



# **Studies on Restricted Suckling in Dual Purpose and Dairy Breed Cattle in Mexico**

Sofie Fröberg

LICENTIATE THESIS

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**Institutionen för husdjurens  
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**Swedish University of Agricultural Sciences  
Department of Animal Nutrition and Management**

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## Abstract

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The aim of this thesis was to investigate the effects of rearing calves by restricted suckling (RS) compared to artificial rearing (AR) in dual purpose and dairy breed cattle in Mexico, milked once or three times a day, respectively. The following parameters were recorded during the first eight weeks after calving: social, abnormal and general behaviours and weight gain of Zebu crossbred and dairy calves; and milk yield, milk composition and udder health in the Zebu crossbred dams and udder health and milk let-down in the Holstein dams.

RS reduced the abnormal behaviours of the calves and resulted in less foraging behaviours compared to AR. AR dairy calves consumed four-fold as much concentrate as RS dairy calves, whereas the amount of milk ingested was similar in the two treatments. A higher fat content in the milk ingested by RS calves compared to whole milk fed to AR calves, resulted in similar total ME intake from milk and concentrate in both treatments. Irrespective of type of animals RS resulted in similar weight gain in comparison to AR. Cow-calf separation five days after calving induced fewer indications of physiological stress in RS cows and calves. RS further increased the saleable and total (saleable and suckled) milk yield and decreased the fat content in saleable milk. RS improved udder health of the Zebu crossbred cows as judged according to elevated CMT scores and lower lactose content in AR cows. RS of Holstein dairy cows did not impair milk let-down and udder health tended to be improved according to the CMT. RS calves showed a front teat preference, which did not impair udder health according to similar CMT scores of front and rear teats.

These studies indicate that RS is economically viable for the farmer as once daily milking combined with twice daily suckling considerably increased the saleable and total milk yield of Zebu crossbred cows and improved animal well-being.

*Keywords:* artificial rearing, behaviour, calf weight gain, Holstein cattle, milk production, Zebu crossbred cattle, udder health.

*Author's address:* Sofie Fröberg, Department of Animal Nutrition and Management, SLU, Kungsängen Research Centre, SE-753 23 Uppsala, Sweden.  
sofie.froberg@huv.slu.se

## **Licentiate degree**

The licentiate degree which requires two years of full-time postgraduate studies, is intended to guarantee, by means of course work and the completion of a dissertation, that the recipient

- has demonstrated an ability to investigate and to solve problems scientifically;
- is conversant with general scientific methodology and is familiar with the more important research methods within his or her subject area;
- is knowledgeable within his or her area of expertise and has contributed to the development of this area through his or her own research;
- is able to utilise the scientific literature within the subject area and relate it to his or her result;
- has in the planning and execution of research, as well as in the analysis of results, worked both independently and in the co-operation with others;
- has experience in presenting and discussing research results, both orally and in writing, e.g., before a board of examiners at a final public seminar.

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# Appendix

## Paper I-III

The present thesis is based on the following papers, which will be referred by their Roman numerals:

- I. Fröberg, S., Aspegren-Güldorff, A., Olsson, I., Marin, B., Berg, C., Hernández, C., Galina, C.S., Lidfors, L. & Svennersten-Sjaunja, K. 2005. Effect of restricted suckling on milk yield, milk composition and udder health in cows and behaviour and weight gain in calves, in dual purpose cattle in the tropics. (Manuscript intended for publication in Preventive Veterinary Medicine)
- II. Hernández, C., Orihuela, A., Fröberg, S., & Lidfors, L.M. 2005. Effect of restricted suckling on physiological and behavioural stress parameters in dual purpose cattle in the tropics. (Accepted for publication in Livestock Production Science)
- III. Fröberg, S., Gratte, E., Svennersten-Sjaunja, K., Olsson, I., Berg, C., Orihuela, A., Galina, C.S., García, B. & Lidfors, L. 2005. Effect of restricted suckling on udder health and milk let-down in Holstein cows and weight gain, feed intake and behaviour of their calves. (Manuscript intended for publication in Applied Animal Behaviour Science)

Paper II is reproduced by kind permission of the journal concerned.

