

Institute of Agricultural Engineering
University of Hohenheim
Livestock Systems Engineering (440b)
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Institute for Sustainability Sciences ISS
Work, Buildings and System Evaluation
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**Investigating dairy cow welfare by optimizing pulsation cycles and improving activity
measurements during milking from a technical perspective**

Dissertation

submitted in fulfillment of the regulations to acquire the degree

"Doktor der Agrarwissenschaften"

(Dr.sc.agr. in Agricultural Sciences)

to the

Faculty of Agricultural Sciences

presented by

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Dachau

2016

This thesis was accepted as a doctoral thesis (Dissertation) in fulfillment of the regulations to acquire the doctoral degree "Doktor der Agrarwissenschaften" by the Faculty of Agricultural Sciences at the University of Hohenheim on the 18th of September 2015.

Date of the oral examination: 2nd of December 2015

Examination Committee

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Published by the author: Franziska E. Blümel
Source of supply: Universität Hohenheim
Institut für Agrartechnik -440-
Garbenstraße 9
D-70599 Stuttgart

In memory of my grandfather

Thomas Bluemel, Agriculturist

(1915 - 2000)



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List of abbreviations

a.m.	ante meridiem
a-phase	opening phase
Acc.	acceleration
b-phase	open phase
BIMO	bimodal milk flow
c-phase	closing phase
d-phase	closed phase
dB	decibel
DIM	days in milk
DP	decreasing phase
Duration DP	time of decreasing phase
Duration MMT	time of main milking process by machine
Duration OP	time of over-milking by machine
Duration PLP	time of plateau phase
Duration PM	time of stripping phase
Duration TMY	total milking period
i.e.	id est
ISO	International Organization for Standardization
e.g.	exempli gratia
Hz.	Hertz
kg	kilogram
kHz	kilohertz
kPa	kilopascal
lme	linear mixed-effects model
Max	maximum

Min	minimum
Min.	minute
mL	milliliter
MPC	mouthpiece chamber
ms	milliseconds
OP	over-milking phase
PFR	peak flow rate
PLP	plateau phase
PM	subsequent milking yield
p.m.	post meridiem
pmol/L	picomole per liter
PMP	post-milking phase
s	second
SE	Standard Error
TMY	total milk yield
vac.	vacuum
Ø	on average

Summary

During machine milking, farmers often encounter milking problems even though milking machine constructions generally comply with the required international standards. Given that the milking process differs from calf suckling, animal welfare and cow comfort may be compromised during this process.

The first aim of this thesis was to investigate the effect of two different durations of the closing and closed phase (c- and d-phase, respectively) on physical processes in the milking cluster. The second aim was to examine the effect of these c- and d-phases on milk removal and hind-leg activity. It was hypothesized that a prolonged c-phase (i.e. slower liner closing) might be gentler and more comfortable for the dairy cow than a short c-phase (i.e. faster liner closing), mainly as a consequence of reduced air acceleration in the transition period and decreased pressure sums on the teat. Consequently, this would lead to optimized milk removal and calmer dairy cow behavior in the milking parlor. Therefore, dairy cows were confronted randomly with two types of pulsation chamber cycles (Treatments A and B) for 12 milkings. The treatments differed in the durations of c- and d-phases. In Treatment A, the c-phase lasted 70 ms and the d-phase 330 ms, whereas in Treatment B, the c-phase lasted 130 ms and the d-phase 270 ms. Using a vacuum measuring device (MT52, BEPRO AG, Güttingen, Switzerland), measurements were taken during milking proceedings. Milk flow characteristics were recorded using milk flow meters (LactoCorder[®], WMB AG, Balgach, Switzerland). Hind-leg activity was recorded during milking using accelerometers attached on the hind-legs (RumiWatch[®] pedometer, ITIN+HOCH GmbH, Liestal, Switzerland).

Treatment B showed the following effects on parameters measured in the milking cluster compared with Treatment A:

- Durations of open liner were on average 26.2 ms longer.
- Durations of closing liner were on average 23.4 ms longer.

- Pressure sum was on average 1 kPa lower.
- No differences were detected in mouthpiece chamber vacuum.
- Values of maximum vacuum on the teat-end were on average 0.2 kPa higher.
- Maximum vacuum accelerations measured on the teat-end were generally lower (liner open: Ø 94 kPa/s², liner closed: Ø 122 kPa/s², liner closing: Ø 6 kPa/s²).

Treatment B showed the following effects on milk flow and hind-leg activity compared with Treatment A:

- Cows produced 0.21 kg higher total milk yield.
- Peak flow rate was 1.04 kg/min higher.
- Durations of plateau phase were 0.1 min shorter.
- No differences between treatments in hind-leg activity were found.
- Decrease in maximum values of hind-leg activity with milk flow curve progression was detected between the phases.

No guidelines of c-phase durations are given in the international standards. However, this thesis showed that c-phase durations influence physical processes in the milking cluster and milk flow characteristics. From a physical perspective, an elongated c-phase could therefore lead to gentler milking due to decelerated air accelerations on the teat-end and decreased pressure sum. In terms of physiology, experimental dairy cows showed higher milk yields and peak flow rates. Thus, this is not only beneficial for dairy cow's health but also from an economic point of view. Statements regarding welfare improvements are rather difficult to make. Therefore, future research should focus on milking machines with dairy cows showing restlessness during milking.

In addition to the first and second aim of this thesis, the third aim was to examine the correlations of hind-leg activity with accelerometers attached to the hind-leg and to the

milking cluster with direct observations. As restlessness during milking is considered as an important parameter for impairments of dairy cow welfare, a standardized measuring procedure for this behavior may be severe for detection of deficient milking conditions. It was assumed that the milking cluster is set in motion as hind-legs of dairy cows move, because the milking cluster is freely suspended on the cow's udder. Therefore, an accelerometer attached to the milking cluster may measure the hind-leg activity of the dairy cow indirectly. This method could replace laborious attaching of accelerometers on the hind-legs of dairy cows and provide a standardized on-line measuring procedure. Therefore, measurements with accelerometers on hind-legs of dairy cows and on the milking cluster were taken once during morning and evening milkings for every cow. In addition, direct observations of dairy cows' motion behavior during milking were made. The differentiation of hind-leg activity took place between active phases and inactive phases. Data from morning milkings were used to create an algorithm to validate data automatically from evening milkings.

The correlation measurements of hind-leg activity were as follows:

- The „level of agreement“ between the „recording of hind-leg activity by means of accelerometers attached to the milking cluster“ and the “recording of hind-leg activity by means of accelerometers attached directly to the hind-legs” was 0.6 units. This means that the output values of sensors on hind-legs were 0.6 higher than the output values of sensors on milking clusters.
- Algorithm is defined as the number of observations using mean values + standard deviation + 0.2 m/s² (Eq.1):

$$\text{Algorithm} = \frac{1}{n} \sum_{i=1}^n x_i + \frac{1}{n-1} \sum_{i=1}^n (x_i - \bar{x})^2 + 0.2 \quad (\text{Eq.1})$$

- Correlation “visually counted number of active phases from graphs” with “number of observations calculated with the algorithm from the milking cluster” was 97 % (morning milkings).

- Correlation “number of active phases recorded with milking cluster using the algorithm” with “number of active phases using direct observations” was 74 %.
- Correlation “number of active phases recorded with the hind-leg using the algorithm” with “number of active phases using direct observations” was 91 %.

It was possible to develop a hind-leg activity measuring method, without attaching accelerometers on the hind-legs of dairy cows. With this novel method, costs and labor can be minimized and objective examination of animal behavior can be guaranteed. In a next step, it can be implemented in the milking parlor or in the automatic milking device as a diagnostic tool providing valuable information to the farmer and consultant in a management program.

Zusammenfassung

Obwohl die Installationen der marktverfügbaren Melkanlagen generell mit den Normen des internationalen Standards übereinstimmen, bekunden Landwirte oftmals Melkprobleme. Mit dem Wissen, dass das Maschinenmelken sich stark vom Milchentzug des Kalbes unterscheidet, könnte darauf geschlossen werden, dass das Wohlbefinden und der Kuhkomfort während des Melkens beeinträchtigt ist.

Das erste Ziel dieser vorliegenden Arbeit war die Analyse des Einflusses zweier unterschiedlich andauernden Belüftungs- und Druckphasen (C- und D-Phasen) im Pulszyklus, auf die physikalischen Vorgänge im Melkaggregat. Das zweite Ziel war die Untersuchung der Auswirkung dieser C- und D-Phasen auf den Milchentzug und dem Kuhkomfort. Es wurde angenommen, dass eine verlängerte C-Phase sanfter und angenehmer für das Milchvieh ist als ein schnellschliessender Zitzengummi aufgrund einer verlangsamten Luftbeschleunigung in den Übergangsphasen und einer geringeren Drucksumme an der Zitze. Eine verlängerte C-Phase könnte zu einem optimierten Milchentzug und zu ruhigeren Milchkühen während des Melkens führen. Um dies wissenschaftlich zu untersuchen wurden die Milchkühe in zufälliger Reihenfolge mit zwei unterschiedlichen Pulskurven (Behandlung A und B) während 12 Melkungen konfrontiert. In Behandlung A betrug die C-Phase 70 ms und die D-Phase 330 ms, während in Behandlung B die C-Phase 130 ms und die D-Phase 270 ms andauerte. Mit einem Vakuummessgerät (MT52, BEPRO AG, Güttingen, Schweiz) wurden Messungen an einem Melkzeug während des Melkens durchgeführt. Milchflussparameter wurden mit einem Milchflussmessgerätes (LactoCorder[®], WMB AG, Balgach, Schweiz) erfasst, und die Aktivität der Hinterbeine wurde mit Beschleunigungssensoren (RumiWatch[®] Pedometer, ITIN+HOCH GmbH, Liestal, Schweiz) untersucht.

Behandlung B zeigte verglichen mit Behandlung A folgende Auswirkungen auf die physikalischen Vorgänge im Melkaggregat:

- Die Dauer des offenen Zitzengummi war im Durchschnitt 26.2 ms länger.
- Die Dauer des schliessenden Zitzengummi war im Durchschnitt 23.4 ms länger.
- Die Drucksumme war im Durchschnitt 1 kPa niedriger.
- Keine Unterschiede zeigten sich bezüglich des Zitzengummikopfvakuums.
- Die Werte des maximalen Vakuums unter der Zitzenspitze waren durchschnittlich um 0.2 kPa höher.
- Die maximalen Luftbeschleunigungen gemessen unter der Zitzenspitze waren generell niedriger (Zitzengummi offen: $\bar{\Delta} 94 \text{ kPa/s}^2$, Zitzengummi geschlossen: $\bar{\Delta} 122 \text{ kPa/s}^2$, Schliessen des Zitzengummi: $\bar{\Delta} 6 \text{ kPa/s}^2$).

Behandlung B zeigte verglichen mit Behandlung A folgende Auswirkungen auf den Milchfluss und die Aktivität der Hinterbeine:

- Die Kühe erzielten eine durchschnittlich 0.21 kg höhere Gesamtmilchmenge.
- Der „höchste Milchfluss“ war im Durchschnitt 1.04 kg/min höher.
- Die Dauer der Plateauphase war durchschnittlich 0.1 min kürzer.
- Die Hinterbeinaktivität zeigte keine Unterschiede zwischen den Behandlungen.
- Die Maximalwerte der Hinterbeinaktivität nahmen mit dem Verlauf der Milchflusskurve ab.

Die Dauer der C-Phase ist im internationalen Standard nicht definiert. Jedoch wird in dieser Studie deutlich, dass diese einen Einfluss auf die physikalischen Vorgänge im Melkaggregat und den Milchfluss hat. Aus physikalischer Sicht betrachtet, kann eine verlängerte C-Phase aufgrund geringerer Luftbeschleunigungen an der Zitzenspitze und geringerer Drucksumme zu einem schonenderen Melken führen. Physiologisch gesehen zeigten die Versuchstiere höhere Milchmengen und höhere Milchflussraten. Somit ist diese verlängerte C-Phase nicht nur für die Tiergesundheit, sondern auch wirtschaftlich gesehen vorteilhaft. Aussagen über

das Wohlbefinden der Kühe sind jedoch schwer zu formulieren. Hierfür sollten Studien mit verlängerten C-Phasen in Melkanlagen mit vermehrt unruhigen Kühen durchgeführt werden.

Zusätzlich zum ersten und zweiten Ziel dieser Studie, galt es als drittes Ziel, die Korrelationen von der direkt beobachteten Hinterbeinaktivität mit Beschleunigungssensoren an den Hinterbeinen und am Melkaggregat zu überprüfen. Es wurde vermutet, dass das Melkaggregat aufgrund der Bewegung der Hinterbeine in Bewegung gesetzt wird, da es frei am Kuheuter hängt. Um die Hinterbeinaktivität am Melkaggregat indirekt zu erfassen, wurde ein Beschleunigungssensor am Melkaggregat angebracht. Diese neue Methode der Hinterbeinaktivitätserfassung könnte ein mühsames Anbringen von Beschleunigungssensoren an den Hinterbeinen der Milchkühe überflüssig machen. Zur Datenaufnahme wurden Messungen mit Beschleunigungssensoren an den Hinterbeinen der Milchkühe und am Melkaggregat während der Morgen- und Abendmelkungen einmal je Tier durchgeführt. Zusätzlich wurde die Hinterbeinaktivität direkt beobachtet. Die Hinterbeinaktivität wurde in aktive und inaktive Phasen unterteilt. Die Daten der Morgenmelkungen wurden dazu benutzt, einen Algorithmus zu erstellen, welcher auf die Daten der Abendmelkungen angewendet wurden, um eine automatisierte Hinterbeinaktivitätserfassung zu testen.

Die Korrelationsrechnungen der Aktivitätsmessung an den Hinterbeinen waren wie folgt:

- Das Maß der Übereinstimmung zwischen der „Erfassung der Hinterbeinaktivität anhand des Beschleunigungssensors am Melkaggregat“ und der „Erfassung der Hinterbeinaktivität anhand der Beschleunigungssensoren direkt an den Hinterbeinen“ betrug 0.6 Einheiten. Dies bedeutete, dass die Sensoren an den Hinterbeinen um 0.6 höhere Werte ergaben als die Sensoren am Melkaggregat.
- Der Algorithmus definierte sich aus „Anzahl Beobachtungen aus Mittelwert + Standardabweichung + 0.2 m/s²“ (Formel 1):
- $$\text{Algorithmus} = \frac{1}{n} \sum_{i=1}^n x_i + \frac{1}{n-1} \sum_{i=1}^n (x_i - \bar{x})^2 + 0.2 \quad (\text{Formel 1})$$

- Die Korrelation von der „Anzahl gezählter aktiver Phasen aus der grafischen Auswertung“ und der „Anzahl aktive Phasen berechnet aus dem Algorithmus am Melkaggregat“ war 97 % (Morgenmelkung).
- Die Korrelation von der „Anzahl aktiver Phasen des Melkaggregats errechnet aus dem Algorithmus“ und der „Anzahl aktive Phasen Direktbeobachtung“ war 74 %.
- Die Korrelation „Anzahl aktiver Phasen der Hinterbeine errechnet aus dem Algorithmus“ und der „Anzahl aktive Phasen Direktbeobachtung“ war 91 %.

Mit dem Erstellen eines Algorithmus gelang es eine Methode zur Hinterbeinaktivitätsmessung zu entwickeln, ohne Beschleunigungssensoren an den Hinterbeinen anbringen zu müssen. Mit diesem neuartigen Ansatz könnten Kosten und Arbeitseinsatz minimiert werden. Es ist vorstellbar, diese Methode als Diagnosewerkzeug in den Melkstand oder in das automatische Melksystem zu implementieren um wichtige Kuhsignale noch besser zu erkennen und an den Landwirt und Berater durch ein Managementprogramm zu senden.

Chapter 1

General introduction

A calf's perspective

One might think that machine milking is based on the suckling of a calf. Alexander Gillies developed the two-chambered teat cup in 1903, which is still in use as the widespread principle today: by applying vacuum on the teat, milk is extracted from the cisterns. Therefore, vacuum is the 'motive force' in milking machines. Pressure changes in the pulsation chamber ensure the rhythmic opening and closing of the liner (Bramley *et al.*, 1992).

