

## Agitation behaviour and heart rate of dairy cows with and without calf-contact during different stimuli in the parlour

KATHARINA A. ZIPP<sup>1</sup>, KERSTIN BARTH<sup>2</sup>, UTE KNIERIM<sup>1</sup>

**Key words:** agitation behaviour, heart rate, calf-contact, stimuli, milking, dairy cows

### Abstract

*Three stimuli were tested to overcome problems in milk let-down in dairy cows with calf-contact. In this contribution, the effect of these stimuli during milking, on heart rate (HR) and agitation (rumination, posture of the head and behaviour during udder preparation) are presented. Olfactory (calf hair), tactile (teat massage) and acoustic (recorded calf calls) stimulation were tested on 15 dairy cows with permanent calf-contact and 22 control cows. Rumination and posture of the head were not influenced by any factor. Agitation and HR in the parlour were not affected by calf-contact. Acoustic stimulation had a decreasing effect on HR in German Holstein independent from calf-contact. German Red Pied showed more agitation behaviour and a higher HR during udder preparation compared to the German Holsteins. Heifers showed more agitation during udder preparation than pluriparous cows. The results did not indicate higher stress reactions in the parlour of dairy cows with calf-contact.*

### Introduction

In dairy production it is common that calves are separated from their mothers soon after birth. In contrast, calves are suckled for about 10 month by their mothers if there is no human intervention (Reinhardt and Reinhardt 1982). There is a growing interest in systems with prolonged cow-calf-contact, where the young is suckled by the dam and the cow is also milked. But despite a good development of calves (e.g. Roth et al. 2009) and a better social ability of calves (Wagner et al. 2010) and heifers which stayed the first weeks in life with their mothers (Wagner et al. 2012), problems with milk let-down during machine milking due to disturbed alveolar milk ejection (reviewed by Barth et al. 2008) constrain the realisation in practice. Udder preparation before milking is an adequate stimulus for oxytocin ejection in dairy cows without calf-contact, but is possibly insufficient in cows with calf contact. Moreover, an increased stress response during milking may lead to inhibition of oxytocin ejection (reviewed by Bruckmaier and Wellnitz 2008). In our project possible effects of acoustic, olfactory and tactile stimulation on milkability of dairy cows with permanent and without calf-contact were investigated. In the following effects of these stimuli on the stress indicators agitation behaviour and heart rate (HR) in the parlour are presented.

### Animals, materials and methods

The experiment was carried out between November 2010 and May 2011 at the Thuenen-Institute of Organic Farming (Germany). There were two herds of 45-48 horned dairy cows of the breeds German Holstein and German Red Pied. They were housed separately but under same conditions (for details Wagner et al. 2012). Fifteen cows had contact to their calf for the first 12 weeks post partum (p.p.) (contact group). They stayed for five days after parturition together with their calf in a calving pen, which they only left for milking and feeding. Thereafter the dairy cows were integrated into their herd. Calves could freely move between the calves' area and the lying area of the cows. Cows of the control group (n=22) were separated within 12 hours p.p.. All cows were milked twice daily in a 2x4 tandem milking parlour (GEA, Boenen, Germany): 38 kPa milking vacuum, 40 s vibration stimulation, automatic stripping and cluster removal. The milking routine, used as control treatment, consisted of manual premilking and cleaning of the udder (approx. 20 s), attaching and positioning of milking cluster, as well as manually check of the udder after cluster removal and if necessary milking cluster was attached again. During the measurements animals were milked by one milker who was familiar to the cows. During olfactory stimulation cows were confronted with hair of the anogenital region, hind legs and tail of their own calf. The hair (approx. 0.8 g per milking) was filled in a thin cloth bag, sprayed with distilled water before milking and put in a stainless steel punnet, installed at the head region of the milking box. For tactile stimulation, after premilking and cleaning of the udder, teats were

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<sup>1</sup> University of Kassel, Farm Animal Behaviour and Husbandry Section, Germany, [www.uni-kassel.de/agrar/fnt](http://www.uni-kassel.de/agrar/fnt), [zipp@uni-kassel.de](mailto:zipp@uni-kassel.de), [knierim@wiz.uni-kassel.de](mailto:knierim@wiz.uni-kassel.de)

<sup>2</sup> Thuenen-Institute of Organic Farming, Germany, [www.ti.bund.de](http://www.ti.bund.de), [kerstin.barth@ti.bund.de](mailto:kerstin.barth@ti.bund.de)

massaged in a standardised way. Udder preparation and massage took 60 s together and no automatic vibration stimulation was applied. Acoustic stimulation consisted of calf-call play-backs from ten 2-12 weeks old calves kept on the farm of the University of Kassel, approx. 10 h after their last milk feeding. Between the fifth and seventh week of lactation the three stimuli were applied in the parlour. In each week at four consecutive milkings each one stimuli and routine milking as a control was carried out. Between different treatments, there were one to two days of routine milking without data recording. The order of stimulation and routine milking during the week was as far as possible randomised. Six cows of the control group had stillborn calves or their young died soon after birth, so no olfactory stimulation was possible. Further, missing data occurred due to udder and technical problems during videotaping or heart rate measurement.

