

## REACTION OF PRIMIPAROUS DAIRY COWS REARED IN EARLY POSTNATAL PERIOD IN DIFFERENT SYSTEMS ON MILKING CONDITIONS

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### ABSTRACT

An experiment was performed in primiparous dairy cows, which were reared as calves in different systems (with own mothers, with nursing cows, in individual hutches) before weaning at day 56. After weaning the animals were kept under similar conditions. During rearing period calves were exposed to some ethological tests except for animals from control group. The aim of the experiment was to determine their milking performance in response to first milking after calf removal and to stressor applied in parlour in the middle of lactation. Responsiveness to milking was monitored during the evening milking on day 8 (first milking after the weaning of calf), and on day 10 (third evening milking). On day 150 of lactation cows were subjected to stressor (unknown human in front of a head in parlour touching the cow). On day 152 of lactation the milking was usual. Rearing conditions and both treatments did not influence the parameters of milking performance. However, both treatments of first milking after calf removal and stressor at 150 day significantly increased residual milk volume ( $3.94 \pm 0.82$  vs.  $1.13 \pm 0.82$  kg,  $P = 0.0218$ ,  $0.58 \pm 0.30$  vs.  $0.11 \pm 0.30$  kg and  $P = 0.0294$ , respectively). In conclusion, the rearing conditions of heifers during the period before weaning could influence their response to milking conditions as primiparous cows fall in the range that is not of high importance for milking performance. However, higher residual milk volume indicates that heifers reared under own nursing cows express higher sensitivity to stressor during whole production life.

**Key words:** primiparous cows, rearing conditions, milking performance, stressor

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### INTRODUCTION

Stress situations occurring in dairy cows have an important influence on their health, welfare and production. Animals are exposed to various emotionally stressful situations during their life. The physiological response to stressful situations depends on the experiences obtained early post partum in cows (Brouček et al., 2000) or rats (Zimmerberg et al., 2003; Becker et al., 2007) or mouse (Cirulli et al., 2006). It was also confirmed in mouse that different housing had impact on emotional behaviour (Zhu et al., 2006). Emotionality significantly influenced the release of oxytocin during stimulation of mammary gland (Tančín and Bruckmaier, 2001). The dairy parlour can be considered as an unknown surrounding for primiparous cows and together with

the loss of calf it may cause emotional stress. In such animals higher residual milk fraction was also detected (Van Rennen et al., 2002). The relocation of animals to unfamiliar and also to familiar surroundings negatively influenced milk yield (Mačuhová et al., 2002; Tančín et al., 2000). Animals usually get adapted to the new surroundings with in a few days to have normal milking performance (Bruckmaier et al., 1996). The welfare of cows during milking is important for fast and complete milk removal (Tančín et al., 2006).

Cows can recognize individual people. Due to this fact it was proved that aversive handling of cows by certain people cause cows to become fearful of those people. The presence of an unknown or aggressive handler at milking can cause a decrease of milk yield, increase of residual milk and lead to the elevation of heart rates (Tančín and

Brouček, 1996; Rushen et al., 1999).

Recently we have shown that rearing conditions of heifers influenced their response to milking in unknown surroundings (Tančin et al., 2004). The goal of this work was to examine whether and how primiparous cows, which were bred in different rearing systems as calves early after birth, can adapt to milking early post partum and respond to stressor applied in parlour during milking in the middle stage of lactation.

## MATERIAL AND METHODS

### Animals, Housing and Management

Animals after calving were used in this experiment. These primiparous cows were reared as calves in three different rearing systems during their first two months after birth until weaning. During this period calves were fed starter mixtures and lucerne hay *ad libitum*.

Experimental primiparous cows after parturition were divided according to their rearing conditions as calves before weaning in day 56 of life into following groups:

- a) With own mother: During the first 21 days of life they were housed in individual pens with own mother. Then they were kept in common group pens in loose housing in balanced age groups until weaning and were fed by milk from a bucket with a nipple.
- b) With nursing cows: First three days they were with own mother in individual maternity pens. Then they were placed in group housing with nursing cows until weaning. The number of calves per nursing cow was determined according to their milk yield (6 kg milk per calf).
- c) In individual hutches: During the first day of life the calves were with their own mother in individual maternity pens. From 2nd to 56th day they were in individual hutches. From the 4th day they were fed with milk replacer from a bucket with a nipple until weaning.

