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Research Article

Effects on Health, Growth and Redirected Suckling Behaviour in Pair-Housed Dairy Calves in Outdoor Hutches

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- Cross-sucking.

Abstract

The aim was to investigate if pair-housing compared to single-housing of dairy calves in outdoor hutches during winter time had any negative effects on health, growth and redirected suckling behaviour. In total 21 calves of Swedish Red and Swedish Holstein cattle were housed in pairs (n=14 calves) or single (n=7) from 10 days to 6 weeks. Each calf was fed 3 l. of whole milk twice per day in teat-buckets. They had ad libitum access to concentrate, silage, hay and water and an empty, clean teat-bucket. Cases of diarrhoea and other diseases were recorded daily and the calves were weighed weekly. Behavioural observations were done on day 14 and then twice per week for each hutch until eight weeks of age. Behavioural observations were made by continuous recording ten minutes before milk feeding and 20 minutes after the calves received their milk in the morning and afternoon. Pair-housed calves had more diarrhoea than single-housed calves ($P<0.05$, Chi2-test). There were no coughing recorded nor any antibiotics prescribed to the calves, and no sucking induced injuries found on any of the calves. Weight gain did not differ between pair-housed and single-housed calves. There was no significant difference in total redirected suckling (non-nutritive sucking and cross-sucking) between treatments before receiving milk, but after milk delivery pair-housed calves performed more total redirected suckling than single-housed calves ($P<0.01$, Wilcoxon rank-sum test). Pair-housed calves performed more cross-sucking than single-housed calves both before and after receiving milk ($P<0.01$). Pair-housed calves had a higher frequency of drinking milk ($P<0.001$) while single-housed calves licked fixtures more often than pair-housed calves before receiving milk ($P<0.05$). In conclusion, diarrhoea and cross-sucking was higher in pair-housed calves', but there were no injuries from this and weight gain was the same as for single-housed calves. As social interactions are important for calf welfare one should still try to pair-house calves.

INTRODUCTION

According to EU-legislation calves above eight weeks must be group housed [1], and for calves in organic farming group housing is a must from one week of age [2]. During single-housing calves must be allowed to have visual and tactile contact with other calves [1]. In single-housing systems, the calves' opportunities for locomotion and social behaviours are restricted and are therefore not recommendable from an animal welfare point of view [3].

One reason for keeping pre-weaning dairy calves in single-housing is the concern that it could have a negative effect on calf health and performance [4,5]. Before weaning, calves are more susceptible to pathogens than older animals [6]. Diarrhoea and respiratory diseases are most common in calves during their first three months of life [7]. They are associated with lower growth rate and/or death and have an economic impact for the farmer in form of treatment costs, lower growth rates and replacement capacity of the herd [7]. Enteric and respiratory pathogens can be transmitted through contact between calves, depending on the management and calf housing system [7]. Minimizing animal-to-animal contact reduces transmissions of pathogens between the calves [8]. An increasing risk factor when keeping calves in outdoor hutches is the impact of the weather; a wet and cold calf in windy conditions can deplete its energy reserves rapidly. If the calf also is sick, it can easily succumb to the disease [8]. Rotavirus is the most common reason for diarrhoea in neonatal calves in Sweden and it is recommend using oral electrolyte solutions as treatments [9]. If a test concludes it to be bacteria causing the diarrhoea then antibiotics can be used [9]. However, the use of antibiotics can enhance the antibiotic resistance in bacteria [9]. Marcé et al. [7], argue that the risk of both enteric and respiratory diseases increases in group-housing of calves compared with single housing. However, Chua et al. [10], found no differences in occurrence of diarrhoea between single- and pair-housed calves. Oral and nasal contact between calves still occurred between the hutches, which allow faecal-oral transmissions of organisms [10]. According to Chua et al. [10], proper management of pens and hutches, such as cleaning, adequate ventilation and feeding, as well as calf immunity is of greater importance to disease susceptibility than housing system.

During the milk-feeding period, pair-housed calves had a higher intake of concentrate than the single-housed calves, which could be because of social facilitation [11]. Chua et al. [10], observed no difference in growth rate between group- and single-housed calves before five weeks of age and de Paula Vieira et al. [11], found no difference in growth rate before or during weaning. On the other hand, Terre et al. [12], found that group-housed calves had a lower growth rate than single-housed calves.

Redirected suckling behaviour is non-nutritive sucking on the surrounding or other calves and is a redirection of natural suckling behaviour. The ingestion of milk stimulates sucking and bunting at a teat-bucket, suggesting that the milk is a stimulus that induces redirected suckling behaviour [13]. Lidfors [14], suggested that redirected suckling behaviour could be a natural redirected response from the calf since calves suckling their mother change to a different teat when the milk flow decreases and finally finish [15]. Bunting is the response to a low milk flow and serves to stimulate the milk let down in the beginning of a meal and to empty the udder at the end [15]. Ingestion of a small amount of milk can trigger redirected suckling behaviour [16]. Redirected suckling behaviour generally starts directly after the milk is finished and decreases in a similar way to when calves suckle their dam [15]. A sucking bout from the cow udder takes 8-12 minutes [15], and ingesting three litres of milk from a teat-bucket only takes a few minutes [17]. The calves' motivation to suckle is then redirected towards either the empty teat (non-nutritive sucking), its surroundings or towards other calves ear, mouth, scrotum, prepuce, tail, udder area or navel (cross-sucking) [14].

