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Dairy cows with mild-moderate mastitis change lying behavior in hospital pens

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ABSTRACT: In dairy production, mastitis is a major problem affecting animal welfare, productivity, and economy. Hospital pens are typically not used for cows with mastitis, except for severe cases involving recumbency. This field trial included 47 cows from three Danish herds followed for 8 d, of which days 1–5 involved the experimental housing. After day 5, all cows were kept with the lactating group. We examined lying behavior in dairy cows with naturally occurring, mild-moderate mastitis in hospital pens [single or group (depending on conditions on the farm), all with deep straw bedding] vs. sick cows kept in the group of healthy herd mates. Within a herd, every other cow fulfilling the inclusion criteria regarding mastitis was allocated to each of the two experimental treatments. Clinical data from involved cases were collected. No significant differences between housing treatments were found in the clinical variables or the daily lying time. During the period of experimental housing, cows kept in hospital pens showed a higher frequency of lying bouts compared with control cows. This difference did not persist after reintroduction to the lactating herd mates. These results suggest that aspects of lying behavior of dairy cows with mastitis are sensitive to the environment as the frequency of lying bouts differed between cows kept in hospital pens and cows kept in control treatment. More controlled studies are needed to examine underlying motivations and evaluate consequences in terms of animal welfare. For such studies, the inclusion of healthy cows for comparison will be valuable.

Key words: cattle, housing, mastitis, sickness behavior, welfare

INTRODUCTION

In dairy production, mastitis is a major problem affecting animal welfare, productivity, and economy (Peterson-Wolfe et al., 2018). Previous studies on the behavior of dairy cows have found that even relatively mild cases of mastitis lead to changed behavior, including decreased lying time (Cyples et al., 2012). However, the vast majority of studies have been based on experimentally induced mastitis—using Lipopolysaccharide (Zimov et al., 2011) or Escherichia coli (De Boyer des Roches et al., 2017), whereas only few studies have involved naturally infected cows [e.g., Fogsgaard et al. (2015) and Sepulveda-Varas et al. (2016)].

In recent years, the use of hospital pens in dairy production has received increased attention (Houe et al., 2016; Thomsen et al., 2019). Mastitic
cows may benefit from softer bedding and reduced competition for resources (Proudfoot et al., 2012). However, mastitic cows are typically not placed in hospital pens unless they are severe cases involving recumbency (Fogsgaard et al., 2016).

This preliminary field trial aimed to examine differences in lying behavior in dairy cows with naturally occurring, mild-moderate mastitis when housed in hospital pens versus kept with the group of lactating cows. We hypothesized that, when kept in deep straw bedded hospital pens, as opposed to the cubicles in the group of healthy cows, the mastitic dairy cows would lie down more and at an increased frequency compared with their herd mates in the group of lactating cows.

**MATERIALS AND METHODS**

This preliminary study was designed as a controlled randomized clinical indoor field trial, involving dairy cows from three commercial Danish herds. Herds were recruited by convenience sampling and the following inclusion criteria: 1) client in veterinary practice in Northern Jutland; 2) indoor loose housing; 3) suitable hospital pens (criteria below); and 4) willing to participate. Inclusion criteria for hospital pens were: 1) group or individual; 2) deep straw bedding; 3) at least one feeding place per cow; and 4) at least 8 m² per cow.

Herd size ranged from 95 to 286 cows. One herd had Danish Holsteins, the others had Danish Red Dairy breed. The herds had slatted concrete floors and one cubicle per cow, with mattresses (two herds; topped with straw or manure solids) or sand (one herd). Space available at the feed bunk ranged from one to two cows per feeding place. Two of the herds had traditional milking parlors (milking two or three times daily) and one herd had an automatic milking system (AMS). All hospital pens had deep straw bedding and maximum one cow per feeding space. Two herds mainly used individual hospital pens, whereas one only used group hospital pens housing up to five cows per pen (of which not all cows were experimental—they may have been there for other reasons than mastitis). Information on the hospital pen type used is not available for all experimental cows. Across herds, the size of hospital pens was 12–43 m². Farmers added straw when deemed needed. Cows in hospital pens were fed the same Total mixed ration for ad libitum intake as the lactating cows, and water was continuously available. Cows in hospital pens were milked in milking parlor or AMS, separated from the rest of the herd with as little waiting time as possible.