The main difficulty in machine milking had been the technical realization of the animals demand on a physiologically optimal milk removal (Worstorff, 1977). Probably hardly any other sector in agricultural engineering finds technology and biology linked as closely as the milk production field. Due to technical constraints, machine milking functions differently than the suckling of a calf. The calf uses pressure and vacuum for milk extraction. Rasmussen and Mayntz (1998) observed that about one-third of the calf's work to extract the milk derives from positive pressure and about two-thirds are extracted with the vacuum applied. They drew the conclusion that there is no biological advantage of a constant vacuum. The calf appears to be able to apply large physical forces in a way rather opposite to that of milking machines (Smith and Petersen, 1945).

McDonald and Witzel (1966) described the suckling of a calf as a cyclic process, divided into active and resting phases: "During the active phase, the calf produces a vacuum at the teat-end within the oral cavity and creates pressure within the teat cistern when the teat is compressed between the tongue and the hard palate. The teat-end is closed by the tip of the tongue, the dental pad, and the hard palate, and therefore the pressure increases within the teat cisterns. In

the resting phase, the mouth of the calf relaxes, and consequently vacuum at the teat-end is relieved and tissue rebound is ensured. The teat cistern refills with milk, when the pressure on the teat-end is relieved at the end of the resting phase. Further, the cyclic rate of 117 cycles per minute during suckling is almost twice as fast as during mechanical or hand milking". McDonald and Witzel (1966) explained that the reasons for the fast milking rate by calves are high differential pressures of 71 kPa and high cyclic rates.

International standards established by the International Organisation of Standardization were set for machine milking: 'These are minimum requirements for milking machine installations. Basic demands for the construction and performance of milking machines were determined by the physiology of the animal and the need for a standard of high hygiene and milk quality' (ISO 5707, 2007).

Challenges in milking technology

Despite international standards, farmers complain about milking problems such as milk-ejection disorders, poor udder health, cow's unwillingness to enter the milking parlor and restlessness during milking. These physiological and behavioral parameters are indicative of reduced cow welfare during milking. Consequences can be performance losses, insufficient udder health or increased milking times (Bruckmaier *et al.*, 1993). Increased milking speed is linked to decreased milking labor time, and labor is one of the major costs in milk production (Boettcher *et al.*, 1998). Dodenhoff *et al.* (1999) stated that the main objective of the evaluation for milkability is to improve labor efficiency. Thomas *et al.* (1991) found the major aim in milking to be gaining maximum milk yield while spending least labor and keeping cows healthy.

However, milking problems can arise from field husbandry (barn, waiting area, milking parlor), electrical emissions, animal nutrition, animal breeding, human beings (labor, hygiene), or milking technique. A survey showed that 21 % of Swiss dairy farms complain about milking problems (Savary *et al.*, 2010).

Pulsation in the milking machine

Although the principle of milk removal by calf suckling and machine milking differs, it is agreed that machine milking requires a relieve phase from the continuous vacuum phase. Therefore, machine milking includes massage and suction components. Until today, neither all complex actions of single components nor the interactions have entirely been illuminated. However, clarification and comprehension of processes during milking are obligatory to improve milking. Physical processes in the teat cup are of great importance, as milking vacuum and pulsation affect the cow directly (Worstorff and Fischer, 2000). In ISO 3918 (2007) pulsation is defined as the cyclic opening and closing of a liner. The main function of pulsation is to circulate blood through the teat and therefore reduce congestion and edema (Worstorff *et al.*, 1985). Typical values show 60 cycles per minute, with 60 suction phases.

The pulsation chamber cycle comprises one complete liner movement sequence and is divided into four phases (Figure 1): “The increasing vacuum phase (opening phase, a-phase) is the period when the vacuum in the pulsation chamber is increasing from 4 kPa to the maximum pulsation chamber vacuum minus 4 kPa. The maximum vacuum phase (open phase, b-phase) is the period when the vacuum in the pulsation chamber is above the maximum pulsation chamber vacuum minus 4 kPa. The decreasing vacuum phase (closing phase, c-phase) is the period when the vacuum in the pulsation chamber decreases from the maximum pulsation chamber vacuum minus 4 kPa to 4 kPa. Finally, the minimum vacuum phase (closed phase, d-phase) is the period when the vacuum in the pulsation chamber is below 4 kPa” (ISO 3918, 2007).

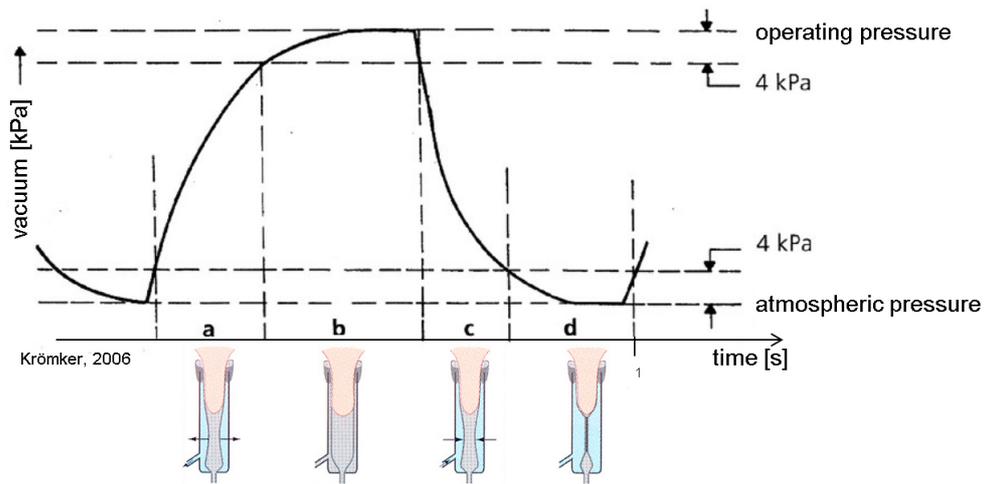


Figure 1: Pulsation chamber cycle (ISO 3918, 2007)

Pulsation chamber cycle consisting of the vacuum phase: opening phase (a-phase) and open phase (b-phase); and the rest phase: closing phase (c-phase) and closed phase (d-phase).

Problem definition

This thesis gives insights in closing and closed phase (c- and d-phase, respectively) durations of pulsation chamber cycles affecting physical processes in the milking cluster and its consequences on milk removal and comfort in dairy cows. Further, a new approach for hind-leg activity measurements was tested on level of agreement and correlations.

In the pulsation chamber cycle, durations of pulsation cycle phases need to comply with the ISO 5707 (2007): open phase (b-phase) ≥ 300 ms, closed phase (d-phase) ≥ 150 ms. Liner movements, which are controlled by pulsators, are important for teat massage and relieve, but can reduce udder health and cause discomfort to the cows' teats. However, between open and closed phases, transition phases are interposed: the opening phase (a-phase), which is the phase when the vacuum builds up, and the closing phase (c-phase), which is the phase when the atmospheric air is let in. Not only the open and the closed phases but also these transition periods appear to be responsible for health and comfort of the dairy cow. Nevertheless, a- and c-phases were neglected and not defined in the ISO 5707 (2007).

As restlessness during milking is considered an important parameter for impairments of dairy cow welfare, a standardized measuring procedure for this behavior may be severe for detection of deficient milking conditions. Common in use for restlessness behavior measurements during milking are hind-leg activity measurements with accelerometers attached on dairy cows' hind-legs. However, attaching accelerometers for measuring hind-leg activity is time consuming (attaching, detaching for reading-out data, checking functionality, recharging battery) and thus measuring hind-leg activity in dairy cows constantly at every milking time for diagnostic reasons is not practicable. Therefore, an innovative method for measuring hind-leg activity attached to the milking cluster was investigated.

Aims of this study

- The aims of this thesis in the experiments presented in the second and third chapters were to investigate the effect of two pulsation chamber cycles differing in the duration of closing and closed phases (c- and d-phases, respectively) on:
 - physical processes in the milking cluster (second chapter)
 - milk flow characteristics and hind-leg activity in dairy cows (third chapter)

It was hypothesized that a slower liner closing, induced by a prolonged c-phase, could be gentler and more comfortable for the dairy cow than a faster liner closing. This improvement would be the result of reduced decelerated air acceleration in the transition period and decreased pressure sums on the teat. This could lead to optimized milk removal and calmer dairy cows in the milking parlor.

- In the experiment presented in the fourth chapter, the aim was to investigate the correlations of hind-leg activity with accelerometers attached to the hind-legs with the direct observations and the correlations of hind-leg activity measured by accelerometers attached to the milking cluster with direct observations of hind-leg activity.

Hind-leg activity is a representative for restlessness behavior during milking and thus, it is an important parameter for impairments of dairy cow welfare, which might be a result of deficient milking conditions. Dairy cows and milking clusters coalesce during milking. The milking cluster is freely suspended on the cows' udder and is set in motion as hind-legs of the dairy cow move. Therefore, attaching an accelerometer to the milking cluster could measure the hind-leg activity of the dairy cow indirectly. Given sufficient accuracy of this indirect measurement, tedious attaching of accelerometers on the hind-legs of dairy cows *would* no longer be necessary, and activity measurement could be implemented in the milking parlor and in the automatic milking device as a diagnostic tool.

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Chapter 2

Effects of an extended c-phase on vacuum conditions in the milking cluster

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Submitted to ELSEVIER BIOSYSTEMS ENGINEERING

Abstract

The objective of this study was to examine how a prolonged closing phase in the pulsation chamber cycle would affect vacuum conditions on the teat and liner movement. Vacuum conditions and liner movements, controlled by pulsators, could cause discomfort on the teats and have negative effects on udder health. Therefore, 18 focal dairy cows were confronted randomly with two types of pulsation chamber cycles (Treatments A and B) for 12 milkings in a low-line 2 × 3 auto tandem milking parlor. The treatments differed in the durations of closing and closed phases (c- and d-phases, respectively). In Treatment A, the c-phase was 70 ms and the d-phase was 330 ms, whereas in Treatment B, the c-phase lasted 130 ms and the d-phase 270 ms. Using a vacuum measuring device, measurement series with 5 s intervals were conducted during three phases of the milking process: the plateau phase, the decreasing phase, and the over-milking phase. Differences between the treatments were detected in the pressure sum and maximum vacuum on the teat-end. Treatment B showed on average a 1 kPa lower pressure sum than Treatment A, whereas values of maximum vacuum at the teat-end were on average 0.2 kPa higher in Treatment B than A. Further, differences could be detected

between the three phases during the milking proceedings, showing that the observed parameters were related to the milk flow rate. Concluding, results indicate that Treatment B might be gentler and more comfortable for dairy cows, which was established in a next step.

Key Words: c-phase duration, pulsation cycle, teat-end vacuum, mouthpiece chamber vacuum, liner movement

Introduction

Teat-end Vacuum Conditions

Milking machine installations need to comply with the International Standards (ISO 5707, 2007) as a minimum requirement to ensure functionality of milking machines. However, variable constructions of single elements are possible and occur on dairy farms. Different adjustments of single components can shift the air and milk streams in the milking machine and lead to changes in vacuum behavior on the teat-end and mouthpiece chamber. This might have a negative influence on the teat conditions. Furthermore, bacteria could be back-sprayed and penetrate the teat canal (O'Shea and Walshe, 1970). Consequently, health and well-being of dairy cows can decline (Stanley *et al.*, 1962; Thiel *et al.*, 1973). When the liner is opening, air can flow reversely. Tiny droplets of milk move within the air and are accelerated according to the rapidly moving air flow. The striking of these milk droplets on the teat-end is called "impact" and is considered a risk for new infections (Schlaiss, 1994). At low milk flow, cyclic vacuum fluctuations are considerably lower than at high milk flow, and the air can move nearly unhindered through the air lines and balance pressure differences. Because of these rapid air movements, there is a high "impact" when cyclic vacuum fluctuations are small and milk flow is low (Nordegren, 1980).

Liner Movement

Worstorff (2000) noticed that neither the pulsation chamber cycle nor the machine's vacuum on the udder allows accurate estimates of the associated liner movement. The differential pressure curve (resulting from the difference between the pulsation chamber vacuum and the teat-end vacuum) and the milk flow control the liner movement. Spencer and Jones (2000) illustrated liner wall movement by data acquisition and laser technology and detected that the

durations of liner movement were not proportional to the pulsation. The pulsation chamber cycle consists of one whole liner movement sequence and is divided into four phases: “the increasing vacuum phase (opening phase, a-phase) is the period when the vacuum in the pulsation chamber is increasing from 4 kPa to the maximum pulsation chamber vacuum minus 4 kPa. The maximum vacuum phase (open phase, b-phase) is the period when the vacuum in the pulsation chamber is above the maximum pulsation chamber vacuum minus 4 kPa. The decreasing vacuum phase (closing phase, c-phase) is the period when the vacuum in the pulsation chamber decreases from the maximum pulsation chamber vacuum minus 4 kPa to 4 kPa. Finally, the minimum vacuum phase (closed phase, d-phase) is the period when the vacuum in the pulsation chamber is below 4 kPa” (ISO 3918, 2007). As durations of pulsation cycle phases (a-phase:b-phase:c-phase:d-phase) were divided in 98:601:42:259 ms, the real liner movement was 87:483:23:407 ms. Thiel *et al.* (1964) found that liner movement was delayed during milking compared with liner movement without milk flow, and Kochman *et al.* (2008) found the duration of the chamber vacuum in the c-phase was longer than the actual movement of the closing liner. As milk flow increased, liner closing decelerated, and therefore duration of the c-phase increased although the overall pulsation cycle progression remained the same (Mayer and Grimm, 2003a). However, Mayer and Grimm (2003b) noticed that the closing of the liner started in the beginning of the c-phase and was completed within the first half of the c-phase. Regarding the well-being of dairy cows during milking, Kochman *et al.* (2008) assumed that a faster liner closing will apply pressure faster to the teat and at higher levels. This will result in an initial spike in pressure causing physical discomfort to the cow and potentially causing milk to be injected back into the udder. Furthermore, Mein *et al.* (2003) explained that the transient peak pressure is at a higher level when the teat-end is compressed faster, and therefore the duration of liner closing is shorter. Consequently, teat health could be impaired by the closing of the liner (Reitsma *et al.*, 1981).

Pulsation and Pressure Sum

Not only vacuum conditions but also liner movements controlled by pulsators can cause discomfort on the teats and have negative effects on udder health, as several studies show (Reitsma *et al.*, 1981; Worstorff *et al.*, 1985; Hamann and Mein, 1996; Billon and Gaudin, 2001; Albers, 2011). The durations of pulsation cycle phases are defined as follows: the b-phase should last at least 300 ms in a pulsation cycle and the d-phase at least 150 ms (ISO 5707, 2007). However, Billon and Gaudin (2001) noted that the a- and c-phases are not mentioned in the International Organization for Standardization due to lack of scientific results on their influence on milking and udder health of dairy cows.

The function of the pulsating liner is to avoid disturbances of blood circulation in the teat (Rasmussen and Madsen, 2000), and therefore the main intention of pulsation is to prevent congestion and edema in the teat tissue during machine milking (Reinemann *et al.*, 1994). When the liner is closed, forces apply to the teat and consequently induce a massage effect. Mein *et al.* (1973) were able to explore over-pressure with radiographic recordings. With the liner closed, the force increased near the end of milking. Consequently, a severe over-pressure can damage the teat or lead to hyperkeratosis (Mein *et al.*, 2003; Zucali *et al.*, 2008; Mein and Reinemann, 2009). As measuring over-pressure is complex and less practicable, further insights into forces acting on the teat-end can be gained by the pressure sum, which is estimated from the area between the differential pressure curve and the compressive force in the massage phase (Spohr, 2012). Spohr (2012) recommends a pressure sum that ranges between 6.75 and 9 kPa*s.

Mouthpiece Chamber Vacuum

Borkhus and Ronningen (2003) observed the different milk flow phases in the milk flow curve progression, focusing on mouthpiece chamber vacuum. They detected a mostly stable mouthpiece chamber vacuum during peak flow rate but an increased mouthpiece chamber vacuum and increased fluctuations due to pulsation during low flow rate.

In this study, we evaluated the effect of vacuum conditions on the teat and in the milking cluster when the closing phase (c-phase) of the pulsation chamber cycle was prolonged.

Materials and Methods

The experiment was performed in July 2013 at the Agroscope Taenikon Research Station in Ettenhausen, Switzerland. Eighteen experimental dairy cows (7 Brown Swiss and 11 Red Holstein x Swiss Fleckvieh), housed in a loose-housing system and milked twice per day at equal intervals in a low-line 2 × 3 auto tandem milking parlor (GEA Farm Technologies GmbH, Bönen, Germany), were confronted randomly with two treatments for 12 milkings. Lactation stages ranged from 3 to 596 days in milk (DIM), and lactation numbers ranged from 1 to 9. Average total milk yield per milking was 16.4 kg and ranged from 6.2 to 25.2 kg.

