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Effect of rank, group size and number of group changes ante partum on the occurrence of milk ejection disorders in primiparous cows – a field study

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Abstract

In the present study causes for the development of milk ejection disorders (MED) in primiparous cows of two herds were investigated. We hypothesised that MED are provoked by social problems within the herd that appear close to calving. Data were collected on 67 heifers of herd A and 60 heifers of herd B. In herd A, heifers were introduced into existing groups of cows 2-4 weeks prior to calving. They were kept in a group housing system with deep bedding. All animals were hornless. In herd B, heifers were kept separated from the cows from 2-4 weeks a.p. in a three-row cubicle barn. In contrast to herd A, all animals had horns.

The animals´ reactions regarding the milking process and MED were analysed during the first four milkings p.p. We evaluated the correlation between social rank, group size and frequency of group changes in the antepartal period and the occurrence of MED.

12 % of the animals in herd A and 47 % of the animals in herd B showed MED. In herd A, low-ranking heifers had significantly more often MED than higher-ranking heifers. A correlation between low rank, increasing strain due to rank order fights and increasing frequency of MED can be assumed here. In herd B, a correlation between stocking rate and frequency of MED could be observed.

Keywords: dairy cow, heifer, milk ejection, rank, group constellation, social stress, housing conditions

Zusammenfassung

Die Auswirkung von Rangstatus, Gruppengröße und Anzahl der Gruppenwechsel ante partum auf das Auftreten von Milchejektionsstörungen bei primiparen Kühen – Ergebnisse einer Feldstudie

In vorliegender Studie wurden die Ursachen für das Auftreten von Milchejektionsstörungen (MES) bei primiparen Kühen in zwei Produktionsherden untersucht. Die Hypothese war, dass die MES durch vor der Kalbung auftretende soziale Probleme innerhalb der Tiergruppen ausgelöst werden. In die Untersuchung waren 67 Färsen der Herde A und 60 der Herde B einbezogen. In Herde A wurden die Färsen ca. 2-4 Wochen vor der Kalbung in die Kuhgruppen eingegliedert und in Gruppenboxen auf Tiefstreu gehalten. Alle Tiere waren hornlos. In Herde B waren die Färsen ab 2-4 Wochen a.p. getrennt von den Kühen in einem 3-reihigen

Hochliegeboxenlaufstall untergebracht. Im Gegensatz zur Herde A waren alle Tiere behornt.

Die Analyse der Tierreaktion hinsichtlich des Melkverlaufes und die Bewertung von MES erfolgten während der ersten 4 Melkungen p.p. Ausgewertet wurden die Zusammenhänge zwischen dem sozialen Rang des Tieres, der Gruppengröße sowie der Häufigkeit von Gruppenwechseln im Zeitraum vor der Abkalbung und dem Auftreten von MES.

12 % der Tiere in Herde A und 47 % der Tiere in Herde B wiesen eine MES auf. In Herde A hatten niederrangige Jungkühe signifikant häufiger MES, als hochrangige. Hier kann ein Zusammenhang zwischen niederem Rang, ansteigender Beanspruchung durch Rangkämpfe und einer zunehmenden Häufigkeit von MES vermutet werden. In Herde B war ein Zusammenhang zwischen Belegungsdichte und der Häufigkeit von MES festzustellen.

Schlüsselwörter: Milchkuh, Färse, Milchejektion, Milchejektionsstörung, Rang, Gruppierung, Sozialstress, Haltungsbedingungen

Introduction

Since 1996 an increase in milk ejection disorders (MED) in herds with Holstein Frisian cows was observed in Saxony, Germany. Primiparous cows were more often affected than multiparous. The reasons for this increase were unknown (Schulz & Petzold 2000, Thümmler 2003, Geidel *et al.* 2005).