Data were collected during 12 mo starting October 2016. Cows, identified daily by the farmer during milking, with visible changes in the milk (mild mastitis) or visible changes in the milk combined with a swollen or hard quarter (moderate mastitis) were included. Cows with affected general appearance were excluded. The study did not involve healthy cows. Cows with mastitis were randomly assigned to experimental treatments: hospital pen (HOSPITAL) or kept together with lactating cows (CONTROL). The exposure period lasted 5 d, after which all cows were kept with the lactating cows again.

On the day of diagnosis (day 1), the farmer scored changes in milk, examined the udder, measured rectal temperature, and took a milk sample for bacteriology and a California Mastitis Test (CMT) test for baseline measures. If it was part of a normal herd routine, the farmer initiated antibacterial treatment. The number of cows included was determined by the availability of cows with mastitis and the number of vacant places in hospital pens. Within a herd, every other cow included was allocated to each experimental treatment. All measures taken on day 1 were repeated on day 5 (where HOSPITAL cows moved back to the lactating cows) and day 8 (data loggers removed).

A subsample of 24 cows was equipped with IceQube loggers (IceRobotics, Edinburgh, Scotland) on one hind leg. For technical reasons, we did not have access to loggers for all cows in the study. The number of cows from each herd varied but always included experimental treatment pairs (one HOSPITAL and one CONTROL). Excluding the day of logger attachment, of logger removal, and the day when HOSPITAL cows were transferred back to the lactating cows, all cows had logger recordings for 5 d, 3 during the treatment period (days 2–4) and 2 after return to the normal group (day 6 and 7). Lying time was recorded continuously and the frequency of lying bouts was calculated based on the continuous device-recorded number of transitions between standing and lying. The device stored data every 15 min, allowing calculation of lying time per 24 h and number of lying bouts per 24 h (expressed as daily median of the two periods).

Statistical analyses were performed using SAS (Version 9.3, SAS Institute Inc., Cary, NC). Effects of experimental treatment on clinical measures were evaluated using Wilcoxon rank-sum test (PROC NPARIWAY) for rectal temperature and Fisher’s exact test (PROC FREQ) for the rest of the measures. Results are presented as mean rectal
temperature ± SE and the proportion of cows within experimental treatment with the given findings. Effects of experimental treatment on lying time and frequency of lying bouts were analyzed using a generalized linear mixed model (PROC MIXED), including rectal temperature at day 1 as explanatory variable during the exposure period and CMT score and rectal temperature, both at day 5, as explanatory variables after the exposure period. Herd was included as random effect. The model included possible interactions and was reduced by backward elimination. Results are given as least square means ± SE, as well as \( F(\text{df}_{\text{treatment}}, \text{df}_{\text{error}}) \). Across analyses, a significance level of \( P < 0.05 \) was used.

**RESULTS**

Forty-seven cows completed the study: 22 HOSPITAL and 25 CONTROL. Of these, 43 had mastitis in one quarter and 4 in two quarters. The number of cows in the three herds was 6, 12, and 29. Table 1 lists clinical measures and the proportion of cows scored, with each on days 1, 5, and 8, and mean rectal temperature. At day 1, the rectal temperature varied from 38.2 to 41.9 °C, with a mean of 39.0 °C. Across treatment groups, the proportion of cows showing signs of mastitis decreased numerically during the 8 d. No differences in clinical measures between experimental treatments were found.

Table 2 shows the median daily lying time: during (days 2–4) and after (days 6 and 7) the exposure period. For the median daily lying time, no differences between treatments were found in either period. During the exposure period, the HOSPITAL cows had more lying bouts compared with CONTROL cows. No differences in the frequency of lying bouts were found for days 6 and 7.

**DISCUSSION**

This preliminary field trial examined hospital pens for dairy cows with mild-moderate naturally occurring mastitis. No differences were found in the time spent lying, but HOSPITAL cows had more lying bouts during the exposure period. Offering the cows a softer bedding and fewer obstacles as compared with cubicles with mattresses or sand in the CONTROL group, as well as a smaller group size, this result is not unexpected. Studies on healthy (Tucker et al., 2009) and lame cows (Bak et al., 2016) have shown similar results. Sick behavior has earlier been described as a highly organized behavioral strategy (Hart, 1988) but, as discussed by Aubert (1999), there are also other examples, for example, in rodents, showing that behavior during sickness is sensitive toward the environment. There may be more than one explanation for the increased number of lying bouts in cows kept in hospital pens. As discussed by Campler et al. (2018) for dry cows on deep straw, it may be bedding softness making it easier for the cows to get up and/or down but may also be fewer obstacles in the deep straw pen compared with, for example, free stalls, facilitating the forward lunge needed to gain momentum for a normal rise. It is possible that regrouping with unfamiliar cows (only experienced by HOSPITAL cows) led to increased aggression and, thus, disturbances in the lying behavior. In Campler et al. (2018), increased aggression was documented, whereas, in our study, no recordings of social behavior were included. However, whether this is a likely explanation is unclear as von Keyserlingk et al. (2008) reported a decreased frequency of lying bouts the first day after regrouping in healthy dairy cows. Another alternative explanation is different milking routines in HOSPITAL and CONTROL groups, which also may have influenced lying behavior.