Treatments A and B were programmed to differ in the durations of closing and closed phases (c- and d-phases, respectively) in the pulsation chamber cycle (Figure 2). The c-phase of the pulsation chamber cycle in Treatment B was prolonged to 130 ms and the d-phase shortened to 270 ms in contrast to the settings in Treatment A (reference, c-phase: 70 ms, d-phase: 330 ms). In both treatments, durations of opening and open phases (a- and b-phases, respectively) were set to 140 and 460 ms, respectively. Pulsation chamber cycle phase durations were verified with a dry test of pulsation before each treatment to ensure proper duration settings (ISO 6690, 2007).

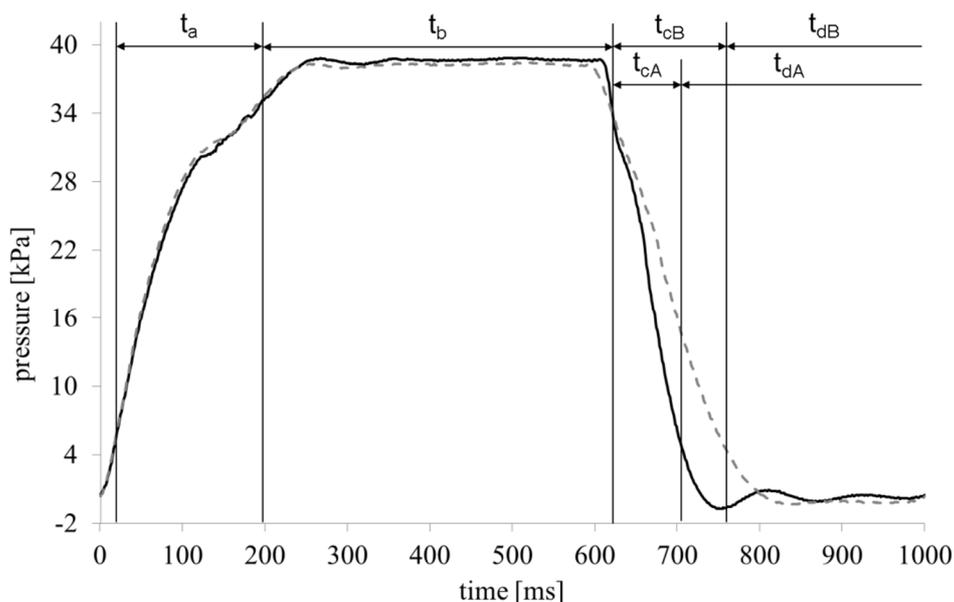


Figure 2: Pulsation chamber vacuum during one pulsation chamber cycle with the different pulsator settings.

Treatment A: reference ($t_{cA} = 70$ ms; $t_{dA} = 330$ ms), Treatment B: prolonged c-phase ($t_{cB} = 130$ ms) and shortened d-phase ($t_{dB} = 270$ ms). A- and b- phases, similar for both treatments: $t_a = 140$ ms; $t_b = 460$ ms.

Milk flow meters (LactoCorder[®], WMB AG, Balgach, Switzerland) and pulsator devices (RotoPuls[®] integral, BITEC Engineering, Romanshorn, Switzerland) were installed at each milking stall during test-period milkings. Treatments were preceded by a 2 week visual habituation period for dairy cows to measuring devices, routine, and observer. Due to warm weather conditions (average ambient temperature 22.5 °C) and high appearances of flies at 4 p.m. during the evening milkings, measurements took place only during the morning milkings at 4 a.m. to reduce those disturbing factors.

In the milking system, machine vacuum was set to 38 kPa. Milking cluster types used were the GEA “Classic 300” (GEA Farm Technologies GmbH, Bönen, Germany). Nitrile rubber teat cup liners (GEA Farm Technologies GmbH, Bönen, Germany) were renewed 6 weeks before experiment start. The pulsation ratio was set to 60:40 with a pulsation rate of 60 cycles/min. Automatic stripping was set to a flow threshold of 0.8 kg/min and automatic

cluster removers were set to a flow threshold of 0.3 kg/min. The touch point pressure was on average 8.5 kPa (± 0.3).

The two milkers used consistent preparation procedures regarding pre-milking, cleaning, and unit attachment. In total, milkers needed 45 s for the preparation procedures. The automatic stimulation lasted approximately 30 s for dairy cows in the first lactation stage (1–100 DIM), approximately 40 s for dairy cows in the second lactation stage (101–200 DIM), and approximately 50 s for dairy cows in the third lactation stage (201–300 DIM).

The six novel and patented pulsator devices (RotoPuls[®] integral, BITEC Engineering, Güttingen, Switzerland: patent number: CH 701646B1) consisted of three main components: two servo motors, a valve block, and a buffering tank. Each servo motor controlled one rotary disc in the valve block and worked separately from the other. The circumference of the rotary disc allowed the cyclic switch between vacuum and atmospheric pressure in the pulsation chamber. Therefore, the length of a cycle phase was defined by the rotation speed of the disc, which could be programmed accurately. The buffering tank was embedded between the valve block and pulsator airline to reduce the vacuum fluctuations in the air lines.

Three 5 s measurement series were performed during milking in three different phases of the milk flow curve: plateau phase (PLP), decreasing phase (DP), and over-milking phase (OP). Therefore, five repetitions per measurement series were carried out, as one second represented one repetition. The three different phases were chosen according to different milk flow rates. PLP and DP were read by LactoCorder[®] displays, and the OP was recognized by a sight glass integrated in the teat cup (Table 1).

Table 1: The three vacuum measurement phases plateau phase (PLP), decreasing phase (DP), and over-milking phase (OP) were chosen according to different milk flow rates

Measurement series	Milk flow rate
Plateau phase (PLP)	Highest milk flow rate
Decreasing phase (DP)	Milk flow rate lower than in PLP
Over-milking phase (OP)	Milk flow rate < 0.8 kg/min

Pressure conditions at the teat-end and in the mouthpiece chamber (MPC) were measured in one teat cup of a milking cluster at a front quarter of the cow's udder. Furthermore, the pressure in the short milk tubes of the cluster was recorded. Pressure was measured by pressure sensors (frequency band: 5 kHz) and recorded by a MT52 measuring device (BEPRO AG, Göttingen, Switzerland) with a sampling rate of 1 kHz.

The pressure sensor measuring the teat-end vacuum was connected to a tube, which was installed instead of the sight glass. The tube had the same dimension as the sight glass. Five holes distributed equally in the inner area of the tube assured precise vacuum measurements. The MPC vacuum was measured by connecting a pressure sensor to a tube that was attached to the inner side of the liner in the MPC to prevent interferences. For measuring the pressure in the short pulse tubes, pressure sensors were interposed between the short pulse tubes. Vacuum conditions on the teat-end, in the MPC and in the short pulse tubes and liner movement were analyzed by means of parameters, shown in Table 2. Figure 3 demonstrates the calculation models for the parameters pressure sum and liner movement.

Table 2: Parameters for analyzing the vacuum conditions on the teat-end, in the mouthpiece chamber, and in the short pulse tubes and the liner movement, calculated for the three phases in Treatments A and B.

Parameter	Abbreviation
Opening duration of the liner	duration liner opening
Open duration of the liner	duration liner open
Closing duration of the liner	duration liner closing
Sum of pressure per cycle in the liner	pressure sum
Minimum/least vacuum in the mouthpiece chamber	min vacuum MPC
Maximum/highest vacuum in the mouthpiece chamber	max vacuum MPC
Mean vacuum in the mouthpiece chamber	mean vacuum MPC
Bandwidth in the mouthpiece chamber vacuum	bandwidth MPC
Mean vacuum on the teat-end during open phase of the liner	mean vacuum liner open teat-end
Mean vacuum on the teat-end during closed liner	mean vacuum liner closed teat-end
Maximum vacuum on the teat-end during closing liner	max vacuum liner closing teat-end
Minimum vacuum on the teat-end	min vacuum teat-end
Maximum vacuum on the teat-end	max vacuum teat-end
Bandwidth of the teat-end vacuum	bandwidth vacuum teat-end
Maximum acceleration of the vacuum in the open liner on the teat-end	max acc. vac. liner open teat-end
Maximum acceleration of the vacuum in the closed liner on the teat-end	max acc. vac. liner closed teat-end
Maximum acceleration of the vacuum in the beginning of the closing liner on the teat-end	max acc. vac. liner closing teat-end

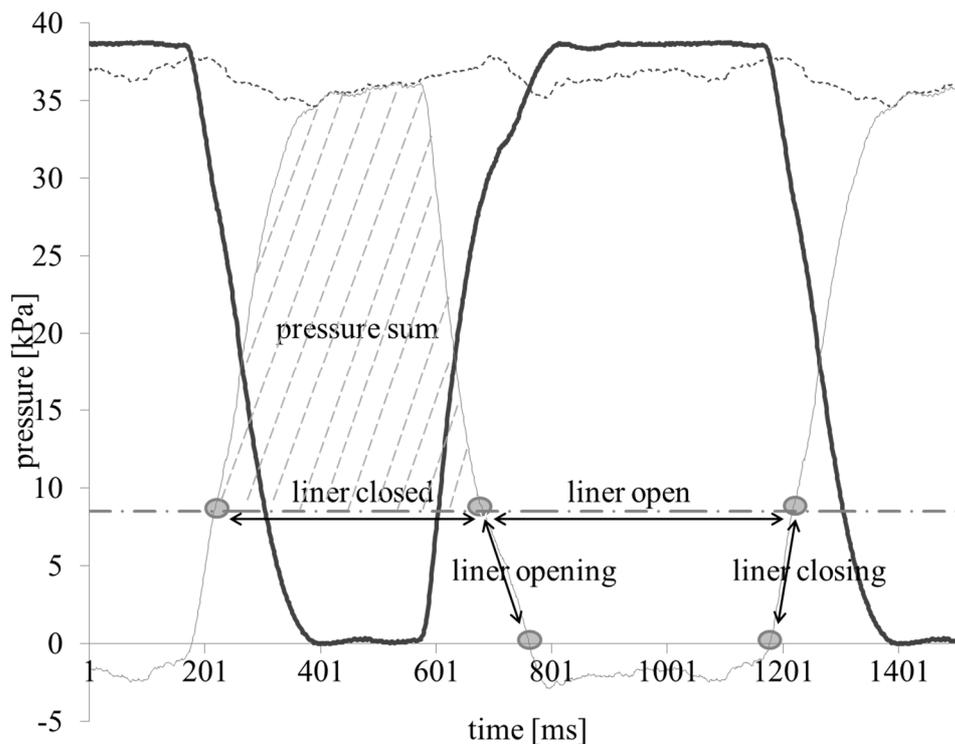


Figure 3: Calculation model for the parameters: pressure sum [kPa*s], duration liner opening [ms], duration liner open [ms], and duration liner closing [ms] expressed for the pulsation chamber (black line), the differential pressure (thin grey line), and the touch point pressure (grey patterned line, 8.5 kPa). Teat-end vacuum (thin dotted black line) subtracted from pulsation chamber equals the differential pressure.

The statistical analysis was performed in the statistic program system R 1.9.1 (R Development Core Team, 2011) with the package lme4. The linear mixed-effects model (Bates *et al.*, 2012) was used to evaluate differences between the Treatments A and B in the different phases PLP, DP, and OP. Target variables were pressure sum, duration liner opening, duration liner open, duration liner closing, mean vacuum liner open teat-end, mean vacuum liner closed teat-end, max vacuum liner closed teat-end, min vacuum teat-end, max vacuum teat-end, bandwidth vacuum teat-end, max acceleration vacuum liner open teat-end, max acceleration vacuum closed teat-end, max acceleration vacuum liner closing teat-end, min vacuum MPC, max vacuum MPC, mean vacuum MPC, and bandwidth vacuum MPC (Table 2). Following responses were log transformed: duration liner opening, bandwidth vacuum teat-end, max acceleration vacuum liner open teat-end, max acceleration vacuum liner closed teat-end, max acceleration vacuum liner closing teat-end, and bandwidth MPC. The model contained the Treatments A and B as explanatory variables. Animals and measuring days were regarded as random effects (nested).

Results

Duration Liner Opening, Duration Liner Open, and Duration Liner Closing

Values for duration liner opening were between 96.4 and 117.0 ms. Significant differences could be detected in the interaction of treatments and phases ($F_{2,543} = 8.77, P = 0.0002$). Duration increased continuously as phases progressed in Treatment A, whereas in Treatment B, duration was similar in PLP and DP but longer in OP than in PLP and DP (Table 3). For duration liner open, values ranged from 551.7 to 603.0 ms. There was a significant difference in the interaction of treatments and phases ($F_{2,543} = 3.12, P < 0.05$). With phase progression, duration decreased in both treatments. However, the durations in Treatment B were longer than in Treatment A (Table 3). Duration liner closing ranged between 37.4 and 66.3 ms and was significantly longer (by 23.5 ms) in Treatment B than A ($F_{1,545} = 1.010, P < 0.001$). In addition, phases showed significant differences ($F_{2,545} = 19.4, P < 0.001$). The duration of liner closing in DP was slightly shorter than in PLP and OP in both treatments (Table 3).

Table 3: Mean values \pm SE of vacuum parameters in the phases plateau (PLP), decreasing (DP), and over-milking (OP) for the Treatments A and B

Phase	Treatment A ¹			Treatment B ²		
	PLP	DP	OP	PLP	DP	OP
Duration liner opening [ms]	96.4 \pm 11.5	102.5 \pm 16.6	117.0 \pm 17.0	98.7 \pm 9.2	98.5 \pm 13.1	109.5 \pm 18.3
Duration liner open [ms]	576.7 \pm 18.0	557.0 \pm 16	551.7 \pm 5.57	603.0 \pm 18.5	586.0 \pm 17.0	575.0 \pm 6.9
Duration liner closing [ms]	42.7 \pm 10.1	37.4 \pm 5.9	44.9 \pm 1.7	66.2 \pm 10.5	62.9 \pm 10.5	66.3 \pm 11.2
Pressure sum [kPa*s]	7.1 \pm 0.7	8.5 \pm 0.7	9.3 \pm 0.3	6.1 \pm 0.8	7.3 \pm 0.9	8.1 \pm 0.3
Min vacuum MPC [kPa]	10.3 \pm 5.0	12.8 \pm 4.6	13.9 \pm 5.4	9.5 \pm 4.8	11.8 \pm 5.4	14.8 \pm 5.6
Max vacuum MPC [kPa]	11.9 \pm 5.5	24.3 \pm 6.7	27.8 \pm 3.6	12.0 \pm 6.7	24.6 \pm 6.1	26.9 \pm 4.8
Mean vacuum MPC [kPa]	11.2 \pm 5.2	19.5 \pm 4.5	22.2 \pm 2.5	10.9 \pm 5.7	19.7 \pm 4.3	22.1 \pm 3.7
Bandwidth vacuum MPC [kPa]	1.6 \pm 0.9	11.5 \pm 8.1	13.9 \pm 7.6	2.5 \pm 3.3	12.8 \pm 8.1	12.1 \pm 7.3
Mean vacuum liner open teat-end [kPa]	32.7 \pm 1.6	35.0 \pm 1.5	36.7 \pm 0.4	32.8 \pm 1.6	34.9 \pm 2.0	36.8 \pm 0.5
Mean vacuum liner closed teat-end [kPa]	31.7 \pm 1.7	34.7 \pm 1.6	36.6 \pm 0.6	31.8 \pm 1.9	34.7 \pm 2.0	36.6 \pm 0.7
Max vacuum liner closing teat-end [kPa]	33.8 \pm 1.7	36.5 \pm 1.4	37.9 \pm 0.5	33.9 \pm 1.7	36.5 \pm 1.8	37.9 \pm 0.5
Min vacuum teat-end [kPa]	28.8 \pm 3.5	32.4 \pm 1.8	34.4 \pm 0.8	29.8 \pm 2.2	32.2 \pm 2.5	34.7 \pm 0.9
Max vacuum teat-end [kPa]	35.5 \pm 1.2	37.1 \pm 1.1	38.2 \pm 0.4	35.9 \pm 1.1	37.2 \pm 1.2	38.3 \pm 0.4
Bandwidth vacuum teat-end [kPa]	6.7 \pm 2.7	4.7 \pm 1.1	3.8 \pm 0.8	6.1 \pm 1.6	5.0 \pm 1.7	3.6 \pm 0.8
Max acc. vac. liner open teat-end [kPa/s ²]	343 \pm 163	464 \pm 516	431 \pm 345	221 \pm 92	436 \pm 455	300 \pm 309
Max acc. vac. liner closed teat-end [kPa/s ²]	352 \pm 317	364 \pm 489	416 \pm 365	144 \pm 62	365 \pm 473	256 \pm 296
Max acc. vac. liner closing teat-end [kPa/s ²]	131 \pm 46	134 \pm 76	130 \pm 60	121 \pm 36	133 \pm 173	125 \pm 140

¹Treatment A: a-phase duration = 140 ms; b-phase duration = 460 ms; c-phase duration = 70 ms; d-phase duration = 330 ms

²Treatment B: a-phase duration = 140 ms; b-phase duration = 460 ms; c-phase duration = 130 ms; d-phase duration = 270 ms

Pressure Sum

The pressure sum ranged between 6.1 and 9.3 kPa. A significant difference existed between the Treatments A and B, as pressure sum was on average 1 kPa lower in Treatment B than A ($F_{1,545} = 393$, $P < 0.001$). Furthermore, a significant difference existed between the phases ($F_{2,545} = 468$, $P < 0.001$), as pressure sum increased with phase progression (Table 3).

