Short communication: Rising and lying behavior of heifers before and after alimentary oligofructose overload

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ABSTRACT

The aim of this experiment was to describe the rising and lying behavior of heifers in the first 5 d after alimentary oligofructose overload to 1) acquire information about consequences of this experimental disease model in terms of animal behavior; and 2) contribute to the evaluation of the influence of disease on cattle welfare. Eight tethered Holstein-Friesian heifers were used and their behavior was observed for 10.5 ± 2.7 hours of light per day. Occurrence of rising and lying behavior and parts thereof were recorded. Clinical and physiological examinations were conducted throughout to confirm presence of ruminal acidosis and lameness. The duration of pre-lying intention movements were increased on d 3 after oligofructose overload compared with d -1 [28 (20 to 40) vs. 13 (10 to 16) s, respectively], and the duration of the total lying sequence was increased on d 1 and 3 compared with the control day [27 (22 to 33) and 36 (25 to 48) vs. 17 (13 to 21) s, respectively]. The frequency and duration of getting up did not differ between days. These results suggest that disease induced by oligofructose overload (i.e., ruminal acidosis and lameness) can affect lying behavior in heifers as shown by increased duration of lying intentions and difficulty in lying down, which may indicate that lying motivation was thwarted, possibly by pain or discomfort.

Key words: rising behavior, oligofructose, lameness, ruminal acidosis

Hoof and leg problems in dairy cows are often long lasting. These problems are most likely associated with pain and are considered a significant welfare problem (Ingvarsen et al., 2006). Consequences of lameness in terms of behavioral welfare indicators have only recently been addressed and, until now, focus was on

measures of activity and lying time (Juarez et al., 2003; O'Callaghan et al., 2003). From other animal species it is known that disease can lead to behavioral changes such as immobility and recumbency, which function to conserve energy and encourage recovery (Hart, 1988), but very few descriptions of sickness or pain behavior exist in cattle (Borderas et al., 2008). Based on studies in clinically healthy animals, lying behavior has a high priority in cattle (Jensen et al., 2005; Munksgaard et al., 2005). Because lying down is a prerequisite thereof, it is considered an important behavioral indicator during evaluation of diseases in terms of animal welfare.

Thoenen et al. (2004) described a model in which alimentary oligofructose overload was used to induce development of ruminal acidosis followed by clinical signs of lameness. This model allowed scientific study of sickness behavior in cattle and was used in the present experiment. The aim was to describe rising and lying down behavior of heifers in the initial 5 d after alimentary oligofructose overload to 1) acquire information about consequences of the experimental model in terms of animal behavior; and 2) contribute to the evaluation of the influence of disease on cattle welfare. The present behavioral study was conducted as part of a larger experiment undertaken to examine the pathophysiology of lameness in heifers given an oral bolus of oligofructose (Danscher et al., 2009).

Eight clinically healthy, nonpregnant, Danish Holstein-Friesian heifers with a BW of 276 to 443 kg (371 ± 55 kg, mean $\pm$ SE) and aged 15 to 28 mo (22.8 ± 4.8 mo, mean $\pm$ SE) from a commercial herd were used. Before the experiment, the heifers were managed in groups of 4 to 12 on deep bedding for at least 6 mo. For practical reasons the animals were allocated randomly to 2 blocks of 4 animals. The first block took place in June 2006 and the second in August 2006.

During the experiment the heifers of each block were kept in tie stalls in a separate room in an experimental barn. The barn was lit with natural light. The heifers were moved to the experimental barn at least 2 wk before initiation of the experiment, and were kept in

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Table 1. Ethogram of behavior recorded before and after oligofructose overload