Unexpectedly, the time spent lying did not differ. Lying behavior is highly prioritized in cattle (Jensen et al., 2005) and often used as the indicator of dairy

| Clinical measures | Day 1 | Day 5 | Day 8 | P value |
|-------------------|-------|-------|-------|---------|
| Rectal temperature |       |       |       |         |
| [mean (SE)]       |       |       |       |         |
| CONTROL           | 39.1 (0.16) | 38.7 (0.07) | 38.6 (0.08) | 0.96    |
| HOSPITAL          | 38.7 (0.16) | 38.5 (0.07) | 38.4 (0.04) | 0.11    |
| P value           | 0.65  | 1.00  | 0.47  |         |
| % of cows with CMT >3 |       |       |       |         |
| CONTROL           | 92%   | 44%   | 24%   | 0.54    |
| HOSPITAL          | 86%   | 45%   | 14%   |         |
| P value           | 0.25  | 1.00  | 0.24  |         |
| % of cows with clots in milk |       |       |       |         |
| CONTROL           | 76%   | 36%   | 12%   | 0.05    |
| HOSPITAL          | 91%   | 32%   | 0%    |         |
| P value           | 0.25  | 1.00  | 0.24  |         |
| % of cows with swollen quarter |       |       |       |         |
| CONTROL           | 44%   | 16%   | 16%   | 0.55    |
| HOSPITAL          | 41%   | 14%   | 5%    |         |
| P value           | 1.00  | 1.00  | 0.35  |         |

The data set included 47 cows from three commercial dairy herds. P values refer to the difference between HOSPITAL and CONTROL.
cow welfare (Ito et al., 2009), with increased lying time interpreted positively. However, if the present results can be repeated in larger, more controlled experiments, they may suggest that associations between lying time and dairy cows’ welfare are not always straightforward and positive. For example, Hernandez-Mendo et al. (2007) found lower lying time on pasture than in free-stall barns, the interpretation of which could not be attributed to the softness of lying surface but was explained by an increased motivation to graze. Thus, even though some studies show clear relationships between the softness of bedding and lying time (Tucker and Weary, 2004), future studies of relations between lying behavior and welfare should include the potential multitude of motivations, which may influence bovine lying behavior.

In the present study, no effects of the experimental treatments were found for the clinical variables rectal temperature, percentage of cows with CMT >3, clots in milk, or swollen quarters. These data were collected on days 1, 5, and 8. There may be several reasons for this finding, such as the low sample size (and cows from different herds) and the short follow-up period, but it is also possible that housing in a hospital pen is not affecting recovery. Further studies are needed to clarify this.

In conclusion, these preliminary findings suggest that lying behavior of dairy cows with mild-moderate mastitis is sensitive to the environment. Keeping cows in deep straw hospital pens was associated with increased frequency of lying bouts. Future, more controlled, studies are needed to examine the motivations underlying this finding and to evaluate the consequences in terms of animal welfare. For such studies, the inclusion of healthy cows for comparison and so-called low-resilience behaviors, such as brush use (Mandel et al., 2017), will be valuable.

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**Conflict of interest statement.** K.K.F. was employed at Aarhus University during the project period and is now employed by SEGES, offering solutions for the agriculture and food sector of Denmark. However, no part of this study has been influenced by commercial interests.

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**Table 2.** The daily lying time (min/24 h) and daily frequency of lying bouts during the two periods of data collection: days 2–4 (during the exposure period) and days 6 and 7 (after the exposure period)

|                         | During exposure (median of day 2–4) | After exposure (median of day 6–7) |
|-------------------------|--------------------------------------|-----------------------------------|
|                         | **HOSPITAL (N = 8)**                 | **CONTROL (N = 15)**              | **P value** |
| Lying time (min/24 h)   | 709 ± 63                             | 668 ± 58                          | 0.42        |
| Frequency of lying bouts| 16.0 ± 1.2                           | 12.0 ± 0.8                        | 0.01        |
|                         | 654 ± 92                             | 671 ± 94                          | 0.82        |
|                         | 12.9 ± 2.1                           | 13.5 ± 2.2                        | 0.82        |
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