Min Vacuum MPC, Max Vacuum MPC, Mean Vacuum MPC, and Bandwidth Vacuum MPC

Concerning min vacuum MPC, we found significant differences in the interaction of treatments and phases ($F_{2,543} = 5.8, P = 0.003$). Values ranged from 9.5 to 14.8 kPa. Values increased with progression of phases and were slightly lower in Treatment B in PLP and DP, but highest in Treatment B in OP (Table 3). For max vacuum MPC and mean vacuum MPC, were detected no significant differences between the treatments but significant differences between the phases in max vacuum MPC ($F_{2,547} = 530, P < 0.001$) and mean vacuum MPC ($F_{2,547} = 552, P < 0.001$), as values increased with phase progression. For max vacuum MPC, values reached from 11.9 to 26.9 kPa, and for mean vacuum MPC, values ranged from 11.2 to 22.2 kPa (Table 3). A significant difference existed in the interaction of treatments and phases for bandwidth vacuum MPC ($F_{2,543} = 3.8, P = 0.02$). Values ranged between 1.6 and 13.9 kPa. Values were slightly higher in Treatment B in PLP and DP than in Treatment A and increased with phase progression (except OP, Treatment B) (Table 3).

Mean Vacuum Liner Open Teat-end, Mean Vacuum Liner Closed Teat-end, Max Vacuum Liner Closed Teat-end, Min Vacuum Teat-end, Max Vacuum Teat-end

Concerning mean vacuum liner open teat-end and mean vacuum liner closed teat-end, we found no significant differences between the treatments but significant differences between the phases for mean vacuum liner open teat-end ($F_{2,545} = 526, P < 0.001$) and for mean vacuum liner closed teat-end ($F_{2,545} = 583, P < 0.001$). With phase progression, both parameters increased in both treatments (Table 3). The values for max vacuum liner closed teat-end reached from 33.8 to 37.9 kPa. Significant differences did not exist between the treatments but between the phases ($F_{2,545} = 480, P < 0.001$). Higher values of max vacuum liner closed occurred with phase progression.

Regarding min vacuum teat-end, we detected significant differences in the interaction of treatments and phases ($F_{2,543} = 4.3$, $P = 0.014$). Values ranged from 28.8 to 34.7 kPa. As phases progressed, min vacuum teat-end increased, and the increase from PLP to DP was greater in Treatment A than B (Table 3). For max vacuum teat-end, we detected significant differences between the treatments ($F_{1,545} = 8.3$, $P = 0.004$) and the phases ($F_{2,545} = 377$, $P < 0.001$). Max vacuum teat-end values reached from 35.5 to 38.3 kPa. Values were on average 0.2 kPa higher in Treatment B than A. With progression of the phases, max vacuum teat-end increased (Table 3). Regarding bandwidth vacuum teat-end, the interaction of treatments and phases was significant ($F_{2,543} = 4.1$, $P = 0.02$). Bandwidth vacuum teat-end values ranged from 3.8 to 6.7 kPa. With progression of phases, bandwidth vacuum teat-end decreased in both treatments. In Treatment A, the differences between PLP and DP were more pronounced than in Treatment B, whereas differences between DP and OP were less distinctive (Table 3).

Max Acceleration Vacuum Liner Open Teat-end, Max Acceleration Vacuum Liner Closed Teat-end, Max Acceleration Vacuum Liner Closing Teat-end

Significant differences existed in the interaction of treatments and phases ($F_{2,543} = 4.6$, $P = 0.01$) for the parameters max acceleration vacuum liner open teat-end (max acc. vac. liner open teat-end), max acceleration vacuum liner closed teat-end (max acc.vac. liner closed teat-end), and max acceleration vacuum liner closing teat-end (max acc. vac. liner closing teat-end). In both treatments, max accelerations were highest in DP, except for max acceleration vacuum liner closed teat-end in Treatment A. Values for max acceleration vacuum liner open teat-end ranged from 221.3 to 463.5 kPa/s², values for max acceleration vacuum liner closed teat-end from 143.7 to 416.0 kPa/s², and values for max acceleration vacuum liner closing

teat-end from 121.4 to 134.0 kPa/s² (Table 3). Generally, accelerations were lower in Treatment B than A. Great differences occurred between the treatments in PLP and OP.

Discussion

Liner Movement

Our results demonstrated that the actual durations of liner movement varied during phase progression for each treatment and were in line with results of Spencer and Jones (2000), who found liner wall movements to differ from the pulsation chamber cycle. In their study, opening and closing of the liner was much faster than the opening phase and closing phase in the pulsation chamber cycle. In our study, duration of liner closing took half as long as the programmed closing phase durations in the pulsation chamber cycle. Duration of liner opening slightly increased during phase progression in Treatment A, but increased only in OP in Treatment B. Liner opening durations are correlated to milk flow rate. One could assume that the milk flow supports the liner opening. This influence was less obvious in Treatment B. Therefore, Treatment B could be a solution for more constant liner opening movement durations during the milking process. Having more constant liner movement opening durations, less unpredictable changes take place, which might be less stressful for the cow. The duration of the liner being open decreased with phase progression. This could be the consequence of the decreasing milk flow rate. The milk flow rate seems to support the liner to open faster, and therefore, open liner durations are highest when milk flow rate is strongest. Furthermore, durations of the liner being open were longer and thus the vacuum penetrated the teat for a longer time in Treatment B than A. This could explain the higher milk yield and the increased highest milk flow rate detected by Bluemel *et al.* (unpublished data). Contrary to the liner opening and liner open durations, the milk flow rate did not influence the liner closing duration in this study. In both treatments, liner closing duration was marginally shorter in DP than in PLP and OP. These results agree with those of Mayer and Grimm (2003b), who found that liner closing decelerated with milk flow decrease. However, liner

closing durations were similar at full milk flow and no milk flow. Furthermore, liner closing durations were longer in Treatment B than A.

Pressure Sum

The pressure sum increased with phase progression and was at a lower level when the c-phase was prolonged compared with the reference duration. The pressure sum can be determined more easily than the compressive load, or over-pressure, by means of touch point pressure, pulsation chamber vacuum, and teat-end vacuum. The massage of the teat, in the literature often expressed as compressive load or over-pressure, is of great importance for the recovery of the teat from the b-phase. A too strong over-pressure can lead to teat damage and can therefore risk the health of the cow (Zucali *et al.*, 2008; Mein and Reinemann, 2009). In this study, the pressure sum was found to be in the recommended range given by Spohr (2012). The increase with phase progression seemed to be clearly related to the milk flow rate. In PLP, milk flow peaked and the pressure sum was lowest, as the liner worked against the milk flow.

Mouthpiece Chamber Vacuum

Results on vacuum progressions of MPC vacuum were similar to observations by Newman *et al.* (1990) and Borkhus and Ronningen (2003). Mean, max, and bandwidth MPC vacuum were greatly increased in the phases DP and OP. The level and pattern of MPC vacuum appeared to be correlated to the milk flow rate, a result also found by Newman *et al.* (1990), and likely influenced teat conditions during milking.

Teat-end Vacuum Conditions

The parameters mean vacuum liner open teat-end, mean vacuum liner closed teat-end, max vacuum liner closed teat-end, and max vacuum teat-end did all increase with phase progression. Similarly, other studies have shown that vacuum on the teat-end increased when milk flow decreased (Schmidt *et al.*, 1964, Mayer and Grimm, 2003a). The parameter bandwidth vacuum teat-end expresses the fluctuating vacuum and decreased with decreasing milk flow rate, which is consistent with Haeussermann and Hartung (2010).

Accelerations on the Teat-end

Accelerations were lower in PLP and OP in Treatment B compared with Treatment A. Furthermore, standard errors in PLP in Treatment B were decreased compared with Treatment A. In addition, accelerations were lower when the liner was closing than when the liner was open or closed. For both treatments, the RotoPuls[®] pulsators were configured for air streams not reaching the critical pressure. Therefore, in contrast to conventional electric pulsators, high accelerations in the beginning of the a- and c-phases and during the transition from a- to b-phase could not occur. However, the data clearly showed that accelerations were less pronounced in PLP and OP in Treatment B than in Treatment A. This could prevent the occurrence of “impact” (Schlaiss, 1994) and keep dairy cows healthier (Stanley *et al.*, 1962; Thiel *et al.*, 1973). Whereas the focus of our study was on the effects of a prolonged c-phase with all other conditions being identical, further research is needed to elucidate the potential advantages of using RotoPuls[®] pulsators compared with conventional pulsators.

Conclusions

We programmed two pulsation chamber cycles on a novel pulsator differing in the closing and closed phase durations (c- and d-phases, respectively). A prolonged c-phase decreased the pressure sum on average 1 kPa*s and increased the duration of the open liner and the closing liner. Furthermore, vacuum accelerations decreased when the liner was open, closed, and closing. From a physical point of view, these results of a prolonged c-phase indicate a gentler and a more comfortable milking. Teat health could be improved due to a decreased pressure sum and udder infections could be minimized due to lower air accelerations on the teat-end. In a follow-up study, investigations were made concerning the physiological and ethological effects (milk flow characteristics and hind-leg activity) of an elongated c-phase.

Acknowledgements

The authors gratefully acknowledge the Swiss machinery association (SLV) for financial support, and Erwin Bilgery, Alfred Kummer, Ralf Fischli, Stefan Mathis, and Christoph Bühler for assistance and advice.

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Chapter 3

Improving dairy cow welfare by extending the c-phase during milking

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Submitted to CAMBRIDGE JOURNAL OF DAIRY RESEARCH

Abstract

We evaluated the effect of two types of pulsation cycles differing in duration of their closing phase (c-phase) and closed phase (d-phase) on the hind-leg activity and milk removal in dairy cows. Despite standardized milking machines, farmers continue to experience milking problems with cows, such as restlessness during milking, a tendency to knock off the milking units, milk ejection disorders, and low milk yields. These behavioral and physiological parameters are indicative of decreased cow welfare during milking. For dairy cows, the movement of the closing liner controlled by the pulsators, and hence the pressure on the teat, can be uncomfortable. Treatments A and B were programmed to differ in pulsation chamber cycle phases c and d during dry tests (Treatment A: c-phase = 70 ms, d-phase = 330 ms; Treatment B: c-phase = 130 ms, d-phase = 270 ms). Forty-two dairy cows were randomly subjected to Treatment A or B during 12 milking sessions. Each cow's lactation stage was assigned to one of three categories (early, mid, late). Lactation number was classified as first lactation and further lactations. Hind-leg activity was observed during milking and evaluated for four milk flow stages. For statistical evaluation, the linear mixed-effects model was used.

Cows milked in Treatment B tended to produce 0.21 kg more total milk yield (TMY) than cows milked in Treatment A. Peak flow rate (PFR) was 1.04 kg/min higher in Treatment B than A. Treatment B led to a slightly higher PFR and appeared to marginally increase TMY. The Treatments did not affect further milk flow characteristics. No differences in hind-leg activity were found between the two treatments. The maximum hind-leg activity decreased with phase progression. Because no significant differences in hind-leg activity were observed between the treatments, no negative influences concerning the gentleness of liner movement and comfort of the dairy cow were detected by measuring hind-leg activity. However, an increased TMY and PFR in treatment B could indicate an improved milk removal. Therefore, Treatment B seems to have a positive effect on milk removal, but no relevant effect in cow comfort.

Key Words: c-phase duration, pulsation cycle, milk flow curves, hind-leg activity

Introduction

The International Organization for Standardization (ISO) gives minimum requirements for milking machine installations. “The basic requirements for the construction and performance of milking machines for animals are determined by the physiology of the animal and the need for a standard of high hygiene and milk quality” (ISO 5707, 2007). Whereas the physiological aspect is mentioned in the ISO, the aspect well-being or welfare is not considered.

The pulsation chamber cycle is responsible for liner movement and consists of four phases: “the increasing vacuum phase (opening phase, a-phase) is the period when the vacuum in the pulsation chamber is increasing from 4 kPa to the maximum pulsation chamber vacuum minus 4 kPa. The maximum vacuum phase (open phase, b-phase) is the period when the vacuum in the pulsation chamber is above the maximum pulsation chamber vacuum minus 4 kPa. The decreasing vacuum phase (closing phase, c-phase) is the period when the vacuum in the pulsation chamber decreases from the maximum pulsation chamber vacuum minus 4 kPa to 4 kPa. The minimum vacuum phase (closed phase, d-phase) is the period when the vacuum in the pulsation chamber is below 4 kPa” (ISO 3918, 2007).

Pulsation Cycle Phase Durations

Configurations of the pulsators in the milking machine could represent possible contributing causes for milking problems and welfare aspects. For dairy cows, the movement of the liner controlled by the pulsators, and hence the pressure on the teat, can be uncomfortable and cause insufficient udder health (Østerås *et al.*, 1995; Billon and Gaudin, 2001; Albers, 2011). Especially when the liner is closing, high air accelerations appear and cause punches on the teat (Kochman *et al.*, 2008; Albers, 2011). However, Billon and Gaudin (2001) noticed that the opening (a-phase) and closing (c-phase) phases are not mentioned in the ISO due to lack

of scientific results on their influence on the milking and udder health of dairy cows. Finding the optimal c-phase duration is a compromise between the c-phase and the closed phase (d-phase), as a short c-phase causes discomfort during liner closing, whereas a long c-phase reduces the d-phase of the liner (Albers, 2011). It is important to maintain blood circulation through the teat to ensure relief for the teat, and therefore liner closure is compulsory (Reitsma *et al.*, 1981; Worstorff *et al.*, 1985). Reitsma *et al.* (1981) found that 90 % of cows had an infection in the udder when milked without pulsation. With an increased duration of the d-phase, the risk of infection decreased. A short d-phase negatively affected the teat condition, reduced the milk flow, and increased the risk for mastitis due to an insufficient relief (Albers, 2011).

Durations of pulsation phases are primarily defined by the International Organization for Standardization (ISO 5707, 2007). The open phase (b-phase) should not be less than 300 ms of a pulsation cycle and the closed phase (d-phase) not to be less than 150 ms. These regulations find support by the studies of Reitsma *et al.* (1981), Østerås *et al.* (1995), and Hamann and Mein (1996). Hamann and Mein (1996) observed that a d-phase duration of at least 150 ms was sufficient to relieve blood congestion, which did not increase with longer d-phase durations. They further found changes in teat thickness as the duration of the b-phase was extended but also as the duration of the d-phase was reduced. Reitsma *et al.* (1981) and Østerås *et al.* (1995) found increased infection rates when the duration of the liner closure was less than 300 ms. Billon and Gaudin (2001) recommended c-phase durations of 120 ms and a-phase durations of 140 to 160 ms of the pulsation cycle in order to avoid increasing milking times and to prevent udder diseases.

Milk Flow Characteristics

Other studies observed the influence of pulsation cycle phase durations on milk flow characteristics, e.g.: Ambord and Bruckmaier (2009) elongated the b-phase of the pulsation cycle with increasing milk flow and found the peak flow rate (PFR) elevated but mean milk flow and total milk yield (TMY) unchanged. However, when Hamann and Mein (1996) and Bade *et al.* (2009) extended the b-phase, TMY increased moderately and PFR increased greatly. Billon and Gaudin (2001) observed longest milking times and lowest flow rates when the a- and c-phases were short.

