State University Teaching and Research Unit (Fargo). Cows were fed a TMR daily at approximately 0630 h and were milked daily at approximately 0430 and 1530 h. All procedures involving animals were approved by the Institutional Animal Care and Use Committee at North Dakota State University.

Each animal was challenged in the right rear quarter with an intramammary infusion of 100 μg of purified bacterial *Escherichia coli* LPS (from *Escherichia coli* 0111:B4; Sigma-Aldrich Co., St. Louis, MO) reconstituted in 20 mL of sterile saline solution. Only quarters with a negative California mastitis test and negative microbiological culture (after 24 h of incubation at 37°C) 3 d before the challenge were eligible. The infusion was performed following morning milking (approximately at 0500 h) on the challenge day after disinfecting the teat with an iodine-based teat disinfectant product. More details of the challenge procedure are described in Cyples et al. (2012).

Animals were randomly assigned to treatment within parity blocks (using a random number generator available at http://www.random.org) within parity blocks. Ten animals received flunixin meglumine (Banamine injectable solution; Merck Animal Health, Summit, NJ) at a dosage of 2.2 mg/kg (dosing volume = 1 mL/22.7 kg of BW), and 9 received an equivalent volume of sterile isotonic saline solution (control group). Animals were weighed (mean ± SD = 635 ± 121; range = 548–715 kg of BW) the day before treatment to calculate individual dosages. The injections were administered by bolus injection in the left jugular vein approximately 4 h after the LPS infusion (at approximately 0900 h).

Body temperature was monitored rectally using a digital thermometer (GLA Agricultural Electronics, San Luis Obispo, CA) 3 d before LPS infusion (at approximately 0830 h), immediately before the LPS infusion and 4, 7, 10, 13, 16, and 28 h after LPS infusion. Data collected before the LPS infusion were averaged to obtain 1 baseline value per cow.

The weight applied to each leg was recorded while cows were standing on a weighing platform (Pacific Industrial Scale Co. Ltd., Richmond, BC, Canada), as described in Chapinal et al. (2010). In brief, the platform contained 4 stainless steel independent recording units (12 cm high × 59 cm wide × 99 cm long), each containing 4 stainless steel load cells (3 mV Shear Beam Load cells; Anyload LLC., Santa Rosa, CA; maximum capac-