## Abbreviations used in this thesis

|     |                          |
|-----|--------------------------|
| AR  | Artificial rearing       |
| CMT | California mastitis test |
| DM  | Dry matter               |
| ECM | Energy corrected milk    |
| LWG | Live weight gain         |
| ME  | Metabolisable energy     |
| RS  | Restricted suckling      |
| SCC | Somatic cell count       |

## **Introduction**

This thesis examines the effects of restricted suckling (RS) in comparison with artificial rearing (AR) applied to cows and calves of two types of cattle: dual purpose (Holstein × Zebu) and dairy cattle (Holstein). In the Zebu crossbred cattle behaviour and weight gain in the calves and udder health, milk production and milk composition in the cows were investigated. In Holstein dairy cattle behaviour, feed consumption and weight gain in the calves and udder health and milk let-down in the cows were evaluated.

### ***Bos taurus* and *Bos indicus* cattle**

Domestication of cattle started about 12 000 years ago (Loftus *et al.*, 1994b). There is evidence that domestication of cattle took place independently in several locations that resulted in two major types of domestic cattle (*Bos taurus* and *Bos indicus*) named as separate species, but often considered as subspecies (Machugh *et al.*, 1997) due to complete interfertility (Loftus *et al.*, 1994a). *Bos taurus* is also called European cattle, while *Bos indicus* is also known as Zebu cattle.

The phenotypic difference between the two types of cattle is that most of the *Bos indicus* breeds have a hump, excess skin under the throat, short hair and large ears. One effect of these attributes is increased body surface for regulation of body temperature. Hence Zebu cattle are more heat-tolerant and this together with their natural resistance to tropical disease enables them to cope with harsh conditions such tropical climates better than European cattle (Phillips, 2001). However, the potential to produce milk or beef under more controlled management conditions is in most *Bos indicus* breeds lower than in *Bos taurus* cattle. Therefore, *Bos taurus* cattle are often kept where climate and feeding conditions allow this, whereas Zebu cattle are the best option in a tropical climate. For many years it has been a common practise to cross the local Zebu breeds with European cattle, predominantly Brown Swiss and Holstein to improve milk production under tropical conditions. According to Preston & Vaccaro (1989) these crosses are generally used in dual purpose cattle production systems where income originates approximately equally from milk and beef.

### **Calf management – restricted suckling versus artificial rearing**

In many developing countries where Zebu cattle and their crosses are common different suckling systems are frequently employed. In the animal husbandry system called restricted suckling the calf is often tethered in front of the cow to stimulate milk let-down (Orihuela, 1990). After milking the calf is allowed access to the dam for a limited period to suckle the residual milk and sometimes the milk in one unmilking quarter. Conversely, in industrialised countries the tradition is artificial rearing of the dairy calf, where the cow and calf are separated immediately or shortly after calving. The calf is usually reared in a single box and

offered milk or milk substitute in buckets. Large farms often raise the calves in groups with automatic milk feeding systems. For various reasons, however, a number of farms during recent years have introduced different suckling systems also in industrialised countries. In some herds the foster cow system is practised, where the cows may nurse three or four calves. On other farms the calves are allowed to suckle the dam restrictedly a few times daily (Anderberg *et al.*, 2004; Lidfors & Berg, 2004). Finally, there is at least one example of a dairy farm with an automatically milking system, where the calves are kept in the barn together with the cows with free access to suckle the dam for the first three weeks of life (Svennersten-Sjaunja, 2002).

The restricted suckling system is often described as labour intensive and has therefore been replaced by artificial rearing elsewhere and also in the tropics, especially in economies where milk is an expensive item in the diet and cheap substitute are available (Preston & Leng, 1987; Galina *et al.*, 2001). On the other hand, the labour involved in feeding and cleaning for artificial rearing is often neglected in the equation. Junqueira *et al.* (2005) found that the additional time needed to milk with the calf next to the dam was similar to the time spent on artificial feeding of calves. Nevertheless, it has been reported, that the restricted suckling or artificial rearing management system influence behaviour and weight gain in the calf (Das, 1999) and udder health (Rigby *et al.*, 1976; Knowles & Edwards, 1983) and milk production (Knowles & Edwards, 1983; Mejia *et al.*, 1998; Das, 1999) in the cow.