A risk factor with pair-housed calves is the occurrence of cross-sucking [18], and a calf which cross-sucks before weaning is likely to continue after weaning [19]. Intersucking in dairy cows, i.e., when cows are sucking on the udder of heifers or other cows, can lead to udder damage, mastitis, milk loss and culling of breeding animals [19]. The behaviour stems from a redirection of the natural sucking behaviour [18]. Keil and Langhans et al. [19], concluded that cross-sucking was most frequent around milk feeding but it was observed nearly all hours of the day. Also single-housed calves have be seen cross-sucking on the neighbouring calves' ears and mouth [10]. Cross-sucking may be reduced by using an artificial nipple or teat-bucket at milk feeding rather than a regular bucket, or by providing a non-nutritive artificial teat following milk feeding [20,21]. The amount of milk fed has an impact on performance of cross-sucking: when calves received either five litres or only 0.1 litres there was almost no cross-sucking compared to when the calves received one or 2.5 litres during weaning [22]. Jensen et al. [18], summarizes that the best way to prevent cross-sucking is to offer the opportunity for the calves to fulfil their need to suck in connection with the ingestion of milk, preferably by teat-buckets which are left for a period of time after feeding.

A health risk with cross-sucking is frostbites occurring in cold weather because of the wet area on the calf that is left after being sucked on [23]. If an ear is sucked on it can for example lead to necrosis arising and part of the ear falling off [23]. Single-housing can prevent most cross-sucking but not the underlying motivation to perform the behaviour. Previous research have shown that welfare is impaired when animals cannot perform strongly motivated behaviours [24].

The aim of this study was to investigate if dairy calves kept in outdoor hutches during the cold season had a higher growth rate, similar health, but higher frequency of redirected suckling behaviour when kept in pairs than individually. It was predicted that pair-housed calves would have an increased feed intake due to social facilitation and keep each other warm when lying together in the hutch, leading to a higher growth rate compared to single-housed calves. It was further predicted that pair-housed calves would be subjected to a higher pathogen load during the first weeks, but that should disappear when the calves' immune system adapted to each other and no difference between pair-housed and single-housed calves should be seen after the first weeks together. Cross-sucking on another calf was predicted to increase when kept in pairs, but instead the occurrence of non-nutritive sucking was predicted to be decreased compared to single-housed calves. Still, the amount of sucking related injuries on the calves was not predicted to increase in pair-housed compared to single-housed calves as they had continuous access to teat-buckets.

MATERIALS AND METHODS

Data collection was conducted at Swedish Livestock Research Centre at Lövsta in Uppsala between November 2015 and March 2016. The experimental procedure, including the animal handling, was within the boundaries of Lövsta's ethical approval (dnr: C332/12).

Materials

A total of 21 healthy heifer calves of Swedish Red (SR) and Swedish Holstein (SH) breed were included (single-housing n=7 calves (2 SR, 5 SH), pair-housing n=7 pairs (8 SR, 6 SH calves).

The calves were separated from their dams directly after calving and fed colostrum with a bottle (SHOOF Easy Feeder 2.5 litres) within four h after calving. The amount suckled as the first meal was at least 2.5 L. If the calves did not want to drink the first meal, it was given through tube feeding (BOVIVET Calf Drencher 2 litres with rigid probe). All calves were marked with ear identity tags with individual numbers before moved out to a calf hutch according to standard farm practise. Calves were fed 3 L of colostrum in a teat-bucket twice a day during their first three days (a total of six meals). From day four until day 56, the calves were fed whole milk. Three litres of whole milk were fed in teat-buckets twice per day at around 7:00 and 18:30 h. Silage, hay, pelleted concentrate (IDOL, Lantmännen) and water were available *ad libitum*. Water was available in open buckets and concentrate was given inside the calf hutch in a container (0.1 m^2). The calves in the study were managed by the personnel of the farm according to regular routines on the farm, except during milk feeding. Regular routines included scraping the outdoor pen

once a day, replenish silage, hay and concentrate, giving fresh water and checking if needing to replenish with fresh straw inside the calf hutch. Calves were dehorned before 30 days of age and before the procedure they were sedated and given pain-relief by injection.

After the calves were fed their first meal of colostrum indoors, they were immediately moved to outdoor calf hutches (Calf-tel PRO 2) where they were housed individually until selected into single or pair-housing. The calf hutches measured 2.12 x 1.14 x 1.22 (~2.4 m^2) on the inside with an outside pen attached measuring 1.36 x 1.26 m [~1.7 m^2, Figure 1]. All calves were housed in the same area and all calves were able to hear and see other calves and also engage in muzzle contact with the calves in adjacent calf hutches. This allowed single calves to perform cross-sucking on the calves muzzle in the adjacent calf hutches and also to perform social grooming. The calf hutch area consisted of a concrete floor measuring 36 x 14 m with a metal roof and open sides. The calf hutches were placed under the roof in two rows of 18 (n=36) with an alley separating the rows. Straw was used as bedding material.