### Agitation behaviour

Behaviour during milking was videotaped (Axis 221 network-cameras, 640x480 pi zoom lens, Axis Communications, Lund, Sweden) and analysed with ObserverXT<sup>®</sup> (Noldus Information Technology, Wageningen, The Netherlands) by two observers. Inter- and intra-observer reliability was acceptable before, during and after the observation phase (Cohen's Kappa Coefficient >0.65). Graphically no influence of daytime could be detected. Thus medians for each animal per treatment were calculated and if applicable rounded to 1. Data were analysed with R 2.15.0 (R Foundation for Statistical Computing) with a generalized linear mixed effects model for binomial data (Bates et al. 2012) with the fixed factors treatment, calf-contact, breed, parity (primiparous vs. pluriparous), and animal as random factor. It was checked that there were no interactions. Through contrasts the routine milking was compared with each stimulus.

parameter	definition
behaviour during udder preparation	0=calm or max. three steps 1=more than three steps and/or kicks
ruminating at milking	0=no ruminating 1=ruminating
posture of the head at milking	0=no or seldom changes of position 1=frequent changes of position

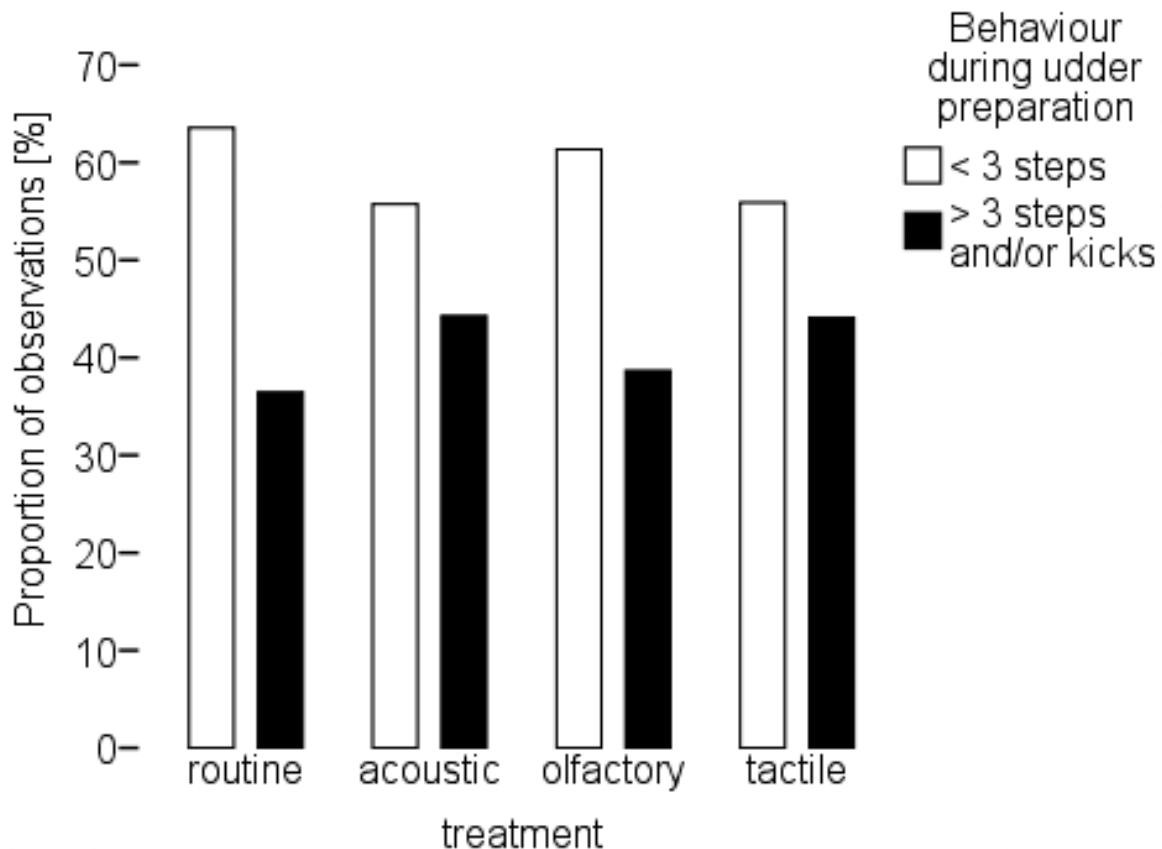
**Table 1: Definition of behaviours observed in the parlour**