### **Influence of calf management on calf behaviour**

From the very first week of life calves are engaged in a number of activities such as social interactions, exploration, locomotion and running (Wood-Gush *et al.*, 1984). How these activities are expressed seems to be influenced by the rearing system. Abnormal behaviours such as excessive licking of the interior (Stephens, 1982) and licking their own bodies (Fraser, 1983; Wood-Gush *et al.*, 1984) have been found in calves kept in single pens. If licking of objects occurs frequently, it is generally considered as a need for exploration (Van Putten & Elshof, 1982). Abnormal behaviour has also been reported in group-reared calves, such as a high frequency of sucking on other calves in the group *i.e.* cross-sucking (Stephens, 1982; Dybkjær, 1988; Lidfors, 1993), which is related to an unsatisfied suckling need. Cross-sucking can cause problems for the animals' well-being as well as financial problems caused by an increased disease transmission (de Passillé, 2001), hair loss (Dybkjær, 1988) or inter-sucking and milk-stealing in adulthood (Keil *et al.*, 2000; Lidfors & Isberg, 2003). Both the performance of sucking and the duration of milk ingestion are important to reduce abnormal sucking (Loberg & Lidfors, 2001). By providing the milk through an artificial teat cross-sucking may be reduced but this is no sure-fire method (Lundin *et al.*, 2000; Veissier *et al.*, 2002) compared to when the calf is allowed to suckle its dam (Lidfors, 1993) or another cow (Lundin *et al.*, 2000; Margerison *et al.*, 2003).



## **Influence of calf management on weight gain and future production**

There are indications that the early interaction between the cow and the calf have influences beyond the beneficial effects of colostrum intake on the calf's immune system (Krohn *et al.*, 1999). Lupoli *et al.* (2000) reported that when the calf suckled, the release of oxytocin and insulin were higher compared to bucket drinking and after the suckling there was a marked decrease in plasma cortisol levels compared to following bucket drinking. It has been shown that injections of oxytocin have a growth-stimulating effect on rats (Björkstrand & Uvnäs-Moberg, 1996; Uvnäs-Moberg *et al.*, 1996). Suckling may therefore enhance growth. There are a number of studies reporting a higher weight gain during the suckling period for restrictedly suckling calves compared to artificially reared calves (Fallon & Harte, 1980; Knowles & Edwards, 1983; Jonasen & Krohn, 1991; Little *et al.*, 1991). In addition, the welfare of the animals might likely be improved, particularly since oxytocin has anti-stress effects (for review see Uvnäs-Moberg *et al.*, 2001).

Management system can have an impact on the calf's future milk-producing capacity. Sejrsen reported in his review (1994) that nutrition during the pubertal mammary growth period can affect the future development and ultimate milk-producing capacity of the mammary gland. Calves allowed to suckle had a higher average daily preweaning weight gain, a higher height at the withers, an earlier age at calving (Bar-Peled *et al.*, 1997) and a tendency to a higher milk production during their first lactation (Foldager & Krohn, 1991; Bar-Peled *et al.*, 1997).

## **Suckling behaviour in calves**

Calves with free access to their mothers suckle for a total period up to one hour daily, distributed over five to eight meals (Hafez & Bouissou, 1975). To obtain access to the milk, the calf has to induce milk let-down by tactile, vocal or visual stimulation. The stimulation evokes the release of the hormone oxytocin, which regulates the milk let-down. This stimulation includes tactile stimulation as sucking and butting with the muzzle against the udder. The teat stimulation performed by suckling calves may enhance milk production (Bar-Peled *et al.*, 1995; Mai Van *et al.*, 1997).

Cattle udders consists of four independent quarters. The production in the front and rear quarters can differ; in general there is a slightly higher yield in the rear quarters. Milk yield of each quarter is an important factor for the calf's choice of teat (Jung, 2001), but also the size and shape of the udder affect teat preference. Jung (1994) found in high-yielding cows that the calf usually becomes satiated when there was still milk available and stopped suckling after an average time of 7 min. In low-yielding cattle, the calf continues to suckle when access to milk declines and performs so-called post-massaging that includes more butting and teat change (Lidfors, 1994). There are indications that "post-massage" stimulates future milk production in pigs (Jensen *et al.*, 1998).