Methods

Calves were single-housed during the period that they received colostrum (six meals, three days). After meal six the calves could be sorted into either single or pair-housing depending on if there was another suitable calf available. Pairing was done when one of the calves was ten days old; the other calf was then from four to ten days of age. In pairing calves, the calves' weight and age was taken into account; if it differed more than 10 kg or more than one week of age between the calves they were not paired. If sorted into single-housing the calf stayed in the same calf hutch while if sorted into pairs the two calves would be moved to a new calf hutch.

The calf hutches were marked with letters in the order that they were included into the study (A, B, C, etc.). The calf hutch was equipped with an empty teat-bucket during all times except during milk feeding [Figure 1]. At milk feeding the personnel was not allowed to be at the calf hutch platform 20 minutes before feeding. The calves in this study were to receive milk first and afterwards the other calves at the platform could be fed. The personnel had to make sure that the calves all started to drink the milk before moving on to another hutch. After the last calf in this study had received milk the personnel was not allowed to interfere with the calves for 20 minutes and had to leave the platform if done feeding the other calves before the 20 minutes had passed. After the 20 minutes and within one hour the milk buckets were taken down and the empty teat-bucket were put back up again. The teat-buckets used for milk was taken apart and washed with water and soap. The teats were placed in Virkon S 1% (DuPont Animal Health) for 5-10 minutes and the buckets and the teats were then assembled and hung up for drying. The teats were controlled regularly to ensure teat quality.

The weaning-period started at six weeks of age with reduction from three litres of milk per feeding to two litres and to one litre at seven weeks of age and the calves were considered weaned at eight weeks of age. Management of calves kept in either single or pair-housing during the study is described in Table 1.

Table 1 Management of calves kept in either single or pair-housing during the study

Age	Treatment
Birth	Received colostrum within 4 hours of birth Weighed Moved out to single-hutch after having received colostrum
1-3 days	Received colostrum during first 6 meals
4 days	Weighed
4-10 days	Paired with suitable calf of 10 days of age
7 days	Weighed
10 days	Weighed and if no suitable calf had been found calf was selected to single-housing
14 days	Based on oldest calf first behavioural observation. Weighed
3-8 weeks	Weighed once a week and behavioural observation twice a week
6 weeks	Weaning started and daily milk amount decreased from 6 to 4 litres
7 weeks	Daily milk amount decreased from 4 to 2 litres
8 weeks	Calves considered weaned and were weighed. Last behavioural observation

Figure 1 Calf-hutches used in the study and a hutch with pair-housed calves (Photo: Alvegard, 2016).

Health

All cases of health disturbances, such as diarrhoea and coughing, and any medical treatment given were recorded daily for each calf by the calf personnel in a health protocol. The occurrence of diarrhoea was divided into four categories [25,26], according to the following diarrhoea scores:

"dry tail…" in under 0. Firm consistency. Brown colour. Clean and dry tail and perineum.

1-Faeces with a paste-like consistency without shape.

2- Watery consistency (flowing out).

3- Watery consistency (flowing out) with blood.

If the calves were diagnosed as ill they were treated according to the current veterinary practice adapted on the farm. If diagnosed with diarrhoea (score 1-3) by the calf personnel, the calf hutch was marked and the calf/calves were given Diakur (Protect Diakur Super, Lantmännen) in their milk. If considered necessary Effydral (Zoetis Effydral Animal Electrolyte Tablets) was also given in their water. Diarrhoea scored for each week was seen as one case of diarrhoea and diarrhoea stretching over several weeks was considered the same case.

Growth and temperatures

Live weight was measured at birth, after the last meal of colostrum, at day 10 and every week after morning milk feeding. Weighing was done with a scale that was brought to the hutches and the calves were moved into the scale at each weighing (Maréchalle Pesage Weighing Crate PM 120). Each calf was examined by the personal as an extra health control in conjunction with the weekly weighing.

Daily air temperature (°C) and relative humidity (RH %) was recorded in four hour intervals 24 hours a day during the study period (HOBO U12-013 data logger). The average daily mean outdoor temperature from November to March was 0.7±0.3 °C. January had the lowest average temperature and also the lowest daily temperature with -21.6 °C.

Behavioural observations

Behavioural observations were done on each hutch twice a week between 2-8 weeks of age, where week was based on the age of the oldest calf in the hutch. Each calf hutch was observed in conjunction with milk feeding at 7:00 and 18:30 h. The observer took place in front of the calf hutch 20 minutes before feeding (6:40 or 18:10 h), giving 10 minutes for the calves to habituate to the observer and then started observing 10 minutes before feeding. After the calves had received milk, the observation continued for 20 minutes. During each observation both calves in the calf hutch, if kept in pairs, were observed. Continuous recording of frequencies in one minute intervals was done. The recorded behaviours and their definitions are presented in Table 2. If other behaviours were performed they were noted under "Other activity".