In all three above-mentioned experimental groups the animals were subjected to behavioural tests during the period from weaning to calving while human contact was required very often. Therefore, to see the possible effect of human presence in above-mentioned groups another "control group" was selected where human contact to animals during rearing period was limited to just necessary interventions of management.

**Control group:** First day after parturition the calves were in individual pens with own mother. From 2nd to 56th day they were in hutches and fed with milk replacer from a bucket with a nipple.

After weaning calves of all groups were kept in loose housing. Each of them received 1.5 kg of concentrate mixture and lucerne hay offered *ad libitum* from weaning

till day 180. From the age of 90 days they received maize silage. From the 181st day, all heifers were fed according to Slovakian recommendations in order to attain 0.75 kg daily gain.

After parturition the primiparous cows were milked in a herringbone parlour 2 x 5 stalls with vacuum level 50 kPa, pulsation rate 55 c. p. m. and pulsation ratio 60:40. The cows were milked twice a day at 05.00 and 16.00 h. The udder preparation for milking consisted of fore-stripping and cleaning. A milking cluster was attached to the udder within 1.5 min of udder preparation. The cows had free access to a mixed ration providing energy and nutrients for the production of 20 kg milk and received additional concentrates depending on their production (0.45 kg concentrates per kilogram milk).

### Experiment I.

The effect of different rearing systems of dairy calves until weaning on their later adaptation to milking was studied in 24 Holstein cows (6 with own mothers, 5 nursing cows, 6 in individual hutches and 7 in control group). Two evening milkings were recorded from each heifer on the first and the third day after calf removal and cow relocation to the herd. The calf removal and relocation of cow to the herd was carried out on day 8 of lactation at 9:00 a.m.

### Experiment II.

The effect of presence of an unknown person during milking was tested on 27 Holstein heifers reared in different conditions (5 with own mothers, 6 nursing cows, 5 in individual hutches and 11 in control group) in the middle stage of lactation. Measurements were taken on day 150 and 152 of lactation during evening milking. On 150th day of lactation cows were milked in presence of an unknown person in front of their heads, who touched their neck or head. On day 152 cows were milked under usual conditions.

During both experimental procedures oxytocin was injected intramuscularly at a dose of 50 IU after stripping. The cows were milked again in order to remove remaining milk, including residual milk 3 minutes after oxytocin injection. The milking performances were recorded using a mobile system Lactocorder®. Milk samples from individual cows were collected to measure somatic cell counts using FOSSOMATIC devices.

### Statistical Analysis

SAS statistical software - procedure MIXED (SAS, ver. 8.2, 2001) was used for statistical analysis. Linear model with fixed and random effect was used to model the studied traits. Statistical model can be expressed in the following form:

$$y_{ijkl} = TRT_i TRT(HOUSING)_{j,i} + cow_k + e_{ijk}$$

where:

$y_{ijkl}$  - were the measurements for time of milking, total yield, yield in first three minutes during milking;

$TRT$  - treatment, first milkings after calf weaning or milking in human presence ( $i=1,2$ );

$TRT(HOUSING)_{j,i}$  - the fixed effect of  $j^{\text{th}}$  housing with in  $i^{\text{th}}$  treatment ( $i=1,\dots,3, j=1,\dots,3$ );

$cow_k$  - random effect of cow,  $cow \sim N(0, I \sigma_c^2)$ , ( $k=1,\dots,33$ );

$e$  - random error, assuming  $e \sim N(0, I \sigma_e^2)$ .

Differences between the levels of the treatment within type of housing were tested by Scheffe multiple range test for studied traits. In tables, data are presented as least square means (lsmeans) and standard error.

## RESULTS

### Experiment I.

Milking performance was not influenced by the order of milking and conditions of rearing (Table 1, 3) with an exception of residual milk volume. There was a significant effect of milking order on the volume of residual milk. The amount of the residual milk during the first milking was  $3.94 \pm 0.82$  kg and during the third milking was  $1.13 \pm 0.82$  kg (Table 1). The same effect was observed within reared groups but results were not significant (Table 3). Although the differences were non-significant, primiparous cows reared as calves by own mother or nursing cows had numerically higher volume of residual milk than the other two groups.

### Experiment II.

Milking performance was not influenced by the presence of an unknown human being in front of the head of cows and also conditions of rearing during milking on day 150 as compared with normal milking on day 152 (Table 2) except for the residual milk volume. There was a significant effect of human presence on the volume of residual milk. The amount of the residual milk during milking in human presence was  $0.58 \pm 0.30$  kg and without  $0.11 \pm 0.30$  kg (Table 2). The same effect was observed within reared groups but results were not significant (Table 4). Numerically, primiparous cows reared by own mothers or nursing cows had a higher volume of residual milk under stress situations as compared to the other two groups of cows.