Not only pulsation cycle phases but also lactation stage and lactation number have shown an influence on milk removal and milk flow characteristics. In the work by Dodenhoff *et al.* (1999) and Antalík and Strapák (2010), milk flow increased as lactation number was increased and decreased as lactation stage was increased. Regarding the milk yield, lowest amounts of milk were produced in late lactation stages. With increasing lactation stage, pre-stimulation should be extended, because the lower the udder filling, the longer it takes to initiate milk ejection due to a delayed response to the oxytocin by the mammary gland (Worstorff *et al.*, 1980; Mayer and Bruckmaier, 1987; Bruckmaier, 2005). Schams *et al.* (1984) found that milk ejection seemed to follow the threshold principle in that small releases of oxytocin. Up to a range of 3 to 5 pmol/L plasma were necessary to induce maximum milk ejection.

Hind-Leg Activity

Stress might cause milk ejection disturbances, which can lead to lower performances and health impairments (Rushen *et al.*, 2001; Rousing *et al.*, 2004). This is explained by Bruckmaier *et al.* (1993) through central inhibition of oxytocin secretion. Thus, it is important

to identify and reduce the acute stress of cows during the milking process (Rushen *et al.*, 2001). Rushen *et al.* (2001) found that cows milked in stressful situations, such as unfamiliar surroundings, have a higher hind-leg activity during milking. Wenzel *et al.* (2003) and Gygax *et al.* (2008) assumed that cows with a higher step activity, more foot-lifting, more kicking, and a higher heart rate suffered an increased level of stress or nervousness. Willis (1983) found a correlation between behavior and milk yield during milking. He assumed that stepping, kicking, and constant movement during milking can be considered indicative of stressful situations and would consequently lead to inhibition of milk ejection and decreased milk yield.

We hypothesized that dairy cows experience a short duration of c-phase, which affects c- and d-phase, to be uncomfortable and therefore show more hind-leg activity during milking—especially with milking phase progression, as milk flow ceases, vacuum on the teat-end increases, and pressure sum is highest.

Therefore, the aim of this study was to evaluate the effect of two pulsation cycles differing in durations of their closing phase (c-phase) and closed phase (d-phase) on milk removal and hind-leg activity in dairy cows.

Materials and Methods

Animals and Housing

This study was carried out at the Agroscope Taenikon Research Station in Ettenhausen, Switzerland. The 42 experimental dairy cows (16 Red Holstein x Swiss Fleckvieh and 26 Brown Swiss) were kept in a loose-housing system, and milking took place twice per day at 4 a.m. and 4 p.m. in a low-line 2 × 3 auto tandem milking parlor (GEA Farm Technologies GmbH, Bönen, Germany). Experiments were restricted to morning milkings to eliminate disruptive influences such as flies and high temperatures since data collection was during the summer period. Average air temperature was 20.0 °C outside and 22.5 °C in the milking parlor, measured at 4 a.m. with an average atmospheric humidity of 63 %. Average noise measured in the milking pit was 59 dB. Lactation stages ranged from 3 to 596 days in milk (DIM) and were classified in three categories: early = ≤ 100 DIM (15 cows); mid = 101–200 DIM (9 cows), late = > 201 DIM (18 cows). Lactation numbers ranged from 1 to 9 and were split into two categories, first lactation (11 cows) and second and further lactations (31 cows). Average total milk yield (TMY) per milking was 16.4 kg and ranged from 6.23 to 25.15 kg.

Machine Settings and Milking

Milking machine vacuum was preset to 38 kPa, and pulsators to a pulsation ratio of 60:40 and a pulsation rate of 60 cycles/min. Further, automatic stripping was set at a flow threshold of 0.8 kg/min, and automatic cluster removers were set at a flow threshold of 0.3 kg/min. Milking cluster types used were GEA “Classic 300” (GEA Farm Technologies GmbH, Bönen, Germany). Installed teat cup liners with commercial nitrile rubber liners from GEA

(GEA Farm Technologies GmbH, Bönen, Germany) were renewed 6 weeks before the experiment commenced.

Milking was performed by two milking parlor operators, using the same agreed preparation procedures including pre-dip application, manual fore-stripping, cleaning the udder with cellulose paper, and unit attachment. Preparation procedures were required to last 45 s. After unit attachment, automatic stimulation lasted approximately 30 s, 40 s, and 50 s for dairy cows in the first, second, and third lactation stages, respectively.

Treatments

The pulsators (RotoPuls[®] Integral, Bitec Engineering, Romanshorn, Switzerland) were programmed with two pulsation cycles (Treatments A and B) differing in the massage phase (c- and d-phase) durations, which were individually adjusted (Table 4). After a 2 week visual habituation period for the cows to pulsators, measuring devices, and observer, cows were randomly allocated to Treatments A and B for 12 morning milkings. Measurements took place during morning milkings. Durations were verified with a dry test of pulsation before each Treatment.

Table 4: Programmed pulsation cycle phase durations for Treatments A and B

Pulsation cycle phase	Treatment A	Treatment B
Opening phase (a-phase) [ms]	142	142
Open phase (b-phase) [ms]	458	458
Closing phase (c-phase) [ms]	70	130
Closed phase (d-phase) [ms]	330	270

Milk Flow Characteristics

To record milk flow characteristics (Table 5) for evaluating physiological aspects, milk flow meters were installed at each milking stall during the test-period (LactoCorder[®], WMB AG, Balgach, Switzerland). Milk flow characteristics were processed by the software package LactoPro Software (LactoCorder[®] Software, Version 6.0.28, 2013; WMB AG, Balgach, Switzerland).

Table 5: Milk flow characteristics and their abbreviations

Milk flow characteristic	Abbreviation	Explanation (Steidle <i>et al.</i> , 2000)
Total milk yield	TMY	milk yield from beginning to end of milking process
Peak flow rate	PFR	maximum milk flow in the main milking process within a time interval of 8 measurement points
Time of main milking process by machine	duration MMT	time from milk flow > 0.5 kg/min at the start of milking until < 0.2 kg/min at the end of milking progress
Time of plateau phase	duration PLP	time from the vertex of the incline to the vertex of the decline
Time of decline phase	duration DP	time from the vertex of the decline until the milk flow falls below 0.2 kg/min
Time of over-milking by machine	duration OP	phase between main milking process (decline below 0.2 kg/min) and subsequent milking (incline to 0.2 kg/min)
Total milking period	duration TMY	time from the automatic start to the switch-off
Time of stripping phase	duration PMP	time needed for stripping
Subsequent milk yield	PMY	post-milking yield
Bimodal milk flow	BIMO	bimodal course of the milk flow incline; milk flow > 0.5 kg/min, a decline of > 0.2 kg/min and an increase of 0.5 kg/min within 38 s after the decline

Hind-Leg Activity

To evaluate ethological aspects, hind-leg activity of 18 focus dairy cows (7 Red Holstein x Swiss Fleckvieh and 11 Brown Swiss) were observed by three-dimensional accelerometer sensors (RumiWatch pedometer, Itin + Hoch GmbH, Liestal, Switzerland). Animals were equipped with an accelerometer on each hind-leg for each treatment once. Hind-leg position was distinguished between hind-leg turned towards milking pit and hind-leg turned away from milking pit. For evaluation of the hind-leg activity, accelerometers ran synchronized with milk flow meters and data were edited in four milk flow phases (Table 6).

Table 6: Milk flow phases for hind-leg activity measurements

Phase	Description	Abbreviation
1	plateau phase	PLP
2	decreasing phase	DP
3	over-milking phase	OP
4	post-milking phase	PMP

The three-dimensional accelerometers measured the gravitational acceleration in three axes. The RumiWatch Converter (Version 0.7.2.0, Itin + Hoch GmbH, Liestal, Switzerland) listed data in form of an activity index with a sampling rate of 10 Hz for the three axes x, y, and z in Microsoft Excel (Microsoft Excel 2010, Microsoft Corp., Redmond, USA). Data were converted into accelerations [m/s^2]:

$$\text{Acceleration} = (\text{activity index} \times (1/56)) \times g$$

Further, accelerations were converted into absolute values, and axes x, y, and z were summed up for each milk flow phase. In addition, mean values and maximum values were calculated for each milk flow phase. The mean values expressed the average activity in the phases, and the maximum values highlighted the activity peaks during milking.

Statistical Evaluation

For the statistical evaluation, the linear mixed-effects model (“lme”; Pinheiro and Bates, 2000) was used for the milk flow characteristics TMY, PFR, duration main milking time (duration MMT), duration plateau phase (duration PLP), duration decreasing phase (duration DP), duration over-milking phase (duration OP), duration total milking time (duration TMY), and the activity parameters mean activity and max activity. The general linear mixed model (“glmm” method; Venables and Ripley, 2002) was used for the milk flow characteristics duration post-milking phase (duration PMP), post-milking yield (PMY), and bimodal milk flow (BIMO) due to distribution differences of the residuals. The statistical analysis was

performed in R 1.9.1 (R Development Core Team, 2011). Using stepwise backward eliminations, upper interactions not reaching significance were excluded. The parameter PFR was log transformed.

In the model calculation milking characteristics, target variables were TMY, PFR, duration MMT, duration PLP, duration DP, duration OP, duration TMY, duration PMP, PMY, and BIMO. The model contained the Treatments (A and B) as explanatory variable and lactation stage (early, mid, late) and lactation number (first, second and further) as co-variables. Animals and measuring days were included as random effects (nested).

The model calculation for activity contained the target variables mean activity and max activity, the explanatory variable Treatments (A and B), and the co-variables milk flow curve phases (PLP, DP, OP, PMP) and position of pedometer (turned towards milking pit, turned away from milking pit). Animals and measuring days were included as random effects (nested).

Results

Milk Flow Characteristics

TMY ranged between 10.25 and 20.45 kg (Table 7). TMY decreased significantly with increasing lactation stage ($F_{2,35} = 14.87$, $P < 0.001$) and increased significantly with increasing lactation number ($F_{1,35} = 15.72$, $P < 0.001$). A tendency was detected between the Treatments ($F_{1,379} = 3.45$, $P = 0.06$). Regarding the model calculation, cows milked in Treatment B showed on average a 0.21 ± 0.10 kg higher TMY than cows milked according to Treatment A.

Table 7: Mean values \pm SE for milk flow characteristics in Treatments A and B, subdivided in lactation number and lactation stage

Parameter	Treatment	First lactation			Second and higher lactation		
		Early stage	Mid stage	Late stage	Early stage	Mid stage	Late stage
TMY [kg/milking]	A	14.96 \pm 0.40	14.66 \pm 0.6	10.71 \pm 0.5	19.39 \pm 0.94	17.47 \pm 0.58	13.23 \pm 0.6
	B	15.00 \pm 0.42	15.45 \pm 0.6	10.25 \pm 0.5	20.45 \pm 0.59	17.74 \pm 0.52	13.72 \pm 0.6
PFR [kg/min]	A	2.84 \pm 0.12	3.46 \pm 0.44	3.71 \pm 0.24	3.91 \pm 0.15	3.90 \pm 0.19	4.11 \pm 0.11
	B	2.96 \pm 0.12	3.97 \pm 0.47	3.93 \pm 0.29	4.12 \pm 0.11	4.00 \pm 0.19	4.28 \pm 0.12
Duration [min]	A	7.38 \pm 0.32	6.32 \pm 0.19	4.70 \pm 0.23	7.41 \pm 0.41	7.06 \pm 0.42	5.41 \pm 0.30
MMT [min]	B	7.07 \pm 0.30	5.78 \pm 0.19	4.25 \pm 0.19	7.72 \pm 0.30	7.09 \pm 0.46	5.47 \pm 0.27
Duration [min]	A	4.11 \pm 0.38	3.16 \pm 0.49	1.62 \pm 0.17	2.81 \pm 0.20	3.27 \pm 0.26	1.63 \pm 0.10
PLP [min]	B	4.05 \pm 0.36	2.58 \pm 0.41	1.41 \pm 0.16	2.98 \pm 0.20	3.11 \pm 0.24	1.58 \pm 0.10
Duration [min]	A	2.53 \pm 0.20	2.41 \pm 0.14	2.10 \pm 0.10	3.76 \pm 0.16	2.89 \pm 0.20	2.65 \pm 0.11
DP [min]	B	2.33 \pm 0.16	2.33 \pm 0.14	1.89 \pm 0.07	3.95 \pm 0.12	3.09 \pm 0.24	2.83 \pm 0.11
Duration [min]	A	0.65 \pm 0.07	0.70 \pm 0.10	0.67 \pm 0.08	0.52 \pm 0.05	0.64 \pm 0.07	0.48 \pm 0.04
OP [min]	B	0.62 \pm 0.09	0.79 \pm 0.09	0.71 \pm 0.11	0.52 \pm 0.05	0.65 \pm 0.07	0.64 \pm 0.05
Duration [min]	A	8.60 \pm 0.29	7.44 \pm 0.27	5.78 \pm 0.27	8.55 \pm 0.28	8.38 \pm 0.32	6.61 \pm 0.15
TMY [min]	B	8.47 \pm 0.29	7.06 \pm 0.30	5.46 \pm 0.22	8.80 \pm 0.21	8.35 \pm 0.35	6.78 \pm 0.15
Duration [min]	A	0.16 \pm 0.09	0.03 \pm 0.02	0.03 \pm 0.01	0.22 \pm 0.08	0.17 \pm 0.07	0.20 \pm 0.04
PMP [min]	B	0.24 \pm 0.09	0.04 \pm 0.02	0.06 \pm 0.05	0.15 \pm 0.05	0.18 \pm 0.06	0.16 \pm 0.04
PMY [kg]	A	0.07 \pm 0.04	0.01 \pm 0.01	0.00 \pm 0.00	0.12 \pm 0.05	0.11 \pm 0.06	0.11 \pm 0.03
	B	0.14 \pm 0.07	0.01 \pm 0.00	0.02 \pm 0.02	0.06 \pm 0.03	0.12 \pm 0.06	0.08 \pm 0.02
BIMO [%]	A	0.13 \pm 0.07	0.36 \pm 0.15	0.29 \pm 0.10	0.25 \pm 0.06	0.15 \pm 0.06	0.33 \pm 0.05
	B	0.20 \pm 0.08	0.18 \pm 0.12	0.37 \pm 0.11	0.29 \pm 0.06	0.12 \pm 0.05	0.22 \pm 0.05

PFR ranged between 2.84 and 4.28 kg/min (Table 7). Lactation stage had no significant influence on PFR ($F_{2,23} = 0.85$, $P > 0.1$) whereas lactation number had a significant influence ($F_{1,35} = 4.80$, $P < 0.05$). The model calculation showed that the PFR at the second and further lactation was at least 1.24 ± 0.09 kg/min higher than at the first lactation. Furthermore, the PFR was significantly higher with 1.04 kg/min in Treatment B than in Treatment A ($F_{1,379} = 27.48$, $P < 0.001$).

Durations MMT took between 4.25 and 7.72 min (Table 7). With increasing lactation stage, duration MMT decreased significantly ($F_{2,35} = 9.72, P < 0.001$). There were no significant differences between lactation numbers ($F_{1,35} = 2.90, P > 0.1$) or between the Treatments ($F_{1,379} = 1.15, P > 0.1$).

Duration PLP ranged from 1.41 to 4.11 min (Table 7) and decreased significantly as lactation stage increased ($F_{2,35} = 7.61, P = 0.002$). Lactation number showed no significant differences ($F_{1,35} = 0.44, P = 0.51$). A tendency could be detected between the Treatments ($F_{1,379} = 3.50, P = 0.06$). Treatment B had a 0.1 min shorter PLP than Treatment A.

Duration DP ranged between 1.89 and 3.95 min (Table 7) and decreased with increasing lactation stage ($F_{2,35} = 3.86, P = 0.03$). A significantly longer duration DP was detected when lactation number increased ($F_{1,35} = 15.39, P = 0.0004$). No significant differences between the Treatments were recorded ($F_{1,379} = 0.84, P = 0.36$).

Concerning duration OP, no significant differences between lactation stages ($F_{2,35} = 1.02, P = 0.36$) and between Treatments ($F_{1,379} = 1.84, P = 0.18$) were found, but there was a tendential difference between lactation numbers ($F_{1,35} = 3.04, P = 0.09$). Duration OP ranged from 0.48 to 0.79 min (Table 7).

Regarding duration TMY, significant differences were detected between lactation stages ($F_{2,35} = 10.12, P = 0.0003$). As stage of lactation increased, duration TMY decreased. Durations lasted between 5.46 and 8.80 min (Table 7). With increasing lactation number, duration TMY slightly increased. There was a tendential difference between lactation numbers ($F_{1,35} = 2.7863, P = 0.1$) and no difference between Treatments ($F_{1,379} = 0.3451, P = 0.56$).