## **The effect of suckling on milk production and milk composition**

It can be difficult to activate milk ejection in Zebu cattle and their crosses in the absence of the calf (Ryle & Ørskov, 1990) and to achieve a sustained lactation (Everitt & Philips, 1971; Alvarez *et al.*, 1980). It has been observed that restricted suckling increases milk production in both Zebu crosses and pure Holstein (Everitt & Philips, 1971; Knowles & Edwards, 1983; Mejia *et al.*, 1998; Bar-Peeled *et al.*, 1995). Milk production is believed to be enhanced due to teat stimulation performed by the calf (Bar-Peeled *et al.*, 1995) and the increased degree of udder emptying when the calf suckles the residual milk after milking (Sandoval-Castro *et al.*, 2000). Moreover, a more frequent udder emptying in early lactation benefits the development of the milk secreting cells (Hale *et al.*, 2003). Increased milk yield can also be related to an improved udder health when cows are suckled (Preston, 1984; Mejia *et al.*, 1998).

Although suckling may induce similar or higher total milk production (saleable milk and suckled milk) in suckled cows compared to unsuckled cows, it has been reported that milk ejection can be disturbed when the cows are machine-milked during the suckling period (Bar-Peeled *et al.*, 1995; Sandoval-Castro *et al.*, 1999; Krohn, 2001). Though *Bos taurus* cows do not normally need the calf presence for milk ejection, these cows in restricted suckling systems may fail to let milk down in the milking parlour (Boden & Leaver, 1994). Preston & Vaccaro (1989) reported that up to 20% of cows in restricted suckling systems may withhold most of their milk during milking and saving it for the calf.

When calves suckle the cow after milking they ingest the residual milk, which has a higher fat content than the machine-milked milk. Consequently, the fat content at next milking is reduced as a carry-over effect (Boden & Leaver, 1994; Tesorero *et al.*, 2001), sometimes to levels undesired by the processing industry. By employing different restricted suckling systems it is possible to manipulate the composition of saleable milk (Sandoval-Castro *et al.*, 2000). Tesorero *et al.* (2001) found that in a restricted suckling system where calves suckled before milking, the saleable milk yield and fat content increased, compared to no suckling before milking. Since the calves were suckling the first milk portion that has the lowest fat content the fat content in saleable milk was elevated. It is a well known fact that milk fat content increases during milking (Johansson *et al.*, 1952).

## **The effect of suckling on udder health**

Mastitis is an inflammation in the mammary gland and is in most cases caused by bacterial infection. Mastitis appears either clinically or sub-clinically. In clinical mastitis there is one or more visible inflammatory signs present in the udder such as little redness, swelling, heat, pain and loss of function. The milk composition may be abnormal, a little watery including dots and blood and the milk somatic cell count (SCC) is increased (Sandholm, 1995). In sub-clinical mastitis, the udder and milk show no visible signs of inflammation but the milk composition might be altered. In particular the lactose content is decreased while the SCC is increased

(Claesson, 1965; Linzell & Peaker, 1972; Korhonen & Kaartinen, 1995; Berglund *et al.*, 2004). An indirect measurement of SCC is the California Mastitis Test (CMT).

Suckling seems to be advantageous for udder health regardless of the length of the suckling period (for a review see Krohn, 2001) or the type of cattle Zebu crossbred cows (Knowles & Edwards, 1983; Mai Van *et al.*, 1997; Mejia *et al.*, 1998) and in dairy cows (Everitt & Phillips, 1971; Rigby *et al.*, 1976). The lower incidence of mastitis has been attributed to mechanical factors in the suckling (Rigby *et al.*, 1976), a better udder emptying and inhibitors in the saliva (Rigby *et al.*, 1976; Mejia *et al.*, 1998). Conversely, Jung (2001) proposed that the udder health of high-producing cows might be at risk due to uneven udder emptying when suckled, since calves suckled most on front teats, which were easier to reach but still had enough milk to satisfy.

## **Aims of the thesis**

The general aim of this study was to evaluate the effect of rearing calves by restricted suckling compared to artificial rearing in an extensive dual purpose system with Zebu crossbred cows milked once daily and in an intensive dairy production system with Holstein cows milked three times daily. There are several reports describing various benefits of restricted suckling systems compared to artificial rearing systems. However few reports have taken into consideration both production and behavioural aspects in the same study.