**Table 1: Milking performance during 1<sup>st</sup> and 3<sup>rd</sup> evening milking after calf removal and relocation cows to the herd**

	Milking after calf weaning		
	first milking	third milking	F - test
Machine milk yield (kg)	$10.42 \pm 0.88$	$10.99 \pm 0.88$	0.4768
Stripping yield (kg)	$0.22 \pm 0.05$	$0.13 \pm 0.05$	0.2654
Residual milk (kg)	$3.94 \pm 0.82a$	$1.13 \pm 0.82b$	0.0218
Time of milking (min)	$5.57 \pm 0.48$	$5.61 \pm 0.48$	0.9541
Milk yield in 2 min	$4.22 \pm 0.53$	$4.87 \pm 0.53$	0.3988
Time to Peak flow (s)	$2.14 \pm 0.20$	$2.14 \pm 0.20$	0.9947
Peak flow (kg/min)	$2.95 \pm 0.33$	$3.28 \pm 0.33$	0.4859
Milk flow phases (s)			
increase	$1.19 \pm 0.16$	$1.24 \pm 0.16$	0.8485
decline	$2.12 \pm 0.27$	$2.09 \pm 0.27$	0.9347
SCC (logx/ml)	$4.72 \pm 0.25$	$4.91 \pm 0.27$	0.6171

a,b Averages in the same line with different letters are different at the level  $P < 0.05$ .

**Table 2: Effect of unknown human presence in front of head on day 150 on milking performance**

	Day 150	Day 152	F - test
Machine milk yield (kg)	$9.35 \pm 0.51$	$9.73 \pm 0.51$	0.6045
Stripping yield (kg)	$0.24 \pm 0.04$	$0.25 \pm 0.04$	0.8218
Residual milk (kg)	$0.58 \pm 0.30a$	$0.11 \pm 0.30b$	0.0294
Time of milking (min)	$5.07 \pm 0.46$	$5.61 \pm 0.46$	0.4186
Milk yield in 2 min	$4.68 \pm 0.40$	$4.75 \pm 0.40$	0.8985
Time to Peak flow (s)	$1.93 \pm 0.21$	$2.15 \pm 0.21$	0.4790
Peak flow (kg/min)	$2.92 \pm 0.22$	$2.90 \pm 0.22$	0.9696
Milk flow phases (s)			
increase	$1.25 \pm 0.12$	$1.46 \pm 0.12$	0.2374
decline	$2.15 \pm 0.28$	$2.26 \pm 0.28$	0.7880
SCC (logx/ml)	$4.88 \pm 0.14$	$4.88 \pm 0.14$	0.9996

a,b Averages in the same line with different letters are different at the level  $P < 0.05$ . Day 152 - control milking

Table 3. Milking performance during 1<sup>st</sup> and 3<sup>rd</sup> evening milking after calf removal and relocation cows to the herd