Duration PMP took between 0.00 and 0.24 min (Table 7), and PMY was between 0.00 and 0.14 kg (Table 7). No significant differences could be observed for duration PMP and for PMY between lactation stages, lactation numbers, or Treatments.

Concerning BIMO, no significant differences were found between lactation stages, lactation numbers, or Treatments.

Hind-Leg Activity

No significant differences in the mean values of hind-leg activity were found between the different milk flow phases. The mean activity expressed in accelerations ranged from 11.6 to 12.2 m/s² (Table 8). Nevertheless, a tendency was detected in the interaction between treatments and positions of the pedometer ($F_{1,351} = 3.49$, $P = 0.06$). Animals in Treatment A showed the highest mean activity in the hind-leg turned towards the milking pit, whereas animals in Treatment B showed higher activity in the hind-leg turned away from the milking pit.

Table 8: Hind-leg activity in accelerations and its SE in relation to the position of the pedometer, the Treatment, and the milk flow phases plateau phase (PLP), decreasing phase (DP), over-milking phase (OP), and post-milking phase (PMP) in m/s²

Position relative to milking pit	Treatment	Activity measurement	PLP [m/s ²]	DP [m/s ²]	OP [m/s ²]	PMP [m/s ²]
towards	A	mean	12.0 ± 0.26	11.8 ± 0.27	11.8 ± 0.28	12.0 ± 0.13
		max	19.9 ± 1.11	18.8 ± 0.87	16.0 ± 1.08	12.2 ± 0.12
	B	mean	11.6 ± 0.23	11.6 ± 0.22	11.6 ± 0.23	11.8 ± 0.32
		max	19.9 ± 1.11	21.5 ± 1.20	17.2 ± 1.31	13.7 ± 0.81
away	A	mean	11.7 ± 0.24	11.7 ± 0.22	11.6 ± 0.22	12.2 ± 0.18
		max	17.7 ± 0.96	20.9 ± 1.28	18.3 ± 0.83	16.5 ± 0.98
	B	mean	11.8 ± 0.26	11.7 ± 0.23	11.7 ± 0.22	12.1 ± 0.23
		max	18.2 ± 1.15	18.9 ± 0.92	19.6 ± 0.72	14.9 ± 0.73

Significant differences were detected between the phases in the maximum values of hind-leg activity, as they decreased with milk flow curve progression ($F_{4,351} = 24.210$, $P < 0.001$). The maximum activity expressed in accelerations ranged from 12.2 to 21.5 m/s² (Table 8). There was a tendency in interaction between Treatments and positions of the pedometer ($F_{1,351} = 3.3149$, $P = 0.07$). Animals in Treatment A showed the highest maximum activity in the hind-leg turned away from the milking pit, whereas animals in Treatment B showed only a slightly higher maximum activity in the hind-leg turned towards the milking pit.

Discussion

The results show that the treatment allocation was based on a good stratification of the sample cows, as TMY of dairy cows (Brown Swiss and Red Holstein x Swiss Fleckvieh) was on average 15.1 kg/milking in Treatment A and 15.4 kg/milking in Treatment B. Milking time (duration TMY) was on average 7.6 min/milking in Treatment A and 7.5 min/milking in Treatment B. These milking times agree with the equation of Stewart *et al.* (1993), where the average milking time per cow yielding 10 to 20 kg of milk per milking is calculated as average milking time per cow [min] = $3.6 + 0.26 \times \text{milk yield per cow per milking [kg]}$. We found that duration TMY decreased as lactation stage increased and thereby confirmed the findings of Bruckmaier *et al.* (1995). Lactation number had no effect on duration MMT, which was contrary to the results of Antalík and Strapák (2010) and Strapák *et al.* (2011), who found longer durations of MMT with advancing lactation number. Our findings that TMY decreased with increasing lactation stage and TMY and PFR increased with increasing lactation number support previous research results (Dodenhoff *et al.*, 1999; Antalík and Strapák, 2010; Strapák *et al.*, 2011).

In this study, milking time durations had a balanced ratio between duration PLP and duration DP. Duration PLP was on average 2.8 min in Treatment A and 2.6 min in Treatment B. Advantageous milk flow curves show a long plateau phase where the major part of milk yield is removed (Wessels, 2002). Especially long decreasing phases can increase milking time, which can be the consequence of a stepwise udder quarter emptying. We found that the decreasing phase duration was on average 2.7 min in both treatments. A long decreasing phase can increase the risk of over-milking of single udder quarters (Steidle *et al.*, 2000), and an increased PMY can be the result of fear reactions (Rushen *et al.*, 1999). Durations OP and PMP were short. Durations OP were on average 0.6 min in Treatment A and 0.7 min in

Treatment B, and durations PMP were on average 0.1 min in both Treatments. Automatic stripping and cluster removal prevented post-milking and serious over-milking.

The low percentage of dairy cows showing BIMO in Treatments A (on average 0.3 %) and B (on average 0.2 %) indicated an appropriate pre-stimulation, as bimodal milk flow curves mostly indicate an insufficient stimulus (Dodenhoff *et al.* 1999). To prevent BIMO, dairy cows were subjected to standardized pre-stimulation procedures, and automatic stimulation was set considering lactation stage. Consequently, neither lactation stage nor lactation number did affect BIMO in this study. Wessels (2002) and Strapák *et al.* (2009) reported increased BIMO in cows with advanced lactation number. Therefore, cows with advanced lactation number need to be stimulated more intensively for the milking process. In Strapák *et al.*'s (2011) study, BIMO was positively correlated with the duration of the incline phase of milk flow curve.

In this study, the c-phase duration was elongated from 70 to 120 ms. Billon and Gaudin (2001) observed elongated c-phase durations by elongating the a-phase duration settings as well. Hence, their results were a consequence of both a- and c-phase changes.

The absolute milking time (duration TMY) is a factor that influences working time during milking, and reducing labor during milking is valuable for farmers (Boettcher *et al.*, 1998). Furthermore, dairy cow welfare is affected by long durations of TMY because they increase the time when the teats are subjected to the vacuum. Billon and Gaudin (2001) found duration TMY and milk flow rates to be the parameters most dependent on the a- and c-phase durations. In Kochman *et al.*'s (2008) study, TMY decreased when c-phase durations were shortened from 120 to 90 ms. Additionally, Billon and Gaudin (2001) and Kochman *et al.* (2008) found that c-phases of 120 ms led to an increased PFR, whereas c-phase durations of 90 ms produced lower flow rates. In this study, c-phase duration setting had no effect on duration TMY. When the c-phase duration was prolonged, TMY tended to be 0.21 kg higher

and PFR was 1.04 kg/min higher. In Treatment B, a higher milk yield was obtained in the same milking time as in Treatment A, showing that PFR was increased. There were no strong differences between shortened and elongated c-phases in the durations of decreasing, over-milking, and post-milking phases. In Bluemel *et al.* (*in prep.*), observations concerning liner movement revealed that the open liner phase increased when the closing phase was decelerated. Thus, higher milk yields might be achieved due to an increased suction phase (a- and b-phase). Additionally, Roşca *et al.* (2010) described that milk flow started at about 25 to 50 % into the a-phase, continued through the b-phase, and extended into the first part of the c-phase. The extended c-phase caused a less rapid closing of the liner after the b-phase and thus prolonged the duration of milk removal. Consequently, we can assume that an elongated c-phase could slightly improve the milk-out grade, which is obligatory to sustain high milk yield and animal health (Worstorff *et al.*, 1980; Bruckmaier and Blum, 1998; Steidle *et al.*, 2000). Hömberg (2013) suggested leaving no more than 300 mL of milk in the udder. He assumed that too slow and incomplete milking resulted from inferior milking technique, such as insufficient stimulation, crooked clusters, pulsator failings, and liners of wrong size or bad quality.

To find out if dairy cows are exposed to stress when milked with a short c-phase or if differences in milk flow characteristics between shortened and prolonged c-phases are only physiological, we measured hind-leg activity of dairy cows. The parameter mean activity should provide information about the general restlessness of the animals during milking. The parameter maximum activity was used to explicitly show outliers. Kochman *et al.* (2008) reported the appearance of physical discomfort and back-spray with increased speed of the c-phase, which would have negative consequences on milking performance. Seabrook (1994) and Rushen *et al.* (2001) considered that dairy cows milked in stressed situations gave reduced milk yields. Wenzel *et al.* (2003) and Gygax *et al.* (2008) concluded that cows with a higher activity during milking were exposed to an increased level of stress or nervousness.

Billon and Gaudin (2001), Kochman *et al.* (2008), and Albers (2011) assumed that the punching of the liner on the cow's teat caused discomfort and led to more activity and milk ejection disturbances. Therefore, Albers (2011) recommended taking particular care in the c-phase so that liners do not close too fast and therefore do not hit the teat promptly.

We found a decrease in maximum activity values during milk flow curve progression, but there were no differences between the mean activity values. Discomfort might be assumed in the decreasing phase and the over-milking phase, as the liner penetrates the teat with machine vacuum although milk flow decreased or ceased, especially when cows are milked with a shortened c-phase. Therefore, an increase in activity would have been expected with milk flow phase progression. Our results might be explained by the non-existent over-milking and post-milking phases in several experimental dairy cows. Activity measurements should be performed with dairy cows having teat problems and disturbed milk removal. A tendency in interaction of c-phase duration and position of the pedometer was observed in this study. When dairy cows were milked with a short c-phase, they showed the highest mean activity in the hind-leg turned towards the milking pit, but the maximum activity in the hind-leg turned away from the milking pit. However, differences were little and might have been a coincidence. Therefore, no clear conclusion can be drawn. Additionally, the aspect of increased activity due to lameness was not considered. Whereas Rousing *et al.* (2004) found no evidence that lameness or other signs of leg disorder were correlated with stepping and kicking during milking, Hassall *et al.* (1993) showed that lame cows kicked less at units but were more restless during milking, suggesting discomfort.

Possible reasons for no clear differences in the results could be the habituation of the experimental dairy cows to a pulsator with a short c-phase and high air accelerations before experiment start. However, it rather seemed that dairy cows were milked in overall good conditions as activity was generally low during experiments in both Treatments. Serious stress

because of pulsation settings can be excluded. Seabrook (1994) noted that acute stress enabled adaptation. It would therefore be useful to run experiments on a dairy farm with cows showing increased activity during milking as increased activity could be an indicator for milking problems.

The influence of the different c-phase durations on teat health was not explicitly observed in this study. The test period was limited to 12 morning milkings to minimize time effects. A long-term study would be needed to observe the influence on teat health,. Billon and Gaudin (2001) found no relevant effects on the udder health and teat conditions when the c-phase was elongated or shortened, although they detected a small tendency for having a poorer teat-end condition when animals were milked with shorter c-phases.

Finally, the pulsator RotoPuls[®] used in the experiments, contrary to commercial electric pulsators, did not reach a critical pressure in air accelerations. Punches in the beginning of the a-phase, during transition from a-phase to b-phase, and in the c-phase were thus eliminated, possibly explaining why experimental dairy cows did not show differences in activity between the shortened and the elongated c-phase in the two Treatments.

Conclusions

An increased total milk yield and peak flow rate was found in dairy cows when milked by a prolonged closing phase, and therefore shortened closed phase of the pulsation chamber cycle. These results indicate to be beneficial for dairy cow's health regarding complete emptying of the cow's udder. Further, higher milk production levels are of economic relevance. However, no improvement in dairy cow comfort was found. This could be attributed to the novel pulsator used in this study, which decreases air accelerations even when transition phases are short. Further, research should focus on milking machines with dairy cows showing restlessness during milking.

Acknowledgements

We gratefully thank the Swiss Machinery Association (SLV) for their funding and Erwin Bilgery, Alfred Kummer, Ralf Fischli, Stefan Mathis, Christoph Bühler, Marianne Cockburn, Yamenah Gomez, and Janika Lutz for their assistance and support.

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Chapter 4**Automated detection of hind-leg activity in dairy cows via the milking cluster during milking****Franziska E. Bluemel, Sebastian Fricker, Pascal Savary, Matthias Schick**

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*Submitted to ELSEVIER APPLIED ANIMAL BEHAVIOUR SCIENCE***Abstract**

The study investigated if it is possible to automatically measure hind-leg activity during milking to decrease labor and costs, by attaching accelerometers to the milking cluster. This would reduce the numbers of accelerometers needed and minimize the additional effort, such as attaching, detaching, charging of battery, and reading out data. The objective of this study was to investigate the correlations of hind-leg activity measured by accelerometers attached to the hind-legs and the milking cluster with direct observations of hind-leg activity. An increased hind-leg activity of dairy cows during milking is considered to be indicative of stress, discomfort and pain, thus impairment of welfare. The present study was performed in the auto tandem milking parlor at Agroscope Taenikon Research Station in Ettenhausen, Switzerland. Hind-leg activity measurements of 35 dairy cows (Brown Swiss and Red Holstein x Swiss Fleckvieh) were recorded during one morning and one evening milking. Direct observations were conducted simultaneously. Hind-leg accelerometer data and milking cluster accelerometer data were graphically evaluated regarding the morning milkings.

Further, the number of active phases, the durations between active phases and an individual threshold for differentiating active and inactive phases were defined. To test the correlation of that dynamic threshold, the algorithm was applied to the evening milking data. The correlation between milking cluster accelerometer data and direct observations was 74 % ($P < 0.001$), whereas the correlation between hind-leg accelerometer data and direct observation reached 91 % ($P < 0.001$). Satisfying high correlations with the direct observations were achieved in both, when attaching an accelerometer to the milking cluster and when attaching an accelerometer to the hind-legs. However, hind-leg activity measurements were most accurate when attaching accelerometers to the hind-legs of dairy cows. Measurements took place with one type of milking cluster and therefore, further investigations are needed to confirm these results. With the successful application of the algorithm, an automatic and indirect activity measurement method has been developed. Therefore, measuring hind-leg activity of dairy cows during milking could be simplified as well as reducing labor and costs. In the future, this innovative method could be implemented in the milking parlor or in the automatic milking device as a diagnostic tool providing valuable information, such as restlessness, to the farmer and consultant in a management program.

Keywords: early warning system, three-dimensional accelerometer, discomfort, health monitoring, restlessness

Introduction

Direct observations and video recordings are often used to observe dairy cow behavior during milking (Breuer *et al.*, 2000; Hagen *et al.*, 2004; Gygax *et al.* 2008). However, a consecutive sampling of direct observations is not practicable as it is time-consuming and costly (Watanabe *et al.*, 2008; Nielsen *et al.*, 2010).

Therefore, hind-leg activity measurements with accelerometers attached to cows' hind-legs are common in use to measure behavior such as resting, eating and walking (Guo *et al.*, 2009; Robert *et al.*, 2009; Blomberg, 2011). Correlations between accelerometers measuring animal behavior and direct observations were $r = 0.75$ in the study of Müller and Schrader (2003) for resting, standing, eating and walking and $r = 0.96$ in O'Driscoll *et al.* (2008) for standing and resting. The high r -values reveal the potential of using accelerometers for cow behavior analysis.

Although recording animal behavior with accelerometers is non-invasive, attaching accelerometers on dairy cows' hind-legs is mandatory for hind-leg activity observations. Consequently, a large quantity of accelerometers is needed for monitoring each dairy cow. Further, it takes additional effort for attaching, detaching, charging of battery, and reading out data. Besides, MacKay *et al.* (2012) recommended a habituation phase for accelerometers of two days before the onset of an experiment.

Hind-leg activity during milking is indicative for stress, discomfort and pain (Willis, 1983; Hemsworth *et al.*, 2000; Hanna *et al.*, 2006). As restlessness during milking is considered an important parameter for impairments of dairy cow welfare, a standardized measuring procedure for this behavior may be important for the detection of deficient milking conditions.

Therefore, the aim of the present study was to investigate the correlation between direct observations of hind-leg activity and hind-leg activity measured by means of accelerometers that were attached to the hind-legs of dairy cows and to the milking cluster for automated restlessness recording during milking. It was assumed that the milking cluster is set in motion when the hind-legs of dairy cows move, because the milking cluster is freely suspended on the cow's udder.