Behavior	Definition
Lying-down behavior	
Intention	The heifer stands still, swinging its head from side to side with the muzzle close to the ground. If the heifer stops this behavior and elevates its head (over 15 cm), the movement is said to be interrupted.
Kneeling	The heifer bends one foreleg and falls down on one or both knees. If the heifer stops this behavior and goes back to standing on both forelegs, the movement is said to be interrupted.
Lying	The body is resting on the ground.
Rising behavior	
Breastbone raises	The heifer moves its head and neck upwards and forward and rises on its breastbone and elbows.
Hind legs stretched	The heifer extends its head and neck upwards and gets up on its hind legs. Both hind legs are stretched.
One foreleg stretched	One foreleg is stretched.
Standing	The heifer is standing with all 4 legs on the ground.
Abnormal getting-up behavior	The heifer gets up by stretching both forelegs and thereafter the hind legs.

tie-stalls (by a chain of 88 cm tied at a height of 20 cm above ground). Each tie-stall measured 182 × 143 cm with a neck bow at a height of 93 cm. The stalls were provided with 3 to 4 cm of sawdust. Each heifer had free access to water from a water bowl and was fed grass hay twice daily at 0800 and 1500 h for ad libitum intake.

The experiment consisted of daily observations of behavior from 1 d before (d −1) until 5 d after oligofructose overload. The experiment was planned and performed in a manner to prevent any unnecessary pain and discomfort for the animals. All animals were cared for and treated according to a protocol approved by the Danish Animal Experiments Inspectorate (file no. 2006/561-1132) before performance.

The heifers were given a carbohydrate overload consisting of oligofructose (Beneo P95, Orafit Active Food Ingredients, Tienen, Belgium). Powdered oligofructose was dissolved in warm water and administered via tube-feeding directly into the reticulum/rumen using an induction bolus dose of 17 g/kg of BW. A priming dose of 5% of the main induction dose was given twice

daily for 3 d before the actual overload to gradually allow adjustment of the forestomach microbiota to the new source of carbohydrate. In the morning of d 0 (at approximately 1000 h) the fructan overloads were given to the heifers in a process taking approximately 10 min per individual. Supportive intravenous fluid therapy was given to the heifers to alleviate metabolic acidosis and dehydration. Calcium borogluconate (14 mg of Ca/mL) was given at 1.4 mL/kg of BW. Bicarbonate (84 g/L) was given in a quantity of 1.5 mL/kg of BW, and Ringer's acetate (Fresenius Kabi, Uppsala, Sweden) was given in a quantity of 15 mL/kg of BW. All heifers were given all 3 solutions at 1800 h on the day of the overload. Six hours later, all heifers received only bicarbonate and Ringer's acetate solutions.

Throughout the experiment, the heifers were subjected to daily walking and trotting by hand and were scored for gait and lameness using the scale of Sprecher et al. (1997) as well as an American Association of Equine Practitioners (www.aaep.org) scale.

The heifers were catheterized for blood sampling in the aural vein on d −7. From d −3 heifers were subject-

Table 2. Behavioral variables used to compared effects of oligofructose overload

Variable	Definition
Lying-down behavior	
Pre-lying intention, s	Duration of period from initiation of intention until the heifer either kneels or the behavior is interrupted
Kneeling-lying-down-period, s	Duration from kneeling until lying down
Total lying-down sequence, s	Duration from initiation of intention until the heifer is lying down
Frequency of lying down	The frequency of lying down behavior per recorded hour
Frequency of intention movements	The frequency of pre-lying intention movements per recorded hour
Frequency of interrupted intention movements	The frequency of interrupted pre-lying intention movements per recorded hour
Frequency of interrupted kneeling	The frequency of lying-down behavior, interrupted after kneeling
Rising behavior	
Breastbone-hind legs, s	Duration from when the breastbone is raised until both hind legs are stretched
Hind legs-foreleg, s	Duration from when the hind legs are stretched until one foreleg is stretched
Foreleg-standing, s	Duration from when one foreleg is stretched until standing
Total getting-up sequence, s	Duration from when the heifer raises its breastbone until standing
Frequency of getting-up	The frequency of getting up per recorded hour
Frequency of abnormal getting up	The frequency of abnormal getting-up behavior per recorded hour

Table 3. Lying-down and getting-up behavior of heifers before and after alimentary oligofructose overload at 1000 h on d 0¹