	Rearing condition												F - test
	Own mother			Nursing cows			Individual hutch			Control			
	first milking	third milking	third milking	first milking	third milking	third milking	first milking	third milking	third milking	first milking	third milking	third milking	
Machine milk yield (kg)	9.58 ± 1.39	10.11 ± 1.39	9.78 ± 1.34	11.27 ± 1.34	11.39 ± 1.33	12.03 ± 1.33	10.95 ± 1.20	10.58 ± 1.20	10.58 ± 1.20	10.95 ± 1.20	10.58 ± 1.20	10.58 ± 1.20	0.8558
Stripping yield (kg)	0.15 ± 0.13	0.13 ± 0.13	0.36 ± 0.11	0.26 ± 0.11	0.34 ± 0.10	0.06 ± 0.10	0.02 ± 0.09	0.07 ± 0.09	0.07 ± 0.09	0.02 ± 0.09	0.07 ± 0.09	0.07 ± 0.09	0.4039
Residual milk (kg)	5.05 ± 1.90	1.27 ± 1.90	5.22 ± 1.70	1.48 ± 1.70	3.02 ± 1.55	1.68 ± 1.55	2.47 ± 1.43	0.08 ± 1.43	0.08 ± 1.43	2.47 ± 1.43	0.08 ± 1.43	0.08 ± 1.43	0.8604
Time of milking (min)	5.07 ± 1.11	5.68 ± 1.11	5.74 ± 0.99	6.30 ± 0.99	6.05 ± 0.90	5.77 ± 0.90	5.42 ± 0.83	4.70 ± 0.83	4.70 ± 0.83	5.42 ± 0.83	4.70 ± 0.83	4.70 ± 0.83	0.8676
Milk yield in 2 min	3.95 ± 1.22	4.13 ± 1.22	3.34 ± 1.09	5.00 ± 1.09	4.63 ± 0.99	5.25 ± 0.99	4.98 ± 0.92	5.11 ± 0.92	5.11 ± 0.92	4.98 ± 0.92	5.11 ± 0.92	5.11 ± 0.92	0.8815
Time to Peak flow (s)	2.70 ± 0.46	2.77 ± 0.46	2.50 ± 0.41	1.94 ± 0.41	1.77 ± 0.37	1.65 ± 0.37	1.61 ± 0.34	2.22 ± 0.34	2.22 ± 0.34	1.61 ± 0.34	2.22 ± 0.34	2.22 ± 0.34	0.4785
Peak flow (kg/min)	2.70 ± 0.76	2.87 ± 0.761	2.54 ± 0.68	3.16 ± 0.68	3.45 ± 0.62	3.99 ± 0.62	3.11 ± 0.57	3.10 ± 0.57	3.10 ± 0.57	3.11 ± 0.57	3.10 ± 0.57	3.10 ± 0.57	0.9526
Milk flow phases (s)													
increase	1.33 ± 0.37	1.52 ± 0.37	1.42 ± 0.32	1.12 ± 0.32	0.83 ± 0.30	0.92 ± 0.30	1.21 ± 0.27	1.40 ± 0.27	1.40 ± 0.27	1.21 ± 0.27	1.40 ± 0.27	1.40 ± 0.27	0.8537
decline	1.28 ± 0.62	3.14 ± 0.47	1.36 ± 0.56	2.44 ± 0.56	3.14 ± 0.47	2.41 ± 0.50	3.14 ± 0.47	1.64 ± 0.47	1.64 ± 0.47	3.14 ± 0.47	1.64 ± 0.47	1.64 ± 0.47	0.0814
SCC (logx/ml)	5.29 ± 0.54	5.44 ± 0.62	4.46 ± 0.54	4.74 ± 0.54	4.58 ± 0.44	4.94 ± 0.44	4.57 ± 0.54	4.54 ± 0.54	4.54 ± 0.54	4.57 ± 0.54	4.54 ± 0.54	4.54 ± 0.54	0.9840

**Table 4. Effect of unknown human presence in front of head on day 150 on milking performance within rearing conditions**

	Rearing condition										F - test
	Own mother		Nursing cows		Individual hutch		Control				
	Day150	Day152	Day150	Day152	Day150	Day152	Day150	Day152	Day150	Day152	
Machine milk yield (kg)	10.77 ± 1.34	11.13 ± 1.34	9.75 ± 0.95	10.28 ± 0.95	7.23 ± 1.04	7.73 ± 1.04	9.66 ± 0.70	9.79 ± 0.70	0.8228		
Stripping yield (kg)	0.40 ± 0.10	0.36 ± 0.10	0.15 ± 0.07	0.25 ± 0.07	0.19 ± 0.08	0.16 ± 0.08	0.21 ± 0.05	0.22 ± 0.05	0.8107		
Residual milk (kg)	1.21 ± 0.54	0.31 ± 0.54	0.95 ± 0.42	0.11 ± 0.42	0.76 ± 0.47	0.09 ± 0.47	0.14 ± 0.36	0.09 ± 0.36	0.4998		
Time of milking (min)	5.86 ± 1.20	6.40 ± 1.20	5.36 ± 0.85	6.18 ± 0.85	4.26 ± 0.93	4.70 ± 0.93	4.81 ± 0.62	5.17 ± 0.62	0.9919		
Milk yield in 2 min	5.16 ± 1.06	5.00 ± 1.06	4.15 ± 0.75	4.11 ± 0.75	4.29 ± 0.82	4.61 ± 0.82	5.10 ± 0.55	5.29 ± 0.55	0.9924		
Time to Peak flow (s)	2.10 ± 0.54	2.03 ± 0.54	2.10 ± 0.38	2.51 ± 0.38	1.52 ± 0.42	2.02 ± 0.42	2.03 ± 0.28	2.03 ± 0.28	0.8554		
Peak flow (kg/min)	3.06 ± 0.57	2.93 ± 0.57	2.63 ± 0.40	2.58 ± 0.40	2.85 ± 0.44	2.91 ± 0.44	3.12 ± 0.30	3.20 ± 0.30	0.9946		
Milk flow phases (s)											
increase	1.13 ± 0.31	1.30 ± 0.31	1.35 ± 0.22	1.66 ± 0.22	1.20 ± 0.24	1.44 ± 0.24	1.33 ± 0.16	1.43 ± 0.16	0.9529		
decline	2.90 ± 0.74	3.26 ± 0.74	1.93 ± 0.52	2.36 ± 0.52	1.88 ± 0.57	1.62 ± 0.57	1.90 ± 0.39	1.80 ± 0.39	0.9022		
SCC (logx/ml)	4.91 ± 0.34	5.16 ± 0.34	4.57 ± 0.30	4.44 ± 0.30	4.69 ± 0.30	4.60 ± 0.30	5.36 ± 0.20	5.33 ± 0.20	0.9403		
Day 152 – control milking											