Materials and Methods

Animals and Design

The present study was conducted at Agroscope Taenikon Research Station in Ettenhausen, Switzerland. Thirty-five dairy cows (18 Brown Swiss and 17 Red Holstein x Swiss Fleckvieh) were monitored during milking in a low-line 2 x 3 auto tandem milking parlor (GEA Farm Technologies GmbH, Bönen, Germany) in March 2014. Milking clusters used were the GEA “Classic 300” (GEA Farm Technologies GmbH, Bönen, Germany) consisting of nitrile rubber liners and short silicon milking tubes. Each milking cluster was attached to a service arm and an automatic cluster removal. Animals showing udder and claw infections were excluded. Dairy cows were habituated to milking procedures and measurement devices two weeks prior to treatments. For 12 days, data were randomly recorded twice for each cow, once during morning milkings (04.00 h) and once during evening milkings (16.00 h). Milking was performed by the same milker using consistent preparation procedures. Preparation time from pre-milking to cleaning, followed by unit attachment was set to approximately one min.

Measuring Method

Three dimensional accelerometers (MSR 145, MSR Electronics GmbH, Seuzach, Switzerland) were used to record hind-leg activity in dairy cows. The sampling rate of sensors was 50 Hz with a sensitivity of $147 \text{ m/s}^2 \pm 1.47 \text{ m/s}^2$. Accelerometers were placed on the cow’s right and left metatarsi and attached by a Velcro strap. Activity was also recorded by attaching accelerometers to the hooks of the six milking clusters by adhesive tape. A pilot study conducted prior to the experiment revealed that attaching the accelerometers to the hooks of the milking clusters was advantageous. At this location, disturbing fluctuations produced by the long milking tube and the automatic disinfection of neighboring milking

clusters were lowest compared to other locations on the milking cluster. Further, accelerometers were protected best in this position.

Data Processing and Evaluation

Initially the 50 values per second from the accelerometer data were transformed in absolute values for the x, y and z axes. Further, absolute values of the x, y and z axes were summed up for each sampled value. For data processing, accelerometer data from the left and the right hind-leg were combined by selecting the maximal value for each value to evaluate the total activity of hind-legs. Subsequently, a moving average for 50 absolute values was utilized and the sampling rate was reduced, selecting the maximum value of the moving 50 absolute values per second for not flattening hind-leg activity.

Data recorded by the milking cluster and hind-leg accelerometers at the morning milkings were used to create the algorithm. Thus, graphs of 18 cows were visually evaluated and data of remaining 17 cows were used for verifying. Subsequently, this algorithm was applied to the data from the evening milkings for validation.

To create the algorithm and validate hind-leg activity by means of accelerometers, direct observations of dairy cows' hind-leg activity were recorded with a sampling rate of 1 Hz and classified as active and inactive phases. Any kind of hind-leg movements, such as stepping and kicking were defined as active phases. Therefore, stepping and/or kicking within 5 s were summarized to one active phase. No hind-leg movement were defined as inactive phases. The edited values of accelerometers were compared to the direct observations.

To examine the level of agreement between the hind-leg accelerometer data and the milking cluster accelerometer data, the Bland Altman Method (1986) was used to determine outliers and the distribution for the morning milkings and plotted the differences between the two techniques (Eq. (1)) against the averages of the two techniques (Eq. (2)). The upper and lower

confidence intervals were calculated for the “limits of agreement”, which were defined as the mean difference plus and minus 1.96 times the standard deviation of the differences.

Differences between the techniques =

$$\text{hind-leg accelerometer data} - \text{milking cluster accelerometer data} \quad (1)$$

Averages of the techniques =

$$(\text{milking cluster accelerometer data} + \text{hind leg accelerometer data})/2 \quad (2)$$

An algorithm was created by means of the morning milking data (reference). Therefore, correlations between hind-leg activity recorded by accelerometers attached to the milking cluster and direct observations were calculated. This algorithm was based on the threshold used in the study of Müller and Schrader (2003). The associated standard deviations were added to the mean values of all recorded accelerations for every milking (Eq. (3)). It was applied on each dairy cow’s morning milking by graphically comparing hind-leg activity of accelerometer data recorded by the milking cluster and hind-legs with its direct observations (Figure 4).

$$\text{Threshold} = \frac{1}{n} \sum_{i=1}^n x_i + \frac{1}{n-1} \sum_{i=1}^n (x_i - \bar{x})^2 \quad (3)$$

To increase the accuracy of the threshold, a constant acceleration value of 0.2 was added, which was derived from the average deviation of the visual threshold and the threshold mean value + standard deviation, creating the algorithm as follows (Eq. (4)).

$$\text{Algorithm} = \frac{1}{n} \sum_{i=1}^n x_i + \frac{1}{n-1} \sum_{i=1}^n (x_i - \bar{x})^2 + 0.2 \quad (4)$$

Accelerations recorded by accelerometers above the algorithm were considered as active phases, whereas accelerations below the algorithm were considered as inactive phases. The number of active phases was compared graphically to direct observations.

Subsequently, this algorithm was applied to the data recorded by accelerometers attached to the hind-legs and to the milking clusters from the evening milkings (validation).

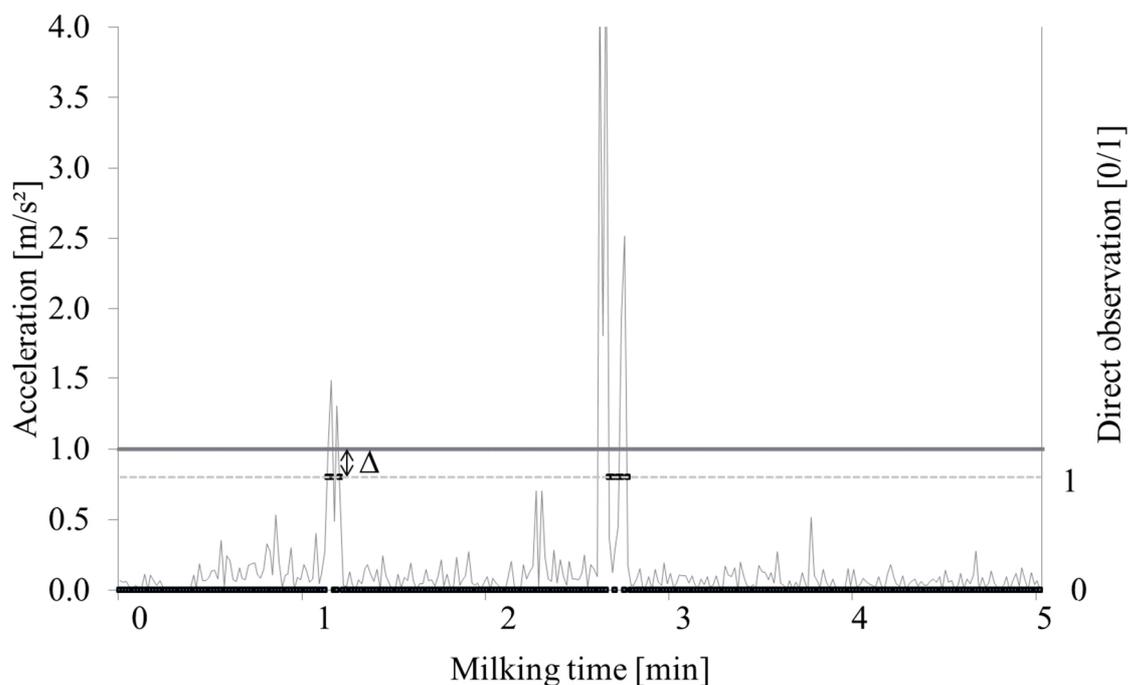


Figure 4: Example of the graphical evaluation by means of the calculated algorithm; the algorithm was derived visually (grey line; threshold + Δ). Δ is the difference between “absolute maximum values per second of the hind-leg activity recorded by the milking cluster (solid grey line)” and the “threshold consisting of mean value and standard deviation (dotted grey line)” in m/s^2 (in average 0.2 m/s^2). Direct observations are marked as black dots (0 = inactive phase, 1 = active phase). Activities taking place within 5 s were counted as one activity.

Statistical Evaluation

The Software R Version 3.1.0 (R Development Core Team, 2014) was used for the statistical evaluation. The graphical analysis for verifying the level of agreement between the measurement methods was established by using the Bland Altman Method (Bland and Altman, 1986). Calculating the correlation of the milking cluster activity data with the direct observations (the morning milkings), the algorithm = threshold + 0.2 m/s² was created and validated with the evening milking data by means of Pearson correlations. All data were visually checked for normal distribution.

Results

The level of agreement examined by means of the Bland Altman Method was 0.6 units. The middle line in Figure 5 indicates the mean difference of 0.6 units. Therefore, the mean difference of 0.6 units demonstrates higher activity values for the hind-legs as for the milking clusters. The lower and upper solid grey lines indicate the confidence interval of 95 %. All values were situated in this interval and therefore highly consistent.

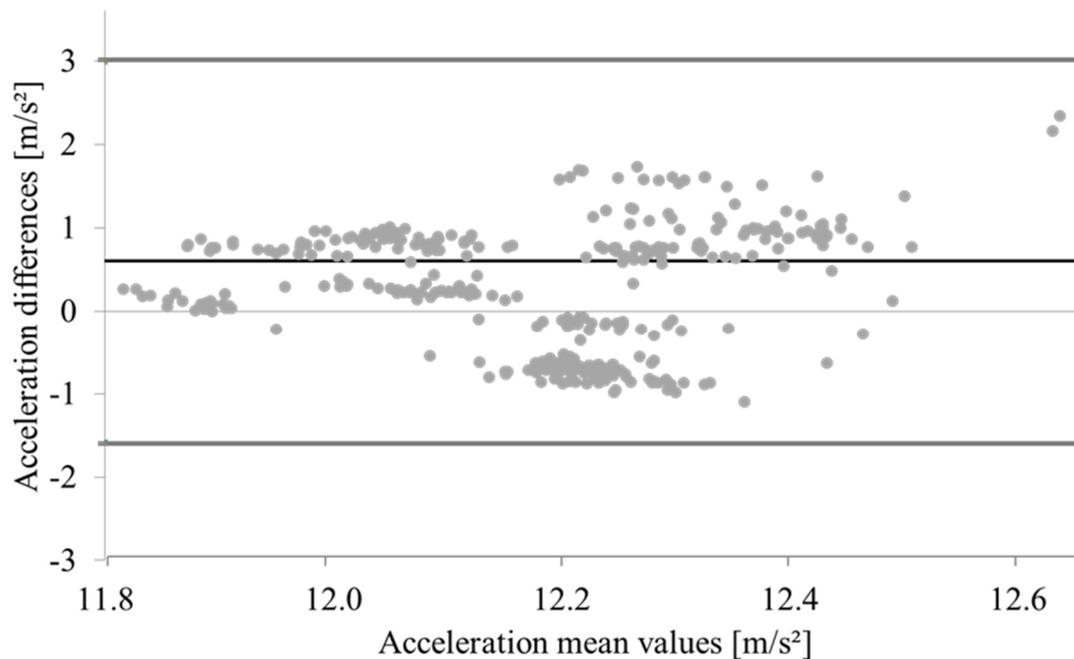


Figure 5: Graphical illustration of level of agreement for morning milkings.

In the Bland Altman Plot, the differences between the two techniques (hind-leg accelerometer data – milking cluster accelerometer data) were plotted against the averages of the two techniques $((\text{milking cluster accelerometer data} + \text{hind leg accelerometer data})/2)$ (grey dots). The mean value of differences was 0.6 units (black line) in the range of the 95 % confidence interval (lower and upper grey line).

The algorithm was verified with the data recorded by accelerometers attached to the milking cluster from the morning milkings, which were visually evaluated before. The correlation of “visually counted number of active phases from graphs” with “number of observations

calculated with the algorithm from the milking clusters” (morning milkings) was 97 % ($r_s = 0.97$, $n = 17$, $P < 0.001$; Figure 6).

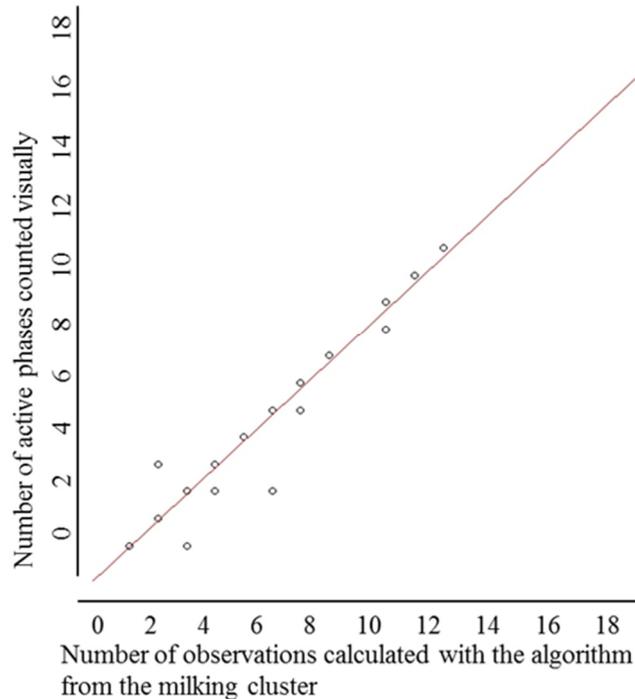


Figure 6: Correlation of “counted number of active phases from graphs” with “number of observations calculated with the algorithm from the milking clusters” (morning milkings).

The algorithm was validated by using it on the accelerometer data of the evening milkings. The result of the correlation between the “number of active phases on milking clusters using the algorithm” and the “number of active phases on hind-legs using direct observations” was 74 % ($r_s = 0.74$, $n = 35$, $P < 0.001$; Figure 7). The correlation between “number of active phases on hind-legs using the algorithm” and “number of active phases using direct observations” was 91 % ($r_s = 0.91$, $n = 35$, $P < 0.001$; Figure 7).

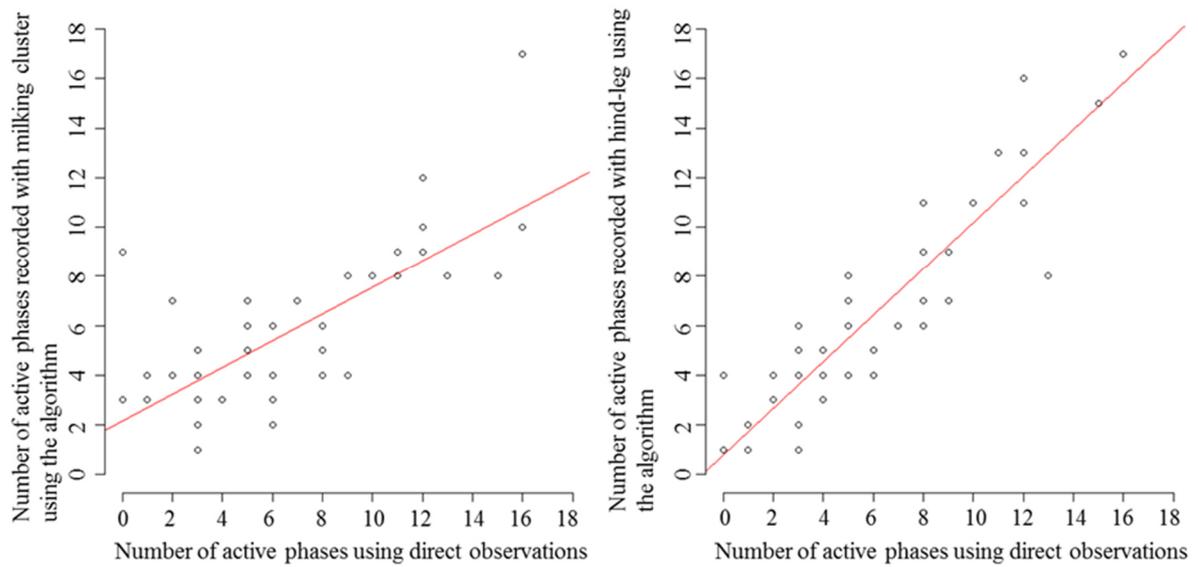


Figure 7: “Correlation between number of active phases recorded with milking cluster and hind-leg using the algorithm” and “number of active phases using direct observations” (evening milkings, $n = 35$).

Discussion

An accelerometer attached to the milking cluster may measure the hind-leg activity of the dairy cow indirectly. Therefore, accelerometer data was recorded for both hind-legs and compared to accelerometer data of the milking cluster. For data processing and evaluation of this indirect hind-leg activity measurement method, the axes x, y and z were summed up as in Bluemel *et al.* (in prep.) and the sampling rate of accelerometers was reduced from initially 50 to 1 Hz by selecting the maximum value out of the 50 values per second. In a previous study by Pastell *et al.* (2008), it was found out that kicking lasted 0.5 s on average. However, by selecting the maximum value out of every second when using a sampling rate of 50 Hz, activities such as kicking and stepping were automatically detected in the defined active phase in the present study.