Table 2 Ethogram adapted from Jensen and Budde (2006)

Behaviour	Definition
Ingesting milk	The calf is ingesting milk by sucking a teat.
Sucking empty bucket or teat	The calf is sucking on an empty teat or an empty bucket, but no milk is ingested. Sucking movements are performed with part of bucket or teat in the mouth
Cross-sucking head or neck	The calf is sucking on the head (muzzle, ear or skin) or on the skin of the neck of another calf. The sucking movements are performed with the body part in the mouth
Cross-sucking under belly	The calf is sucking under the belly of another calf, mainly on navel, scrotum or udder base. The sucking movements are performed with the body part in the mouth
Bunting calf	The calf is pushing its forehead with a rapid and forceful movement against another calf's head, neck or body
Bunting bucket or teat	The calf is pushing its forehead with a rapid and forceful movement against a bucket or a teat
Licking fixtures	The calf's tongue is out of its mouth and in contact with any fixtures of the pen, except teat or bucket
Social grooming	The calf's tongue is out of its mouth and in contact with the head, neck or body of another calf
Self-grooming	The calf's tongue is out of its mouth and in contact with its own body
Inactive	The calf is standing or lying still
Other activity	The calf is performing any activity not described above

Statistical analysis

Data analysis was done in R (version 3.3.1 2016-06-21), using packages lme4 (version 1.1-12) for mixed models and LmerTest (version 1.0) to obtain P values on all data except for diarrhoea. Hutch values (7 single-housed calves and 7 pairs of pair-housed calves) were used to generate means for behaviour, cough, fever, diarrhoea, and weights for statistical analysis. Week 7-8 was excluded from the statistical analysis since not enough calves were included in the study for that period. Week 1-6 was included when analysing weight and diarrhoea. For diarrhoea, age in weeks was compiled into period 1 (week 1-3) and period 2 (week 4-6) and significance of treatment on cases of diarrhoea during each period was tested with a Chi^2-test.

Differences in weight as a response to treatment over time were tested by the Residual Maximum Likelihood (REML) analysis, with a treatment x day interaction, temperature and breed as fixed effects, hutch identity as a random effect and birth weight as a covariate. An analysis of variance (ANOVA) was done to assess the effects of treatment on weight gain between week 2 and 6, with treatment as a fixed effect and birth weight as a covariate.

Behaviour data analysed included week 2-6; week one was excluded when analysing the behaviours since the first behavioural observations were done at two weeks of age and all calves included were then in their hutch. The average temperature of the day was categorized in three categories: above +5 °C (1), between +5 °C and -5 °C (2), and under -5 °C (3).

Behaviours were checked for normality of the residuals (Shapiro-Wilk test) and homogeneity of variance with Bartlett's test and all variables were square root transformed to provide normal residuals. Redirected suckling behaviours performed

before receiving milk and sucking on empty bucket after receiving milk had normal residuals but were square root transformed to provide a better pass of the Shapiro-Wilk test. The result was not back-transformed. If data was not normally distributed after data transformation (before milk: sucking on empty bucket or teat, bunting on bucket or teat, cross-sucking or social grooming; after milk: drinking milk, redirected suckling total, cross-sucking and social grooming) a non-parametric Wilcoxon rank-sum test with continuity correction was performed where the sum of the different behaviours performed before and after receiving milk was analysed separately.

For behaviour, the weekly means are the average of the two observations per calf hutch per week. The mean value of each behaviour performed per observation before and after milk feeding was used and analysed separately. Behaviours bunting calf, cross-sucking under belly, cross-sucking on head or neck and licking fixtures was compiled to categorise all redirected suckling behaviours (redirected suckling total). Bunting calf, cross-sucking under belly and cross-sucking on head or neck was compiled to categorise all behaviours redirected towards another calf (cross-sucking) excluding licking fixtures. Differences in behaviour as a response to treatment over time were tested by REML analysis, with a treatment x day interaction, temperature, and breed as fixed effects and hutch identity as a random effect.

Data are presented as weekly means ± standard error (SE) of the weekly mean. Significant level was set to 0.05.

RESULTS

Health

Pair-housed calves had significantly higher number of hutches with diarrhoea (score 1-3) in both period 1 (weeks 1-3, pair: 5 hutches, single: 1 hutch, p=0.03, Chi^2=4.7) and period 2 (weeks 4-6, pair: 4 hutches, single: 0 hutches, p=0.02, Chi^2=5.6). All pair-hutches had at least one case of diarrhoea; two hutches had two cases each, whereas only one single-hutch had diarrhoea. Diarrhoea was recorded 42 times and the longest period for a calf to continuously have diarrhoea was eight days and the shortest one day. The occurrence of diarrhoea was most common at three weeks of age for pair-housed (8 calves) and least common in week six (0 calves).

No coughing or fever was recorded and no antibiotics had to be prescribed to the calves during the study. No sucking related injuries were found.

Growth

Birth weights were similar for the two treatments (pair-housed: 39.9±15.1 kg, single-housed: 40.6±15.4 kg, p>0.1). No significant effect of treatment on weight between pair- and single-housed calves was found [Figure 2a] but there were significant main effects of week ($F_{(5,58)}$=58, P<0.001) and birth weight ($F_{(1,11)}$=75.1, P<0.001). Weight gain from birth to week six (pair-housed: 5.6±2.1, single-housed: 5.6±2.1 kg/week) or from week two to week six (pair-housed: 5.1±1.9, single-housed: 5.0±1.9 kg/week) were not affected by treatment. In pair-hutches, one calf often had slightly higher weight gain than the other calf in the hutch [Figure 2b].