## DISCUSSION

During the first milking the machine milk yield was numerically lower than during the third one by all groups of heifers reared in different rearing systems except for the control group. This was supported by a significantly higher volume of residual milk after first milking in comparison with the third one indicating less effective milk removal. In general, transition to exclusive machine milking is associated with inhibition of oxytocin release and decreased milk production (Tančin et al., 2001; Marnet and Negrao, 2000). In our experiment higher residual milk volume can be caused by emotional stress induced by the calves weaning from cows. Also, their movement from individual maternity pen to a production group of cows could induce emotive stress. Milking of dairy cows under emotional stress from novelty and calf loss normally evokes a central inhibition of the oxytocin release (Tančin et al., 2001; Bruckmaier et al., 1993, Van Rennen et al., 2002). However, in our experiment it was beyond our scope to see whether higher residual milk volume during the first milking was caused by milk keeping for calf or due to the stress from new housing conditions. From our earlier results it appeared that higher residual milk volume during the first milking after calf removal is related to voluntary milk keeping for calf (Tančin et al., 2001). On the other side, the rearing conditions early after birth could contribute to the differences in residual milk volume among our experimental groups. Although not significantly, we could demonstrate numerically higher volume of residual milk in primiparous cows reared as calf by own mothers or nursing cows as compared to other two groups.

In the middle of lactation the presence of an unknown person in the front of cow's head before and during milking resulted in significant lower volume of residual milk as compared to milking in the absence of unknown human being. Though not significantly, the same tendency was seen in all groups of cows. Also, the cows reared by own mother or nursing cows showed higher volume of residual milk as in the beginning of lactation. When the aversive person was present, less milk and a higher residual volume was obtained during milking than during absence of human (Rushen et al., 1999). The same study also pointed out that higher residual volume of milk was found only in some cows that probably are able to distinguish between human approaches to them. The results from open field tests revealed that calves which were nursed by cows moved less in open field than their contemporaries from other groups (Brouček et al., 2000). The delayed occurrence of milk ejection in cows raised by nursing cows could indicate that such animals are exploring the new situation more than cows from other groups (Tančin et al., 2004).

The experiences with human during rearing

period due to ethological tests did not significantly affect the cow's response to human presence during milking. There were no significant differences between control group and a group of cows reared in individual hutches. However, numerically higher residual milk volume in primiparous cows from group of individual hutches could indicate some negative impact with human while performing tests as compared to control group.

Higher volume of residual milk under both stress situations in our experiment in primiparous cows reared by own mothers or nursing cows, their lower activities in open field test (Brouček et al., 2000), delayed milk ejection after relocation (Tančin et al., 2004) could indicate that such animals are more sensitive to stressful situations. In both experiments we demonstrated numerically higher residual milk volume under stress conditions in a group of cows reared by own or nursing cows compared to individual or control groups. Recently, it was described in rodents that exposure to neonatal separation stress increased exploratory behaviour compared to controls, which might indicate reduced anxiety (Becker et al., 2007). However, primiparous cows reared as calves by nursing cows had significantly higher milk production during the first lactation (Brouček et al., 2005, Uhrinčat' et al., 2007), which was confirmed in our experiment, too.

In conclusion, the rearing conditions of heifers during the period before weaning could influence their response to milking conditions as primiparous cows fall in the range that is not of high importance for milking performance. More investigations are needed to explain the effect of rearing conditions of calf heifers early after birth on their sensitivity to stressor during their whole life.

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