### Heart rate

During the fourth week of lactation animals were habituated to wear the belts for HR measurement. Only during morning milkings HR was measured with Polar S810i and RS800CX (Polar Electro Oey, Kempele, Finland) in beat-to-beat interval. The HR equipment (without monitor-watch) was attached after the afternoon milkings, when the cows were locked at the feeding rack. The monitor-watch was activated and fastened at the belt after a cow had entered the milking box in the morning. Approximately 5 minutes after this manipulation the milking started. Sequences for analysis were selected and corrected with the software Polar Trainer 5.0. Data where more than 5% should be corrected were excluded from analysis. Due to missing values HR of 15 contact and 20 control cows was analysed. For the phases 'udder preparation', 'milking' and 'preparation+milking' means for every milking were calculated and these were averaged for each treatment per cow. The HR-differences between treatments were analysed separately for the two breeds with Friedman's ANOVA and Wilcoxon signed rank test as post-hoc test comparing the stimuli with routine milking. The differences in breed were analysed with Mann-Whitney U-test in SPSS (IBM<sup>®</sup> SPSS Statistics 20.0).

## Results and Discussion

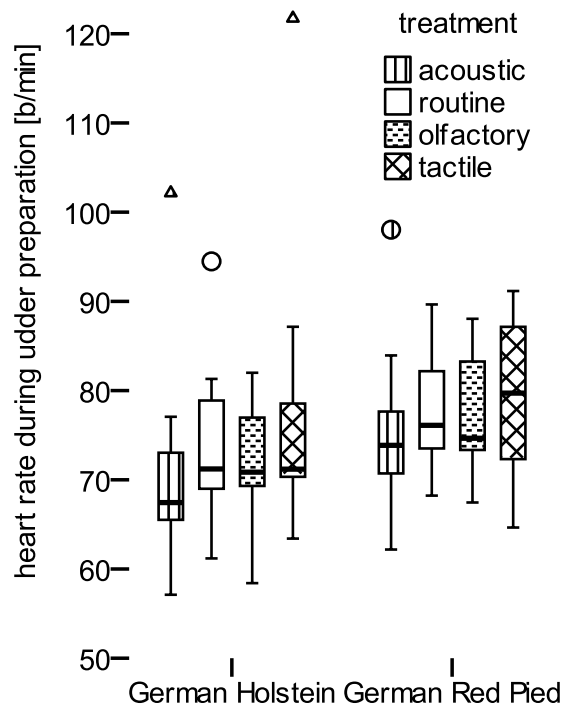
There was no significant impact of calf-contact on the investigated variables. Posture of the head and rumination were not influenced by any factor. HR during acoustic stimulation was lower than at routine milking ( $p < 0.01$ ), but only in German Holstein ( $n = 14$ ); there was no significant difference in HR between milkings with olfactory or tactile stimulation and the control (Fig. 2). Thus calf calls had a more relaxing influence on German Holstein cows with and without calf-contact. Concerning agitation behaviour during udder preparation no effect of the different stimuli was found (Fig. 1). German Red Pied ( $n = 14-15$ , median = 78.5 b/min) had in tendency a higher HR during udder preparation than German Holstein ( $n = 21-22$ , median = 72.3 b/min,  $p = 0.052$ ) and showed more agitation during this phase (57.4% vs. 27.5% of observations agitated,  $p = 0.003$ ). In both breeds HR decreased during milking (German Red Pied: median = 74.7 b/min, German Holstein: median = 70.5 b/min) and there was no difference between breeds any longer. Beside differences in reaction towards human contact between breeds, measurements of activity in the stable (unpublished Barth 2013) point to a greater reactivity in German Red Pied in general. Also agonistic behaviour in the waiting area could have influenced the data. Heifers ( $n = 9$ ) were agitated during udder preparation in 65.7% of observations which was significantly more often than in pluriparous animals ( $n = 28$ , 30.3%,  $p = 0.003$ ). HR did not significantly differ between parities, but as HR is influenced by body weight (Hagen et al. 2005), which was not considered in the analysis, possible differences in HR between parities could be concealed.



**Figure 1: Relative frequency of agitation behaviour at udder preparation according to treatment**

## Suggestions to tackle with the future challenges of organic animal husbandry

Cows with calf-contact did not show more agitation behaviour in the parlour. The effect of the stimuli on the parameters of the project in total (milk yield, milk flow, HR, heart rate variability, agitation in the parlour) must be considered before final recommendations can be given. Acoustic stimulation had beneficial influence on HR independent from calf-contact.



**Figure 2: Heart rate (HR) at udder preparation in b/min during different treatments for the two breeds. Box-plots with median (lines in boxes), 25 and 75% quartiles (boxes), 10 and 90% range (whiskers), outliers (dots) and extremes (triangle)**

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