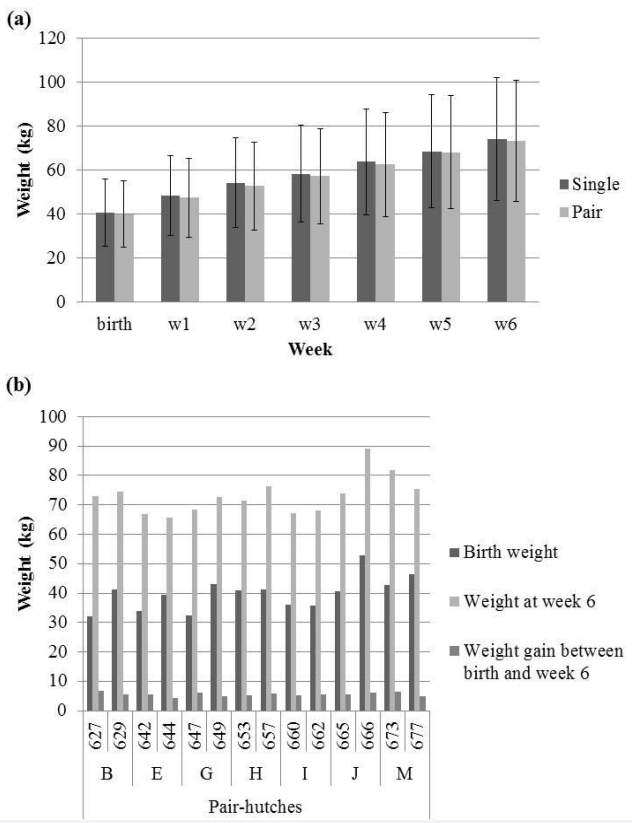

Figure 2 Mean (± SE) of weights (kg) in calves from birth to 6 weeks of age when housed single or in pairs in hutches (a) and weights of individual calves when housed in pairs (b)

Behaviours

There was no significant effect of housing treatment on total redirected suckling before receiving milk. After receiving milk, however, pair-housed calves performed total redirected suckling more often than single-housed calves [Table 3, Figure 3] There was a significant effect of treatment on cross-sucking both before (P<0.01) and after receiving milk (P<0.001), where single-housed calves did not perform any cross-sucking [Table 3, Figure 3].

Single housed calves had a higher frequency of licking fixtures than pair-housed calves before receiving milk (P>0.05), but there was no significant effect of treatment after receiving milk [Table 3] No significant effect of treatment on bunting bucket or teat before receiving milk was found. However, there was an effect of treatment (P<0.001), day (P<0.05) and temperature ($F_{(2,108)}$=5.2, P<0.01) on bunting bucket or teat after receiving milk where single-housed calves performed the behaviours more frequently [Table 3]. Pair-housed calves had a higher frequency of drinking milk than single-housed calves [P<0.001, Table 3] but there was no significant effect of treatment on sucking on empty bucket or teat neither before nor after receiving milk [Table

Table 3 Mean values (±SE) of number of recordings week 2-6 of behaviours before and after receiving milk for single vs. pair housed dairy calves kept in outdoor calf hutches and results from statistical testing on the effects of housing treatment, observation day and treatment-day interaction for normally distributed behaviours tested with a REML model and for not normally distributed data tested with Wilcoxon rank-sum test. Significant results are marked with bold and – means that it was not tested due to the behaviour was not performed or it being tested with Wilcoxon rank-sum test.

Behaviour	Before receiving milk					After receiving milk				
	Single mean	Pairs mean	Effect of treatment	Treatment and day interaction	Main effect of day	Single mean	Pairs mean	Effect of treatment	Treatment and day interaction	Main effect of day
Redirected suckling total	2.86±1.08	1.83±0.69	$F_{(1,11)}$=1.6	$F_{(9,101)}$=1.5	$F_{(9,101)}$=0.89	7.14±2.70	16.03±6.06	**W=6**	-	-
			P=0.23	P=0.16	P=0.23			**P=0.02**	-	-
Licking fixtures	2.86±1.08	1.37±0.52	**$F_{(1,12)}$=5.1**	$F_{(1,102)}$=1.4	$F_{(9,102)}$=1.5	7.14±2.70	3.08±1.16	$F_{(1,10)}$=1.74	$F_{(9,100)}$=0.48	$F_{(9,101)}$=1.25
			P=0.04	P=0.19	P=0.17			P=0.22	P=0.89	0.28
Bunting bucket or teat	0.38±0.14	0.06±0.02	W=33.5	-	-	24.41±9.22	8.64±3.26	**$F_{(1,10)}$=11.5**	$F_{(9,101)}$=0.56	**$F_{(9,101)}$=2.1**
			P=0.23	-	-			**P=0.006**	P=0.83	**P=0.05**
Cross-sucking	0.±0.	0.46±0.17	**W=3.5**	-	-	0.±0.	12.95±4.89	**W=0**	-	-
			P=0.004	-	-			**P=0.001**	-	-
Drinking milk	-	-	-	-	-	3.91±1.48	6.51±2.46	**W=0**	-	-
			-	-	-			**P=0.0006**	-	-
Sucking on empty bucket	2.28±0.86	1.79±0.67	W=20	-	-	30.22±11.42	30.49±11.53	$F(_{1,10})$=0.13	$F(_{9,100})$=0.48	$F_{(9,100)}$=0.58
			P=0.62	-	-			P=0.73	P=0.89	P=0.81
Self-grooming	1.97±0.74	1.86±0.70	$F_{(1,110)}$=0.06	**$F_{(9,110)}$=4.4**	**$F_{(9,110)}$=2.5**	4.17±1.58	3.16±1.19	$F_{(1,11)}$=0.94	**$F_{(9,101)}$=2.1**	$F_{(9,101)}$=0.86
			P=0.81	**P=0.00006**	**P=0.014**			P=0.36	**P=0.03**	P=0.54
Social grooming	0.±0.	0.27±0.10	**W=3.5**	-	-	0.±0.	0.26±0.10	**W=3.5**	-	-
			P=0.004	-	-			**P=0.004**	-	-