Behavior	Day						P-value ²
	-1	0	1	2	3	4	
Lying-down behavior							
Duration from initiation of pre-lying intention movements until kneeling, s	13 (10-16) ^a	16 (10-21) ^{ab}	21 (16-25) ^{ab}	16 (11-23) ^{ab}	28 (20-40) ^b	18 (14-38) ^{ab}	0.005
Duration from kneeling until lying down, s	6 (2-21)	5 (4-7)	5 (4-7)	6 (4-6)	6 (5-7)	6 (5-9)	NS
Duration of total lying-down sequence, s	16 (13-21) ^a	21 (14-28) ^{ab}	27 (22-33) ^b	21 (16-28) ^{ab}	36 (25-48) ^b	26 (19-45) ^{ab}	0.0001
Frequency of lying down per hour	0.4 ± 0.1	0.4 ± 0.2	0.4 ± 0.2	0.5 ± 0.3	0.4 ± 0.2	0.4 ± 0.1	NS
Frequency of bouts of intention movements per hour	0.7 ± 0.3	1.0 ± 0.6	0.8 ± 0.3	0.7 ± 0.4	0.7 ± 0.3	0.5 ± 0.3	NS
Frequency of interrupted bouts of intention movements per hour	0.1 ± 0.03	0.3 ± 0.2	0.3 ± 0.3	0.2 ± 0.2	0.1 ± 0.1	0.2 ± 0.2	NS
Frequency of interrupted kneeling, total	0	3	2	1	0	0	NT
Rising behavior							
Duration from breastbone raised until hind legs are stretched, s	2 (2-3) ^a	2 (1-3) ^{ab}	3 (2-5) ^b	3 (2-3) ^b	3 (2-3) ^b	2 (1-3) ^{ab}	0.068
Duration from hind legs stretch until 1 foreleg is stretched, s	1 (1-2)	1 (1-1)	1 (1-2)	1 (1-1)	1 (1-3)	1 (1-2)	NS
Duration from 1 foreleg stretch until standing, s	1 (1-1)	1 (1-1)	1 (1-2)	1 (0-1)	1 (1-1)	1 (1-1)	NS
Duration from breastbone being raised until 1 foreleg is stretched, s	3 (3-4)	2 (2-4)	5 (4-6)	4 (3-5)	4 (3-6)	4 (3-5)	NS
Duration from when the breastbone is raised until standing, s	4 ± 1	4 ± 2	7 ± 3	5 ± 2	5 ± 2	5 ± 1	NS
Frequency of total getting-up sequence per hour	0.4 ± 0.1 ^{ab}	0.3 ± 0.2 ^{ab}	0.2 ± 0.2 ^a	0.4 ± 0.2 ^b	0.3 ± 0.1 ^{ab}	0.3 ± 0.2 ^{ab}	0.053
Abnormal getting-up periods, total	1	0	1	1	0	0	NT

^{a,b}Values in rows with different superscript letters indicate $P < 0.10$; the exact P -value is noted.¹The control day (d -1) started 24 h before the overload. Data are presented as means ± SE or medians followed by 25 to 75% quartiles in brackets.²NS = P -value > 0.10. NT = not tested.

ed to clinical and physiological examinations according to a predefined protocol. Examinations were carried out every 24 h until the carbohydrate overload, after which examinations took place every 6 h for the next 48 h. From d 2 to 5, the heifers were examined every 24 h. Two individuals handled the heifers at examination times. Further details of the main experiment are described by Danscher et al. (2009).

All occurrences of rising and lying-down behavior were recorded using time-lapse video recording. An ethogram of the recorded behavior is shown in Table 1. Recording was only possible during daylight (from approximately 0700 to 2000 h), and all periods when research personnel were present within the stalls were excluded, leading to a mean recording period of 10.5 h/d (range: 4.8 to 12.8 h/d). Calculated behavioral variables are in Table 2.