MED is the complete failure or incomplete process of milk ejection during proper milking, sucking of the calf or other adequate stimulation of the teat receptors (Mielke & Brabandt 1963). Numerous studies were conducted to identify the physiological causes for MED (e.g. Kokorina 1956 and 1959, Mielke & Brabandt 1963, Mielke 1992, Bruckmaier *et al.* 1992, Kraetzl *et al.* 1999 and 2001a,b, Wellnitz *et al.* 1999, Inderwies *et al.* 2001, Macuhova *et al.* 2002). However, the physiological interactions are not yet fully understood.

Less often housing conditions causing MED under production conditions were investigated (Thümmler 2003, Geidel *et al.* 2005, Schulz & Petzold 2000, Belo *et al.* 2009). In these studies tendencies were identified, but no causality for individual animals could be determined. Furthermore, the definition of MED-status was made by the herd managers and was thus not consistent within one study.

A third group of studies dealt with MED that were generated under experimental conditions (Tröger 1978, Lefcourt *et al.* 1985, Kraetzl *et al.* 2001a) or under standardised conditions similar to production (Bruckmaier *et al.* 1993, Tancin *et al.* 2001, Kraetzl *et al.* 2001 b, Mačuhová *et al.* 2002, Das & Das 2004, Dzidic *et al.* 2004). MED was triggered by medication or otherwise mostly by acute emotional stress. Such stressors were for example a divergent milking routine, unknown environment or a late separation of the calf. All these stressors have a peripartal and a postpartal effect and can also occur under production conditions. However, based on these studies no sufficient explanation for the increased incidence of MED-cases in Saxon herds could be found.

The aim of this study was therefore to identify concrete reasons for MED in primiparous cows of two production herds. Other studies conducted at the same time showed that in the examined herds the age at first calving, the duration of calving, stillbirth, the person of the milker at first milking and medication with oxytocin for birth relief could be ruled out as a reason for MED (Heidig 2007). Therefore we hypothesised that emotional stress before calving does lead to the postpartal development of MED.

It is known that the integration of heifers into the group of nonlactating cows is an emotional strain for the animals. The arrangement of new groups, especially when unfamiliar animals are mingled, leads to an increase in rank order fights (z.B. Raussi 2005, Kucevic *et al.* 2010). The hierarchy is stable after approximately three weeks (Brakel & Leis 1976). After integration into existing groups cows can show decreasing milk yield (Hasegawa *et al.* 1997), shorter feeding and resting times (Hasegawa *et al.* 1997) and increased blood cortisol, albumin and urea (Gupta *et al.* 2005).

Regrouping can lead to loss of rank, especially when the animal held a high rank until that time (Hasegawa *et al.* 1997). Loss of rank is a severe strain, which can lead to decreasing milk yield (Hasegawa *et al.* 1997) and fertility disorders (Dobson *et al.* 2000). When heifers are introduced into a group of multiparous cows, a loss of rank for the heifer can be expected, because due their higher body weight cows are usually dominant over heifers (Arave *et al.* 1977, Baehr 1983). Especially low-ranking animals suffer from social problems within a group. The mere presence of a higher-ranking conspecific can cause measurable physiological stress reactions (Lefcourt *et al.* 1999).

Social conflicts within animal groups can be triggered or reinforced by housing and management conditions. An insufficient number of feeding places or exceeding stocking rates lead to an increase in aggression towards low-ranking animals and induce temporal and spatial changes of behaviour in these animals (Gygax *et al.* 2009b, Huzzey *et al.* 2006).

Insufficient adaption periods increase the sensitivity towards stress (Takeda *et al.* 2003). Frequent regrouping decreases the resting time, induces an increase in activity and therefore results in constant restlessness within the group (Raussi *et al.* 2005). Regrouping of single cows leads to more agonistic behaviour against newly introduced animals than the simultaneous integration of several familiar cows (Neisen *et al.* 2009).

In the following it will be investigated if these associations accounted for by literature are the reason for the increased occurrence of MED in production herds.

Material and methods

Housing conditions

Table 1
Housing conditions of heavily pregnant heifers a.p.