The following objectives were to be investigated:

- calf behaviour in RS in comparison to AR system
- calf performance in RS in comparison to AR system
- milk production of cows in RS system in comparison to cows in AR system
- possibilities and limitations of RS and AR in dual purpose and dairy bred cattle

## **Material and methods**

The papers included in this thesis describe two different types of cattle management; an extensive dual purpose system with Zebu crossbred cows and an intensive dairy production system with Holstein cows.

## Location

The three studies included in this thesis were carried out at two different locations in Mexico: El Clarin Research Centre of the National University of Mexico, located outside the city Martinez de la Torre, Veracruz State, in the tropical lowlands (paper I and II) and La Escondida farm, a private dairy farm, located in the centre of State of Aguascalientes in a semi-arid zone in the highlands (paper III).

## Experimental design

In all studies the cow-calf pairs were allocated to two treatments – restricted suckling (RS) and artificial rearing (AR). Within the groups of primiparous and multiparous cows, cow-calf pairs (except for twins) were alternately distributed to the two treatments according to order of calving.

## Animals

In papers I and II, the animals used (Fig. 1) were F1 cows (Holstein × Zebu) and their calves (F1 × Simmental). In paper I, 24 cow-calf pairs were followed from calving until eight weeks of age. The cows calved from April 29th to June 8th 2002, and ranged between the first to seventh lactation. In paper II data were collected from 30 cow-calf pairs from day four after calving until the calves were nine days old. The cows calved from April 17th to June 28th, 2002, and ranged between the first to fourth lactation. A number of the cows studied in paper I were also used in paper II. The average yearly milk production of the herd was about 2400 kg per cow.



*Fig. 1. A Zebu crossbred cow suckled by her calf (Photo: Carlos Hernández).*



Fig. 2. A Holstein cow suckled by her calf (Photo: Emma Gratte).

In paper III, 27 Holstein cows and their calves (Fig. 2) were followed from calving until weaning at eight weeks. The cows calved from May 19th to July 9th 2003 and ranged between the first to fifth lactation. The average yearly milk production of the herd was about 8 000 kg per cow.

### **Feeding and management of cows**

In paper I and II all cows were grazing together on a pasture of *Paspalum spp.* and *Axonopus spp.* (90%), and *Cynodon nlemfuensis* and *Brachiaria arreceta*, including some leguminous plants as *Desmodium spp.* Minerals and water were provided *ad lib.* Cane sugar molasses was given during milking and concentrate after milking (approximately 6 kg altogether). Cows were milked once daily in the morning, in a four-unit milking parlour. During milking calves in both treatments were tethered in front of the cow such that physical contact was possible to stimulate milk let-down.

In paper III, the cows in the two treatments were housed together in an open paddock, with a ground of soil and a resting area with a shelter and a ground of peat-like dried faeces. The cows in the experiment were kept in the same paddock as newly-parturated cows. The cows were fed 23 kg dry matter (DM) daily, divided into three meals, of a total mixed ration. Half of the mixture consisted of alfalfa hay, corn silage and green chopped alfalfa, the other half was composed mostly of corn grain, and included minerals. Water was provided *ad lib.* The cows were milked three times daily in an 8×2-unit herringbone parlour.

## Feeding and management of calves

In the studies described in paper I and II, all calves were kept together with their dams on pasture until cow-calf separation five days after parturition. The calves in both treatments were kept together in a paddock including a roofed pen with a concrete floor, troughs for feed and water, and three smaller grazing areas (Fig. 3). Water, concentrate and minerals were provided *ad lib*. The AR calves were individually fed fresh whole milk with a nipple bottle. Calves were offered 3.6 litres of milk twice daily. However, five weeks after the start of the study the diet was changed and the calves were fed 3.6 litres per meal of a commercial milk substitute reconstituted as 1.1 kg milk substitute powder and 10 litres fresh whey. The RS cows were machine-milked, but one udder quarter was left to be suckled by the calves after milking. The RS calves were allowed to suckle 30 min two times daily after milking in the morning and in the afternoon. After suckling calves were separated from the cows. The calves were weaned at four months of age.