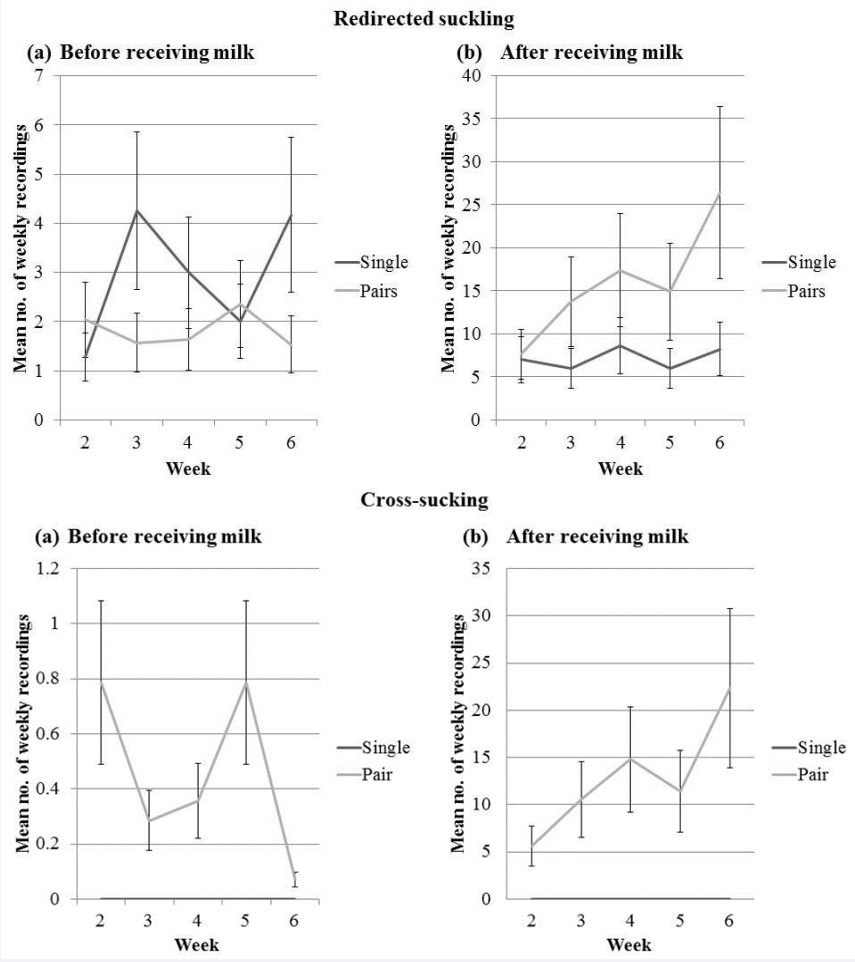

Figure 3 Mean (+SE) number of weekly recordings of redirected suckling behaviours and cross-sucking before (a) and after (b) receiving milk in dairy calves housed in single- or pair-housing from 2-6 weeks of age.

3]. Self-grooming was significantly affected by a treatment-day interaction both before (P<0.001) and after (P<0.05) receiving milk [Table 3]. There was also a main effect of day on self-grooming before receiving milk [P<0.05, Table 3]. Social grooming was significantly affected by treatment where pair-housed calves performed social grooming both before (P<0.01) and after (P<0.01) receiving milk whereas single-housed calves never performed social grooming [Table 3]. There was no effect of breed on any of the tested behaviours.

There was a difference between the calves in the pair-hutches where one calf generally cross-sucked more than the other [Figure 4a]. Cross-sucking was mostly performed on the other calves head or neck both before and after receiving milk (52.5% total or 2.1±1 and 9.3±3 respectively) or as bunting at the other calf (46.5% or 1.0±0 and 8.1±3 respectively). Sucking under belly was performed least of the three behaviours (1.0% or 1.0±0 and 1.8±1 respectively) [Figure 4b].