	Herd A	Herd B
Integration of heifers in the production barn	2-4 weeks before calving	2-4 weeks before calving
Stabling	straw bedding box (6×13 m)	cubicle barn (44 cubicles, rubber mattresses) with narrow alleys (2.65 m and 2.10 m) with blind ends
Grouping	together with unknown multiparous cows	only primiparous cows
Regroupings	2× per week	1× per week
Horns	all animals without horns	all animals with horns

The experiments were conducted on two farms with an increased occurrence of MED in primiparous cows. Both farms kept the heifers in separated rearing barns and moved them into a special section of the dairy barns shortly before calving.

Table 1 describes the housing conditions of the heavily pregnant heifers after moving. It needs to be mentioned that all animals of herd B had horns.

Definition of MED-status

To define the MED-status of every cow the milking process was recorded from the first milking after calving. In the first instance, the milk removal was performed according to the milking routine of the farm (foremilking, cleaning of the udder, automatic stimulation). After finishing the milking, the milker recorded the quantity of milk and estimated the filling degree of the udder. If the udder was not fully milked out, milk ejection was tried to be initiated by different means (rectal stimulation, oxytocin treatment). Afterwards the animal was milked a second time. The treatment, the quantity of milk gained at the second milking and the filling degree of the udder were recorded again. Recording ended after the 4th milking. The definition of milking MED-status was made according to the scheme shown in Figure 1. It was always done by the same person.

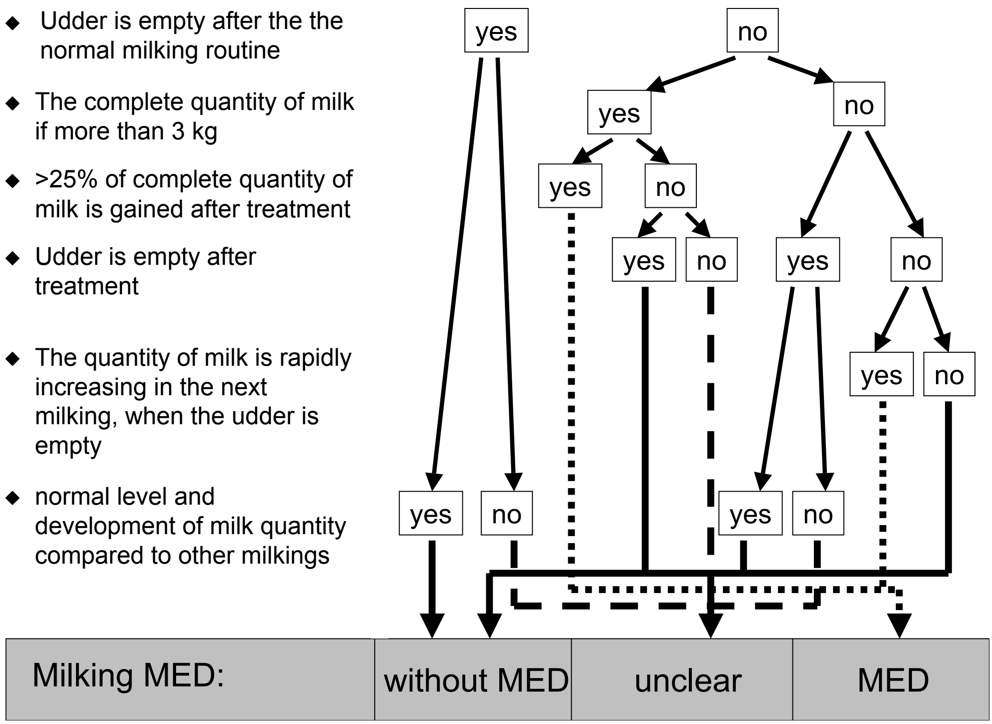

Figure 1
Method of defining the MED-status of a single milking (»Milking MED«), based on the protocol of the milker.