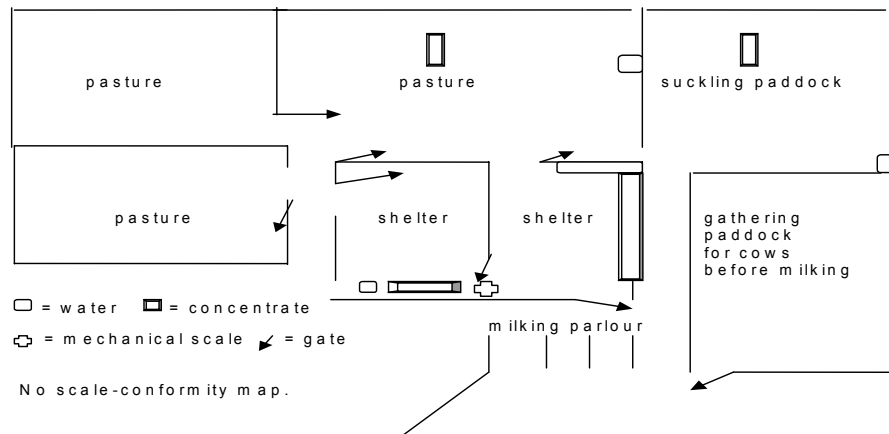
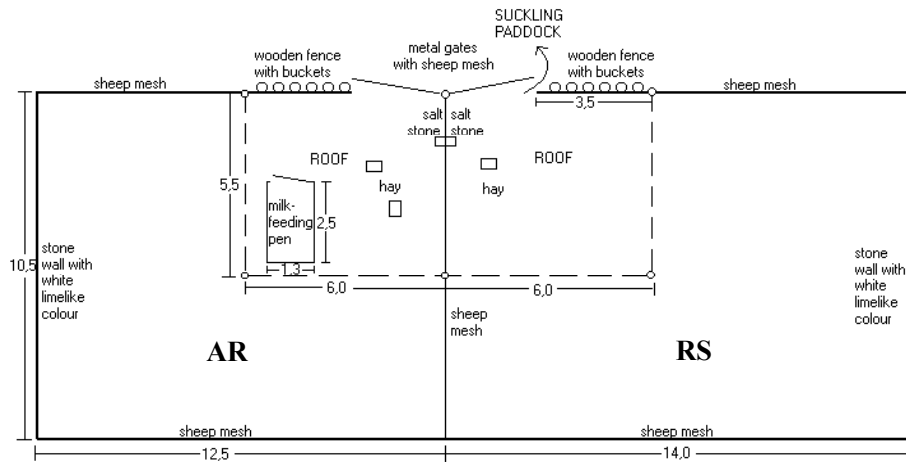


Fig. 3. Illustration of the paddock of the Zebu crossbred calves and the milking parlour.

In paper III, cows were kept with other pregnant cows in a paddock before and until shortly after calving. All calves were separated from the cow as soon as possible after calving and fed one meal colostrum from the dam with a nipple bottle. The calves in both of the treatments were kept in separate paddocks containing an area with a shelter, feeding troughs and water buckets (Fig. 4). Water, alfalfa hay, concentrate and minerals were provided *ad lib*. The AR calves were individually fed fresh whole milk from an open bucket with a floating rubber



[1:175]

Fig. 4. Illustration of the paddocks of the dairy calves. The restrictedly suckling calves were kept in the paddock named RS and the artificially reared calves in paddock AR.

nipple. In weeks 1 to 7, the AR calves were offered milk twice a day and during week 8 once daily. The daily allowance was 4 litres during week 1 and 2, 6 litres in weeks 3 to 7 and 3 litres during week 8. Two hours after morning and afternoon milking the RS calves were allowed to suckle their dams for 30 min. In week 8, calves were allowed to suckle only in the morning. If a calf showed signs of bloat it was not allowed to suckle for more than 15 min. After suckling calves were separated from the cows. The calves were weaned at the end of week 8 after birth.

## Recordings and analyses

### *Behavioural observations*

During behavioural observations cows and calves were observed as focal animals. The behaviours were recorded in a protocol with one-zero sampling.