DISCUSSION

Pair-housed calves had more diarrhoea than single-housed calves during the whole study. It was predicted that diarrhoea should increase during the first week in the hutch (week two) as a result of a higher pathogen load, but that this effect should decrease when the calves' immune system adapted. No such adaption can be seen in the results as the occurrence of diarrhoea is similar during both periods. The higher occurrence can be due to that if one calf in the pair-hutches got diarrhoea the other calf also got it due to sharing of space and that they suckled on the same teats.

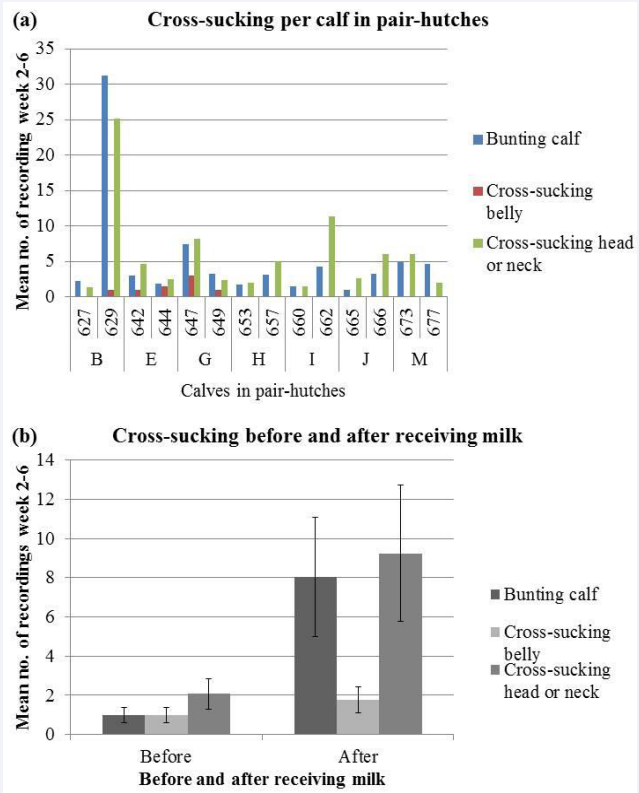

Figure 4 Mean number of recordings of bunting, cross-sucking belly or cross-sucking head or neck of another calf per individual calf (a) and means (± SE) before and after receiving milk in pair-housed calves in hutches.

Diarrhoea has been found to have a negative impact on growth [7]. However, there was no difference in weight or weight gain between single- and pair-housed calves in this study. This is consistent with the result of de Paula Vieira et al. [11], who saw no difference in body weight during the pre-weaning and weaning period between pair-housed and single-housed calves. Chua et al. [10], found that pair-housed calves' continued to gain weight at pre-weaning levels during the weaning period at six weeks of age whereas single-housed calves only gained at half the rate of the pair-housed. Since not enough data was obtained for week 7-8 in this study the effect of weaning on weight was not analysed. Terre et al. [12], on the other hand found that group-housed calves (five calves/pen) had a lower growth rate than single-housed calves. The results in this study thus contradicts the predictions that pair-housed calves should have a higher growth rate than single-housed calves due to social facilitation.

Cold stress has a larger impact on young, sick and injured animals than it has on mature and healthy animals [27]. Perhaps the pair-housed calves had a higher energy reserve due to less energy being used to maintain body temperature, which could be used to maintain weight gain even during a case of diarrhoea. As no significant difference could be found in weight or weight gain between the two treatments, pair-housing could be seen as a viable option during the cold season.

The pair-housed calves could steal milk from each other, which may explain that the pair did not gain weight equally. It also suggests that perhaps the volume of milk was too restrictive. Calves aged 2-6 weeks can drink from 10-16 l. of milk/day when given free access [28,29]. If calves had been given a larger amount of milk than 6 l./day in this study, then their growth may have been better. Furthermore, the concentrate and silage was provided so that only one calf at a time could feed, but hay was available so that both calves could eat together.

In this study, pair-housing did not increase the risk of sucking related injuries, even as the temperature dropped to -21°C, which is opposite to earlier suggestions [23]. If this is due to the teat-bucket that was continuously available, a warmer temperature inside the hutch or due to other factors cannot be concluded.

No significant difference in total redirected sucking was found before the calves received milk, although there was a difference in distribution of which behaviours they performed. After receiving milk, pair-housed calves performed more redirected suckling in total than single-housed calves. Cross-sucking was performed more by pair-housed calves both before and after receiving milk. Redirected suckling is stimulated by the taste of milk [13] and most redirected suckling occurs during the first 10-15 minutes after milk feeding [14]. But this does not answer the question why single-housed calves displayed a higher frequency of licking fixtures before receiving milk than pair-housed calves. It may be due to that pair-housed calves directed their non-

nutritive sucking towards the other calf (cross-sucking) instead of licking the fixtures. The finding that no significant difference in licking fixtures was found after receiving milk can be due to that single-housed calves were able to satisfy their need for sucking on the teat-bucket while pair-housed calves did not because of competition about the teats and therefore cross-sucked instead.