Based on the definition of the MED-status of a single milking event (»Milking MED«), the MED-status of the animal (Animal MED) was determined, following the scheme shown in Table 2. The Animal MED score was used for further calculation.

Table 2
Method of defining the MED-status of an animal (»Animal MED«)

Animal MED =	if	Milking MED
Score 1 (negativ)		No MED at all milkings
Score 2 (not clear)		MED at one or more milkings unclear, no MED at all other milkings
Score 3 (positiv)		MED at one or more milkings

Definition and database of the property rank a.p.

All animals were videotaped during the whole day from the moment of introduction into the group until calving (herd A 172 observation days, herd B 120 observation days). On the first two or three days after integration into the group the number and outcome of agonistic interactions as well the partner of interaction was registered. All those interactions were defined as agonistic, during which the submissive animal showed clear reactions of inferiority or flight. According to the results of the agonistic interactions of all group members a graphic overview of the hierarchy within the group was generated. The assignment to the rank categories »high-ranking«, »medium ranking« and »low-ranking« was made based on the formation of subgroups (Figure 2).

The rank category represents the rank of the heifer in relation to the other heifers of the group. Animals that did not show any interactions with group mates were recorded as »poor in interaction« and not further analysed.

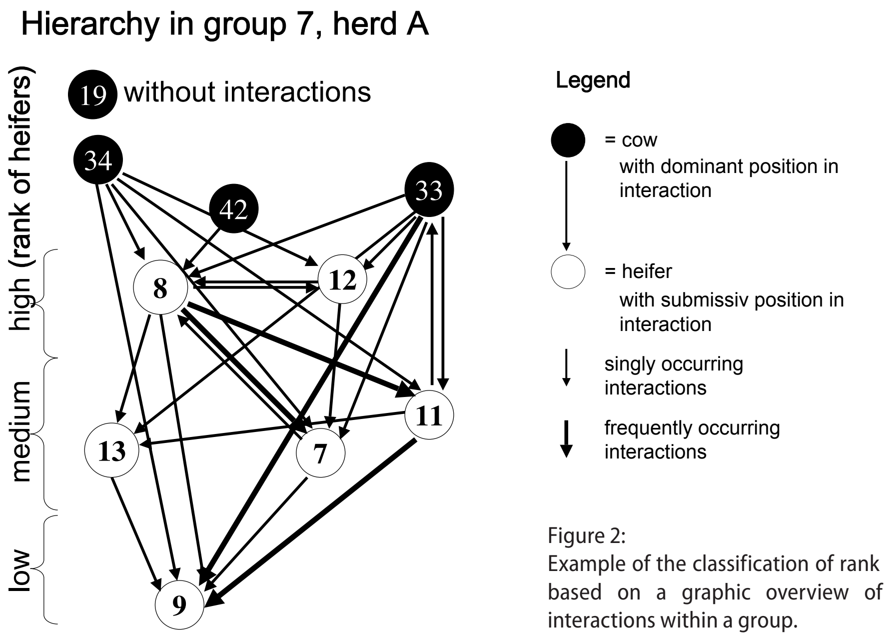

Figure 2:
Example of the classification of rank categories based on a graphic overview of agonistic interactions within a group.

The assessment of ranks was redone for every new group composition. Therefore some animals were allocated to several different rank categories. The rank occupied most often was used for further analysis.

The classification of ranks was not done by using rank indices (e.g. Sambraus 1978), because despite the small groups not all animals were interacting and unclear dyads led to the misclassification of some animals.

Table 3
Number of animals per rank category

	Included in the analysis Rank category			total	Not included Poor in interaction
	low	medium	high		
Farm A	13	38	16	67	6
Farm B	10	45	13	68	3

Definition and database of the property group size a.p.

In both herds, new animals were integrated into existing groups. Every change of group composition caused by the introduction of at least one new animal was recorded as new group. The number of animals per group was assessed.