An algorithm for automatic data recording was applied to determine individual thresholds in a similar way as in the study of Müller and Schrader (2003) described. The authors achieved a correlation of 75 % between accelerometers attached to hind-legs and video recordings when observing resting, standing, eating and walking. We adjusted the threshold with the constant value of 0.2 m/s^2 to increase the accuracy of the algorithm for the activity types during milking. Therefore, the correlation of 91 % was achieved between hind-leg activity recordings by accelerometers attached to the hind-legs of dairy cows. Further, the correlation of 74 % was still achieved with indirect hind-leg activity recordings by accelerometers attached to the milking cluster and direct observations.

However, regarding the counting of hind-leg activity, the interruptions between activity phases were not clearly identifiable. Consequently, the separation of successive peaks was not practicable for counting activity. Therefore, active and inactive phases were deemed to be completed when taking place in a time period of 5 s.

In previous studies, the frequency of restlessness behavior was applied to define stress in dairy cows (Wenzel *et al.*, 2003; Gygax *et al.* 2008). It was assumed that by means of recording the frequency of active and inactive phases, the indication of restlessness behavior could be given. Therefore, classifying hind-leg activity as active and inactive phases seemed to be a practicable method for recording restlessness behavior. The differentiation of activity patterns, such as stepping and kicking, was not possible at the milking cluster due to high variations between animals and within animals. Kicking could not be detected graphically by means of high acceleration occurrences. Reason for these variations could be the level of free suspension of the milking cluster as udder shape and udder conditions vary between cows and the different udder fillings, which change during the milking process. Further, the milking cluster is attached to the milking machine by the long air and milk tube and the service arm, which could lead to distortion of the accelerometer data. Additionally, twisted short air and milk tubes could lead to different activity patterns. Nevertheless, milking clusters touch the floor, when the base of the dairy cows' udder is low. In this case, problems with automatic activity recordings could occur, as the milking cluster would not be suspended freely to the cow's udder, making the recording impossible under these circumstances.

While investigating correlations, it was to be kept in mind that the accelerometers attached to hind-legs and to milking clusters would not show the same acceleration values at any time. Accelerometers attached to the milking cluster record hind-leg activity time-delayed and indirect as accelerometers are not directly attached to the animal. Hence, accelerations were lower at the milking cluster as at the hind-legs. Further, other disturbing movements, such as coughing, snorting, and twitching can lead to movements of the milking cluster as it was freely suspended to the cow's udder, although these movements are not indicating restlessness. Therefore, these movements had to be considered. However, we examined these movements in a pilot study and found no accelerations above the algorithm. Consequently, these movements were classified as inactive phases.

Rousing *et al.* (2004) concluded that measuring behavior parameters, such as hind-leg activity during milking, could reveal milking technology deficits and udder health problems. A continuous hind-leg activity monitoring during milking has the potential to draw conclusions on dairy cow's well-being and health status. If detecting average values for hind-leg activity, deviations could be a signal for impairments of welfare. Such impairments could be udder and teat infections or injuries, which might be expressed as increased hind-leg activity during milking due to pain or discomfort.

Conclusions

Laborious attaching of accelerometers on the hind-legs of dairy cows is no longer necessary with this innovative hind-leg activity monitoring method presented in this study. Recording hind-leg activity during milking with an accelerometer attached to the milking clusters was successful and enabled automatic calculation by means of an adjusted algorithm. With this novel method, costs and labor can be minimized and objective examination of animal behavior can be guaranteed. Implementing this innovative method in a management program as a diagnostic tool in a milking parlor or an automatic milking device, cow comfort, health status of dairy cows, milking routine and milking technology conditions could be analyzed, thus providing valuable information to milk producers.

Acknowledgements

We would like to thank Christoph Bühler and Stefan Mathis for their support in preparing and conducting the experiment. Our thanks also go to Janika Lutz and Yamenah Gomez for their support with the data analysis and proofreading.

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Chapter 5

General discussion

Chapter overview

Chapter 1

Machine milking does not function according to the same principle as calf suckling. Therefore, international standards need to be complied to ensure dairy cow's health and a high hygiene (ISO 5707, 2007). However, these international standards are minimum requirements for milking machine installations and the aspect "welfare" is not explicitly mentioned. Nevertheless, dairy cow welfare plays a major role in machine milking. Milking problems, such as unwillingness to enter the milking parlor, milk ejection disturbances, restlessness, defecation and urination often arise due to discomfort of dairy cows during milking (Savary *et al.* 2010). Milking problems can have many reasons, thus poorly adjusted pulsators might appear secondarily. But being exposed to an average of 60 pulsation cycles per minute for more or less 8 minutes twice per day for on average 305 days per year can be a serious problem for the cow and may not be underestimated.

However, durations of transition phases (opening and closing phases; a- and c-phases, respectively) are not defined in the ISO 5707 (2007) as previously mentioned by Billon and Gaudin (2001). But the present study shows that the c- and d-phase durations affect physical processes in the milking cluster and milk flow characteristics. Consequently, the transition phases in the pulsation chamber cycle need to be considered in the international standards and need to be defined on the basis of this and subsequent scientific studies.

Chapter 2

Results in the second chapter showed that the real liner movement varies during phase progression, which was also shown in Spencer and Jones (2000). Liner opening durations were correlated to milk flow rates. This effect was less obvious when the c-phase was prolonged (Treatment B) and, liner opening movement durations during the milking process were more stable with phase progression. With stable liner open movement durations, less unpredictable changes took place. This might be less stressful for dairy cows. However, durations of the liner being open were increased, and thus the vacuum penetrated the teat for a longer time in Treatment B than in Treatment A (i.e. short c-phase). This might explain the higher milk yield and the increased highest milk flow rate for Treatment B detected in the experiment presented in the third chapter.

The pressure sum was lower in Treatment B than in Treatment A. A too tremendous over-pressure can lead to teat damage and can therefore risk the health of the cow (Mein and Reinemann, 2009). In this study, the pressure sum was found to be in the recommended range given by Spohr (2012).

For both treatments, the RotoPuls[®] pulsators were configured for air streams not reaching the critical pressure. Therefore, in contrast to conventional electric pulsators, high accelerations in the beginning of the a- and c-phases, and during the transition from the a- to b-phase could not occur. However, the results showed that accelerations were less prominent in the plateau phase (PLP) and over-milking phase (OP) in Treatment B than in Treatment A. This could prevent the occurrence of “impact” (Schlaiss, 1994) and keep dairy cows healthier, particularly the udder (Stanley *et al.*, 1962; Thiel *et al.*, 1973). Whereas the focus of this study was on the effects of a prolonged c-phase with other conditions being constant, further research is needed to elucidate the potential advantages of using RotoPuls[®] pulsators compared with conventional pulsators.

Chapter 3

The extended c-phase caused an increased total milk yield (TMY) and peak flow rate (PFR) and, thus appeared to be advantageous. Billon and Gaudin (2001) observed stretched c-phase durations by elongating the a-phase duration settings, as well and found milking time (duration TMY) and milk flow rates to be the parameters most dependent on the a- and c-phase durations. In this study, TMY tended to be 0.21 kg higher and PFR was 1.04 kg/min higher, when the c-phase was prolonged. In chapter two, observations concerning liner movements revealed that the opened liner phase increased, when the closing phase (c-phase) was extended. The extended c-phase caused a less prompt closing of the liner after the b-phase and thus prolonged the duration of milk removal. Consequently, it can be assumed that an elongated c-phase could slightly improve the milk-out grade which is obligatory to sustain high milk yield and animal health (Worstorff *et al.*, 1980; Steidle *et al.*, 2000).

Kochman *et al.* (2008) considered the appearance of physical discomfort and back-spray with increased speed of the c-phase, which would have negative consequences on milking performance. Seabrook (1994) and Rushen *et al.* (2001) considered that dairy cows milked in stressed situations were reduced in milk yields. Wenzel *et al.* (2003) and Gygax *et al.* (2008) concluded that cows with a higher activity during milking are exposed to an increased level of stress or nervousness. Billon and Gaudin (2001), Kochman *et al.* (2008), and Albers (2011) presumed that the punching of the liner on the cow's teat causes discomfort and leads to more activity and milk-ejection disturbances. The present study found a decrease in maximum activity values during milk flow curve progression, but there were no differences between the mean activity values. However, it appeared that dairy cows were milked in overall good conditions as activity was generally low during milking before experiment start and during experiments in both treatments. Serious stress due to pulsation settings can be excluded.

Furthermore, the RotoPuls[®] pulsator used in the experiments, contrary to commercially available electric pulsators, did not produce emissions of structure-borne sound (Bilgery, 2011). Punches in the beginning of the a-phase, during transition from a-phase to b-phase, and in the c-phase were thus eliminated. Due to these eliminations in both treatments, dairy cows might not have shown activity changes between the shortened and the elongated c-phases.

Chapter 4

With diagnostic tools, milk producers and consultants receive detailed insights to incidents during milking concerning restlessness. Restlessness is attributed to discomfort in dairy cows. Conclusions concerning stress in animals during milking can be drawn by knowing the frequency of restlessness (Wenzel *et al.*, 2003; Gygax *et al.* 2008). Rousing *et al.* (2004) found that measuring behavior parameters during milking could expose milking problems concerning milking technology, udder health and milking routine quality.

As previously mentioned by Gygax *et al.* (2008), comparing studies on hind-leg movements was difficult because researchers used different types of classifications for stepping, foot-lifting, and kicking. Additionally, direct observations during milking is not advisable, because observing many dairy cows simultaneously is rather difficult, hence time consuming and disturbing for the milking routine.

Thanks to accelerometers, automatic data logging of hind-leg activity is possible during milking. However, as conducted in chapter two and three, attaching accelerometers on hind-legs is time consuming (attaching, detaching, charging, reading out data, programming). Additionally, two sensors per cow were in use, which was cost intensive. Further, the risk of data losses due to sensor defects or sensor loss in the barn was given. Hence, suggestions to simplify these procedures were considered in chapter four.

It was hypothesized that the milking cluster is set in motion when the hind-legs of dairy cows move, because the milking cluster is freely suspended on the cow's udder. Hence, in chapter four, hind-leg activity was measured indirectly by attaching an accelerometer on to the milking cluster. The objective of this experiment was to examine the correlation between direct observations of hind-leg activity and hind-leg activity measured by means of accelerometers that were attached to the hind-legs of dairy cows and to the milking cluster.

Using an algorithm, the correlation of hind-leg measurements by accelerometers attached to the hind-legs was 91 % and the correlation of hind-leg measurements by accelerometers attached to the milking cluster was 74 %. Such deviations were also found by Moreau *et al.* (2009).

Going through further stages of development, accelerometers attached to the milking cluster may serve as a useful diagnostic tool for closer characterization and identification of impairments in dairy cow welfare during milking, such as lameness, health issues, and estrus, as well as for detection of deficient technical installations in the milking parlor, hence, could be useful when comparing and evaluating milking routines, and milking equipment. Additionally, further research should also consider feasibility of automatic differentiation of hind-leg activities indicating restlessness, such as stepping and kicking. Consequently, scientists could use this innovative method for research in milking parlors or automatic milking devices and therefore, results could easier be compared with other scientific studies.

Customer benefit

The International Organization for Standardization delivers minimum requirements for the construction and performance of milking machines focusing on the animal's physiology and milk quality (ISO 5707, 2007). Additionally, national guidelines enhance these international standards. However, a study of Savary *et al.* (2010), in which dairy farmers complain about milking problems, shows that milking technology still needs to be optimized.

Benefits of this thesis are as follows:

- Offering detailed information about the effect of changing the duration of the closing phase in the pulsation chamber cycles. International standards should provide information concerning closing phase duration and incorporate dairy cow welfare. Guidelines play a major role for milking machine companies and consultants to install and adjust milking machines conscientiously.
- Securing teat health from a technical perspective. RotoPuls[®] pulsators may have particular suitability for automatic milking devices, due to their capability to keep air accelerations at the teat-end below the level of commercial pulsators. Hence, in an automatic milking device, in which milkings per cow and day exceed the number of milkings in conventional milking systems, adequate milking conditions are of particular importance.
- Providing solutions for optimized diagnosis. Laborious attaching of accelerometers on the hind-legs of dairy cows is no longer necessary with the diagnostic tool presented in this study. Thus, costs and labor are minimized, the risk of losses and defects is reduced and objectivity is guaranteed. Activity measurement could be implemented in the milking parlor or in the automatic milking device as a diagnostic tool sending valuable information to the milk producer and consultant in a management program. Thus, conclusions can be drawn concerning cow welfare and health, estrus, milking routine, and functionality of the milking parlor.

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- Ensuring animal welfare and health by minimizing milking problems. Optimized parlor installations and early detection of impairments in dairy cow welfare are expected to result in higher milk yields, reduced labor time and costs, and thus higher profitability of dairy farming.
 - Based on the results of this thesis, recommendations for optimizing milking machine installations and constructions should be developed. These recommendations could be used when planning new milking parlors and automatic milking devices or when restoring existing milking parlors and automatic milking devices.
 - Limit values for the transition phases of the pulsation cycle should be implemented in the Swiss national guidelines to ensure high milking standards.

Perspectives

The transition phases, i.e. opening and closing phases (a- and c-phases, respectively), are not mentioned in the international standards. However, the presented results show that the c-phase affects the physical processes in the milking cluster and the milk flow characteristics. Future research should focus on the effect of a prolonged c-phase on teat and udder health, since a prolonged c-phase might be gentler due to a lower pressure sum and to decreased air accelerations. Though, as the c-phase is prolonged, the d-phase is shortened. Therefore, it must be clarified, if the shortened d-phase is sufficient for a good blood circulation through the teats and if congestion and edema reduction is ensured.

In this study, changes took place in the duration of the c- and d-phase to keep the massage phase set at a duration of 1000 ms. Therefore, statements about elongated c-phases, respectively shortened c-phases in this study, imply a shortened d-phase, respectively elongated d-phase. If d-phase durations would have been remained constant in both treatments, the pulsation ratio would have been differing between the treatments. Examinations of different c-phase durations with constant d-phase durations would be recommended to be illuminated in a next step.

It is advised to run experiments on a dairy farm with cows showing increased activity during milking to outline the possible decrease of restlessness behavior when milked with a prolonged c-phase. Experiments concerning the influence of the a-phase should also be performed as no guidelines are defined, too. Subsequently, recommendations concerning a- and c-phases should be implemented in the international standards.

For hind-leg activity recording during milking, it should be aspired to classify and differentiate hind-leg movements in kicking and stepping. Additionally, it might be possible to draw conclusions from hind-leg activity during milking to detect estrus, lameness, mastitis and further impairments. Supplementary, one needs to investigate the efficiency of defining

active and inactive phases only as an indication of stress-related behavior measurement tool. Consequently, accelerometers on the milking clusters could be implemented as a diagnostic tool. Development of easy handling, regarding read out data, charging, and programming when attached permanently to the milking cluster, needs to be considered. The assumed working time requirement and cost reduction for attaching accelerometers to the milking clusters should be investigated to underline the positive advantages of this innovative method and to introduce it as a common scientific measuring device.

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Acknowledgement

I would like to express my gratitude to:

- *Prof. Dr. Matthias Schick for his unlimited support, guidance and reliance.*
- *Prof. Dr. Eva Gallmann and Prof. Dr. Heinz Bernhardt for the willingness to operate as co-reviewers.*
- *Dr. Pascal Savary for supervising and guiding me through this dissertation.*
- *The Swiss machinery association for its funding to enable this dissertation project.*
- *Erwin Bilgery, Ralf Fischli and Fredi Kummer for their assistance and support.*
- *The whole administration team of Tänikon, in particular: Diana Heer, Alma Modes, Sonja Pfister, Karin Sannwald and Rosmarie Senn.*
- *The work, buildings and system evaluation team.*
- *Sebastian Fricker and Flavio Ferrari for their assistance in this project.*
- *The entire staff of the experimental farm, in particular: Gallus Jöhl, Thomas Hämmerli, Christoph Bühler and Stefan Mathis.*
- *The technicians Hubert Bollhalder and Beat Kürsteiner for all advices, explanations and discussions concerning measurement techniques.*
- *The Ammersee friends for distraction.*
- *All my Tänikon fellows for the mental support, advices, proof readings, discussions and fun, particularly: Yamenah Gomez, Marianne Cockburn, Janika Lutz, Katharina Weber and Nils Zehner for always being on and by my side: "People like us, we don't need that much. Just someone that starts, starts the spark in our bonfire hearts" (Blunt, 2013).*
- *My family for their love, patience, care, support, advices and understanding.*

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Aadorf, 19th of May 2015