One heifer's reaction to the oligofructose overload was markedly more severe than that of the others. Despite intensive aggressive supportive therapy and close observation, she died during the experiment and data from this animal were omitted. The duration of rising behavior was analyzed based on data from 5 heifers because no spontaneous rising behaviors were recorded from 2 of the heifers on d 1 and 3, respectively. Data from all 7 heifers were used in the analysis of the frequency of rising. Interrupted kneeling and abnormal lying-down and rising behavior were not treated statistically because of very low representation in the data set.

Because of the variable registration periods all frequencies were calculated as occurrence per hour. The oligofructose bolus was given at 1000 h on d 0 and each 24-h period (day) started at 1000 h, so that the control day, by definition, was initiated 24 h before the overload. Differences between days were tested using one-way repeated measures ANOVA (Sigmastat, Systat Software, version 3.1, San Jose, CA). The results are presented as means ± SE. In cases of nonnormality, Friedman repeated measures ANOVA on ranks (Sigmastat, Systat Software, version 3.1) was used and the results given as medians followed by 25 to 75% quartiles. For both analyses, the Tukey test was used for all pairwise comparisons of the means or mean ranks of the treatment groups.

Spearman rank correlations of SAS (Version 8e, SAS Institute Inc., Cary, NC) were used to test for correlations between selected clinical and physiological traits for ruminal acidosis and lameness and 1) the duration of pre-lying intention movements; and 2) the duration of lying-down behavior on d 1 and 3 (days differing from control period). The selected variables were rumen pH and contractions, rectal temperature, temperature of coronary band, and lameness scores.

No differences between the days were found for the frequencies of lying-down events (overall mean: $0.4 \pm 0.2/\text{h}$). The duration of the total lying-down sequence was affected by treatment leading to a longer duration on d 1 compared with d -1 [27 (22 to 23) vs. 16 (13 to 21) s, respectively, $P < 0.001$; Table 3], and a longer duration of lying-down on d 3 than on d -1 [36 (25 to 48) vs. 16 (13 to 21) s, respectively, $P < 0.001$; Table 3], but the duration of the last part of lying-down (from kneeling to recumbency) did not differ between the days (overall mean: 7 ± 5 s). No differences were found between the days for the frequency of pre-lying intention movements (overall mean: $0.7 \pm 0.4/\text{h}$) or the frequency of interrupted unsuccessful intention movements (overall mean: $0.2 \pm 0.2/\text{h}$). But the heifers showed longer duration of pre-lying intention movements on d 3 compared d -1 [28 (20 to 40) vs. 16 (10 to 16) s, respectively, $P = 0.005$; Table 3]. A tendency was found for the frequency of rising to be higher on d 2 than on d 1 ($0.4 \pm 0.2/\text{h}$ vs. $0.2 \pm 0.2/\text{h}$, respectively; $P = 0.053$, Table 3), but no differences were found for the duration of the rising sequence (overall mean: 9 ± 12 s). However, the duration of the first phase of the behavior (from breastbone rises and until hind legs are stretched) tended to be longer on d 1, 2, and 3 than on d -1 ($P = 0.068$; Table 3). No significant correlations between clinical or physiological traits and behavior were found.

The results of the present experiment suggest that lying motivation of heifers was thwarted during disease, as indicated by the longer duration of pre-lying intention movements on d 3 than on d -1 and a longer duration of lying down on d 1 and 3 compared with d -1. It is known that rising and lying-down behavior of cattle can be affected by housing (Lidfors, 1989; Krohn and Munksgaard, 1993), but this is one of the first descriptions of the influence of disease on this behavior. Krohn and Munksgaard (1993) showed that the lying and rising behavior of healthy cows in tie-stalls was different from the behavior of cows in loose housing systems, as the tied cows used more time to lie down and had

longer periods of pre-lying intention movements. The present results were obtained with a limited number of animals, but suggest that oligofructose overload can change the behavior comparably. Both metabolic and leg problems appear to lead to a thwarting of the lying-down motivation in cattle and potentially prevent the animals from lying down. This study, however, did not investigate the existence of discomfort or pain.

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