Table 4
Number of evaluated groups and group size

		Herd A	Herd B
Number of groups included in the analysis	n	28	12
Number of animals/ group (cows and heifers)	mean	8.5	-
	min	4	-
	max	12	-
	SD	2.15	
Number of heifers/ group	mean	3.5	14.3
	min	1	6
	max	9	28
	SD	2.27	5.82
Proportion of heifers with Animal MED=2 of heifers/group in %	mean	0.0	16.9
	min	0.0	0.0
	max	0.0	45.5
	SD	0.0	16.25

The proportion of animals per group with unclear MED status (Animal MED=2) varies widely. Therefore the percentage of Animal MED=3 per group is not a suitable measure for the frequency of MED. We used the MED-index instead:

$$MED\text{-}Index = \frac{\text{number of heifers in the group with Animal } MED = 3}{\text{number of heifers in the group with Animal } MED = 1} \quad (1)$$

A high number of animals with Animal MED=2 does thus not lead to a low frequency of MED, as it would be when percentages are used.

Definition and database of the property number of group changes a.p.

An animal changes the group if
- it is integrated into an existing group
- the group the animal is belonging to changes its composition because new animals are introduced

The number of group changes per animal was recorded. Animals were included in the analysis if all group changes of the animal were recorded and the MED-status was not unclear. The obtained data for this property is shown in Table 5. Sample size for more than three group changes was insufficient for statistical analysis.

Table 5
Number of group changes per animal a.p.

Herd		Number of group changes							Total
		0	1	2	3	4	5	6	
A	Number of animals	10	18	14	12	3	2	1	60
B	Number of animals	8	20	24	3	-	-	-	55

Each group was observed for three hours per day too evaluate the circadian rhythm of the animals and synchronicity of behaviour. The number of animals per group that were lying, standing (in the cubicle or alley including those animals walking) or feeding was recorded. The recording started on the third day after the group change. Only days with undisturbed daily routine were analysed.

Statistical analysis

For the statistical analysis of the data, the software programmes SPSS Statistics 17.0 (SPSS Inc., Chicago, IL, USA) and Microsoft Excel (Microsoft Corporation, Redmond, WA, USA) were used. The properties group size, number of group changes and number of agonistic interactions are approximate normal distributed. The properties animal MED and rank category are available as classes, therefore the procedure of Spearman-Rho (bivariate, two-sided) was used for the calculation of the correlation coefficient. Comparison of means of the metric properties was done using the standard error after Clauß & Ebner (1977) with the formula:

$$SE = \frac{Standarddeviation}{\sqrt{n}} \qquad (2)$$

The comparison of percentages was done based on the standard error after Ferguson 1959 (quoted after Clauß & Ebner 1977):

$$SE = \frac{p(1-p)}{n}, p = percentage\ to\ be\ tested\ in\ decimal\ notation \qquad (3)$$

The consideration of relations between the examined factors and the frequency of MED is based on a mono-causal model. The use of a multivariate model for evaluation was not considered to be advisable because of the small database, the fact that not all properties were assessed for every animal and the disadvantageous distribution of the data among the statistical subgroups.

Results

Relationship between the rank of heifers in the groups a.p. and the occurrence of MED

12 % of the examined heifers in herd A and 47 % in herd B received the Animal MED score 3. As shown in Table 6, the distribution of the animals to the rank categories is comparable in both herds. In herd A a significant correlation can be seen: the lower the rank of an animal, the more frequent it suffers from MED. In herd B no correlation between rank and MED was found.

Table 6
Relationship between the rank of heifers a.p. and the occurrence of MED

		Herd A				Herd B			
			Rank				Rank		
		Total	Low	Medium	High	Total	Low	Medium	High
Included animals	n	67	13	38	16	60	13	33	14
Thereof with	n	8	3	5	0	28	6	14	7
Animal MED = 3	% of incl. animals	11.9	23.1[b]	13.2[b]	0.0[a]	46.7	46.2[c]	42.4[c]	50.0[c]
	SE%	3.96	11.69	5.48	0.00	5.32	11.44	7.21	10.91

Herd A: heifers mixed with cows, Herd B: heifers alone in the group, [a,b,c]Different letters mean significantly different frequencies. Significance was tested after Ferguson (1959).