Cross-sucking could be performed by both treatments but was only recorded in pair-housed calves. Single-housed calves did not perform any cross-sucking at all even though they could cross-suck on the neighbouring calves' muzzle. This could be due to the access to the teat-bucket where the single-housed calf did not have to compete for the teat or milk with another calf. Competition for milk depends on milk allowance where a low milk allowance increases the occurrence of cross-sucking [21]. Competition for the teats increases when access to the teat is reduced, but competition for the teats is seen even when the calves have one teat each [30]. Same could be seen in this study, where the calves competed for the teats. The increase of cross-sucking seen by Lidfors et al. [14], is consistent with the findings in this study where an increase of cross-sucking can be seen in week six at the start of the weaning period. Previous studies have suggested that the occurrence of cross-sucking in calves can lead to welfare problems later on [19]. However, this study ended at 8 weeks of age and we do not have any data on the long-term effects.

There were significant effects of treatment, day and temperature on bunting bucket or teat after receiving milk, where single-housed calves performed the behaviour to a larger extent than pair-housed calves. Bunting has been seen to increase over time as the calf's effort to maintain milk flow from the udder and to empty the teat and udder more completely increases as a result of the growing demand for milk with age [13]. Pair-housed calves might perform less bunting on bucket or teat because of the competition for milk between the calves. The calf that did not get access to the milk bucket could have performed bunting towards the calf instead of the other empty bucket or teat.

At week six, the weaning period started with a decrease from three litres of milk per feeding to two litres of milk per feeding. The increase of total redirected suckling and cross-sucking that can be seen in week six in the figures is therefore not unexpected. A slow milk flow (0.5 litre/minute) decreases the frequency of non-nutritive sucking in comparison to a fast milk flow (1 litre/minute) [17]. As a high amount of milk (5 litres per meal) also has been seen to reduce redirected suckling [22], a combination of a high amount of milk with a reduced milk flow should have a positive effect on reducing the redirected suckling following milk feeding as observed in this study.

Factors that affect the time for milk intake from a teat-bucket is, among others, the age of the calf and the size of the hole in the teat [17]. The frequency of cross-sucking decrease with time and cease within 15 minutes after milk feeding [14]. If the empty teat-bucket is removed after milk intake, it will increase the occurrence of cross-sucking [22]. Feeding by teat-bucket also allows calves to express their natural sucking reflex during feeding and the calves can keep on sucking the empty teat-bucket after the milk is ingested [22]. According to Loberg and Lidfors [17], it seems that the motivation for sucking is reduced with the possibility to perform the behaviour and a longer time spent ingesting milk. According to Rushen and de Passillé [16], the taste of milk stimulates non-nutritive sucking more than access to a teat, and the motivation for sucking is reduced more by performing the non-nutritive sucking behaviour itself than by ingesting milk.

There were individual differences between the pair-housed calves in the frequency of cross-sucking. Most of the recorded behaviours were performed by one calf in each hutch, but all calves performed cross-sucking at least two times. This is in line with results obtained by Loberg and Lidfors et al. [17].

The calves directed 52.5% of the cross-sucking towards the other calves' head or neck, 1.0% towards the belly and 46.5% on bunting the other calf. Jung and Lidfors et al. [22], found that calves directed most of their cross-sucking towards the other calves' belly (52.7%) and less at the calves´ mouth (16.3%) and ears (1.3%). In contrast, Lidfors [14], reported that ~40% of the cross-sucking was directed towards other calves' mouth, ~34% towards their ears and ~3% towards their throat. Cross-sucking towards the head and around the muzzle can be due to it being smeared with milk [21], but in this study also as a response to the other calf stealing the teat. The low frequency of cross-sucking under the belly is extra desirable as excessive cross-sucking under the belly can lead to inter-sucking in heifers and milk stealing in cows [19].

Pair-housed calves had a significantly higher frequency of drinking milk than single-housed calves. Not surprising when one calf had finished its milk it often tried to steal the milk from the other calf, giving a higher recorded frequency of drinking milk when the calves competed over the bucket that still contained milk. In future studies it would be recommended to add a column with the behaviour "stealing milk" to get a more correct result on which behaviours that were performed. It may also be useful to repeat the study with larger quantities of milk and a reduced milk-flow, as we expect that this could minimize cross-sucking behaviours.

CONCLUSIONS AND RECOMMENDATIONS

In conclusion, there was a higher occurrence of diarrhoea for calves in pair-hutches than single-housed calves. However, despite the higher occurrence of diarrhoea, we did not observe any effect of housing type on weight gain. No other effect on health was seen and no injuries due to redirected suckling were recorded during the study, even though pair-housed calves performed all the recorded cross-sucking behaviours and redirected suckling at a higher frequency than the single-housed calves. As both diarrhoea and cross-sucking are considered welfare issues, pair-housing of calves under the conditions that were implemented in this study cannot be recommended as a general on-farm routine even though weight gain can be

maintained. Given the importance of social behaviours for calf welfare and the strong motivation to suckle, further studies on how to reduce the occurrence of diarrhoea and cross-sucking in pair-housed calves during on-farm conditions is needed.

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