The reason for this correlation is supposed to be the unequal strain caused by rank fights, which is depending on the individual rank. As expected, low-ranking animals of both herds showed a significantly higher number of lost agonistic interactions than high-ranking animals (Table 7).

Table 7
Mean number of lost agonistic interactions/ day depending on rank

Number of lost interactions per day	Low ranking heifers		High ranking heifers	
	$\bar{x}$	SE	$\bar{x}$	SE
Herd A				
Interaction with multiparous cows	2.0	0.55	1.5	0.35
Interaction with other heifers	2.3	0.39	0.1	0.05
Total	4.3	0.63	1.6	0.35
Herd B				
Interaction with other heifers	3.2	0.42	0.7	0.10

More lost interactions were recorded in herd A than in herd B, independent of rank category. It must be noticed, that in herd A low-ranking animals were rather dominated by other heifers than by cows.

Relationship between group size a.p. and the occurrence of MED p.p.

Group size strongly varied in both herds (Figure 3 and Figure 4). If we calculate a space requirement of 8 m^2 per animal in an unstructured group calving pen (KTBL 2008), all groups in herd A with more than 10 animals were overstocked. In herd B, the housing compartment contained 44 cubicles, which means that maximum 64 % of the capacity was used.

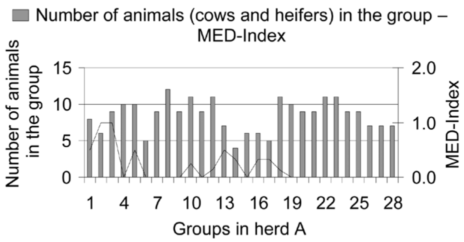

Figure 3
Relationship between group size and the occurrence of MED in herd A (r=−0.218; *P*=0.264)

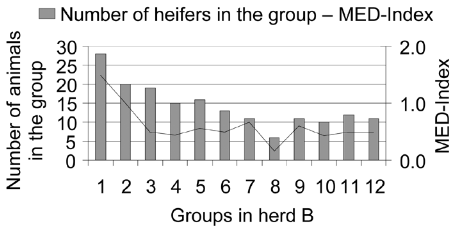

Figure 4
Relationship between group size and the occurrence of MED in herd B (r=0.599; *P*=0.040)

However, no correlation between group size and MED-index could be found in herd A, while there was a close correlation between these parameters of r=0.599 (*P*=0.040) in herd B (Figure 3, Figure 4). The reason for the close correlation between group size and MED-index in herd B was not the insufficient supply of lying cubicles or total cubicle area. In fact, the very narrow or dead ending passageways are the major problem (width of passageways: in the lying area 2.10 m; in the feeding area: 2.65 m). The video recordings clearly show that the heifers have problems changing position within the housing compartment. They fear a meeting with a

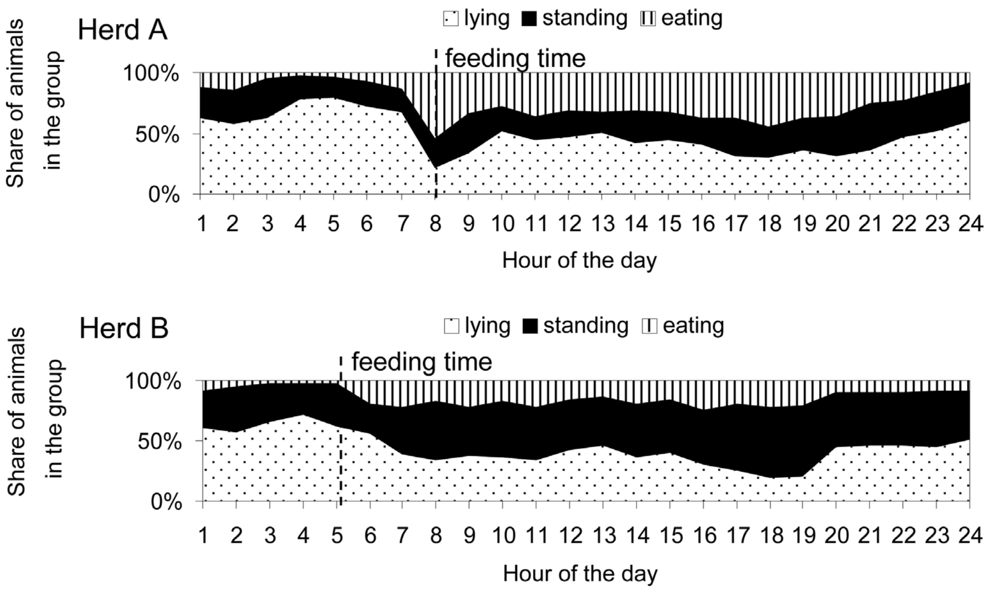

Figure 5
Daily rhythm of group activity in herd A and herd B (means per hour over all days of the observation period)

higher-ranking animal in narrow passageways and at the feeder, because keeping individual distance is not possible. The carrying of horns does further intensify the situation.

The activity rhythm of both herds was compared to evaluate whether the shortage of space in the walking area of herd B is a strain to which animals react with behavioural changes (Figure 5).

The circadian rhythm of the animals in herd A is typical for dairy cows, with a distinct resting period at night and a less distinct in the early afternoon as well as active phases during the morning and evening hours (Sambraus 1978, Bogner & Grauvogel 1984, Tilger 2005). After the distribution of feed most animals go feeding together. A small proportion of the group is standing. Herd B shows a very different behaviour. The animals of one group do not synchronize their behaviour. Even after feed distribution only a small number of animals is feeding immediately. Noticeable is the large number of standing animals during the whole day.

Relationship between the number of regroupings a.p. and the occurrence of MED p.p.

The number of animals with Animal MED 3 in herd A is significantly decreasing with increasing number of group changes per animal (Table 8).

A possible explanation can be the fact that a large number of group changes per animal is synonymous with a long duration of stay in the observed housing compartment, hence a long adaption period. In herd B no trend could be observed.

Table 8
Correlation between the number of group changes per animal a.p. and the occurrence of MED

Herd			Number of group changes			
			0	1	2	3
A	Number of animals	n	10	18	14	12
	Animals with	n	3	3	1	0
	Animal MED= 3	%	30.0^{c}	16.7^{bc}	7.1^{b}	0.0^{a}
		SE$^{\%}$	14.49	8.78	6.88	0.00
B	Number of animals	n	8	20	24	3
	Animals with	n	3	9	8	0
	Animal MED= 3	%	37.5^{b}	45.0^{b}	33.3^{b}	0.0^{a}
		SE$^{\%}$	17.12	11.12	9.62	0.00

^{a,b,c}Different letters mean significantly different frequencies. Significance was tested after Ferguson (1959).

Discussion

The results of the examined herds are diverse: In herd A, two factors led to an increased occurrence of MED: a low rank and a small number of group changes, this means a short adaption period in the new environment.

When comparing both herds, the number of agonistic interactions in all rank categories was significantly higher in herd A. The reason for that is the housing of heifers together with unfamiliar multiparous cows, which provokes strong rank order fights. Heifers that were high-ranking till that point are confronted with higher-ranking cows and must thus deal with a loss of rank. Dobson & Smith (2000) and Hasegawa *et al.* (1997) demonstrated that the decrease if individual rank is a considerable strain. In the present study dominant heifers often and apparently causeless showed aggression against submissive heifers.

Low-ranking heifers are particularly burdened by rank fights: they have to win their rank against unknown cows and are at the same time exposed to aggressive behaviour of dominant heifers. Consequential low-ranking heifers show on average a considerably increased number of lost agonistic interactions per day. In more than 50 % of these cases the rival was another heifer. Rank conflicts are a strong emotional stressor for the submissive interaction partner (Lefcourt *et al.* 1999). The increase of MED occurrence with decreasing rank seems to be a reaction to the heavy strain of rank order fights.

An earlier introduction of heifers to the new group might ease the situation, because an increasing number of regroupings and a thus prolonged adaption period for the heifers decrease the number of animals with MED. Another approach in farm A could be the separate housing of cows and heifers.

The initial situation in herd B is different: Here stable groups of heifers are formed. In the preceding housing area the animals are grouped by parity. Thus animals are already known to each other. Consequentially there is only little correlation between the number of group changes and MED: There is no familiarization period during which the rank order needs to be established. Fewer agonistic interactions are observed than in herd A. In herd B almost 40 % more lying cubicles than necessary are available to the animals. However, the proportion of animals suffering from MED p.p does increase with increasing stocking rate. The reason for that seems to be the restricted space in the walking and feeding area. Cattle require an individual distance to herd mates of 1 m to 10 m from head to head (Sambraus 1978, Kondo *et al.* 1989, Gygax *et al.* 2009). The widths of the passageways are 2.10 m in the lying area and 2.65m at the feeding table. Some passageways are blind ended.

The animals have no opportunity to behave according to their rank in the hierarchy of the group. The horns of the animals intensify this situation. Animals with horns need considerably greater individual distances and therefore broader passageways than dehorned animals. The housing of animals with horns in a barn which passageways are, according to current knowledge (KTBL 2009), too narrow even for hornless animals, as practised in herd B, must be critically assessed and is not in accordance with good farming practice.

Medium-ranking and low-ranking animals try to avoid contact with high-ranking group mates and stay in the walking area as little as possible. The high proportion of standing animals during the whole day results mainly from animals standing in the cubicles. The passageway is only entered when the passage is free. The result of this reduced mobility is a complex change in behaviour: A part of the animals postpones activities to atypical daytimes. The circadian rhythm of the group mates is therefore not longer synchronized.

Bogner & Grauvogel (1984) and Tilger (2005) demonstrated distinct resting phases of dairy cattle at night and at noon. This rhythm was expected in the observed herds, particularly because heavily pregnant heifers are not disturbed in their circadian rhythm by milking or other treatment. Also the feeding time during the 5th hour of the day corresponds with the feeding time under natural conditions during daybreak (Sambraus 1978, Bogner & Grauvogel 1984, Tilger 2005). Contrary to our expectations, the typical biphasic circadian rhythm was hardly recognisable in herd B.

According to Sambraus (1978) the lack of the species-specific circadian rhythm is a strong evidence of severely stressing environmental conditions. Because of the disturbed rhythm there is constant restlessness within the group. The increase of MED with increasing stocking rate shows that large groups intensify this problem. This situation is a strain for animals of all rank categories. Consequential, no correlation between rank and MED, but a close correlation between stocking rate und MED was found. The very large proportion of animals with MED of 46.7 % is a measure for the intensity of the strain.

The situation in herd B could be eased by the use of hornless animals. Fundamental improvement however can only be achieved by structural modification of the walking area in the compartment for heavily pregnant heifers and an adjustment of the width of the passageways to the carrying of horns.

The diverse results of the herds show that the influence of single factors is highly specific for the farms. Therefore, a generalization and transfer of the results to other farms can only be done with reservation. The present analysis of the situation in the examined herds is based on a mono-causal model and does not include interactions between the analysed factors. A verification of the found correlations under standardized conditions and with a larger database and a deeper statistical analysis would be desirable.

The hypothesis, that prenatal load factors might be responsible for MED could be confirmed for both herds. Social stressors in this period of housing seem to cause a predisposition for MED. The results suggest that farms with MED problems should in any case include the period before calving into their cause study.

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