Behavioural observations in paper I were conducted once weekly between 7 and 56 days after parturition. During milking the social behaviours between cow-calf pairs and calves, and calves' abnormal behaviours were recorded. During suckling/milk feeding in the morning, the calves' abnormal behaviours, social behaviours between RS cow-calf pairs and social behaviours between calves were recorded. Observations of the general behaviours of the calves in both treatments were conducted two hours after suckling/milk feeding in the evening.

Behavioural observations in paper III were conducted on two separate days during weeks 1, 3, 5 and 7. During suckling/milk feeding in the morning, the calves' abnormal behaviours, social behaviours between RS cow-calf pairs, social behaviours between calves and RS calves' teat preference were recorded. Two

hours after the evening suckling/milk feeding, the general behaviours of calves in both treatments were recorded.

#### *Weight, concentrate and milk intake recordings of calves*

In paper I, the body weight of calves was recorded at parturition and subsequently once weekly before morning suckling/milk feeding until 56 days of age. Calves in RS group were also weighed after morning suckling (weigh-suckle-weigh procedure), to estimate milk consumption (papers I and II).

In paper III, the weight of calves were recorded at parturition and subsequently twice weekly before morning feeding until day 56. To estimate milk consumption, calves in the RS treatment were also weighed after the morning suckling and again in the afternoon before and after suckling (weigh-suckle-weigh procedure), once weekly. The concentrate intake was recorded at a group level. To estimate the composition of ingested milk, samples were taken from RS cows and from the milk offered to AR calves once a week.

#### *Stress measurements after early cow-calf separation*

In paper II, cows' and calves' heart and respiratory rates and body temperature were recorded 15 min before milking. Blood samples for cortisol analysis were taken after suckling/milk feeding. The data was recorded once daily from day 4 to 9 after calving. Heart and respiratory rates were recorded with a stethoscope and body temperature was recorded with a rectal thermometer. Blood samples for serum cortisol concentration were taken from the cows by venipuncture of the coccygeal vein in the tail and from the jugular vein of calves. The number of steps were recorded on calves by a step-counter from 15 min before milking until the end of suckling/milking feeding. Calf body weight was recorded 15 min before milking and after morning suckling (weigh-suckle-weigh procedure), to estimate milk consumption and weight gain.

#### *Udder health, milk quality and milk yield*

In papers I and III, the CMT-test was used once weekly in the morning for estimation of udder health status. The Scandinavian scoring system was used (Klastrup & Schmidt Madsen, 1974).

In paper I, the milk yield was recorded once weekly with start at day 7 and lasting until day 56 after parturition, and milk samples were collected for analysis of milk composition. In addition, milk samples called fore-milk, were collected from each quarter directly after samples for the CMT-test week 3, 6 and 9.

In paper III milk samples were taken before and after suckling once weekly on all cows in the RS treatment. Samples from the parlour milk fed to calves in the AR treatment, were also taken once a week. Metabolisable energy (ME) content of the ingested milk was calculated from the DM and fat content.



### *Milk let-down*

In paper III, the time to evoke milk let-down was recorded manually once weekly using a stop-watch during the morning or afternoon milking. The time for milk-let down was measured from start of cleaning the teats until the milk flow reached 300 g/min *i.e.* the first recording of milk amount on the display of the milking machine.

### *Milk analyses*

In paper I and III, the milk samples were analyzed for fat, protein and lactose with mid-infrared spectroscopy (FMA2001, Miris AB, Uppsala, Sweden).

### *Blood analyses*

In paper II, the blood samples were analyzed for cortisol concentrations in duplicate by RIA using a commercially available kit (Pantex, Santa Monica, CA) validated by Godfrey *et al.* (1991).

### *Statistical analyses*

The behavioural observations (paper I and III) were tested with analysis of variance using the mixed linear models procedure (SAS, 1999), where the residuals of the output of each behaviour were controlled for normality. If the distribution was not normal, the data was transformed by a square root transformation and thereafter tested again according to the mixed linear models approach. Analyses of behaviours which were not normally distributed even after transformation were done by the Wilcoxon rank sum test or a Chi-square test (SAS, 1999). In paper III, the Wilcoxon signed rank test (Lowry, 1999-2005) was used to test for age difference of some behaviours not normally distributed. The Pearson correlation coefficient (SAS, 1999) was calculated for correlations between cross-sucking and some of the other behaviours (paper III). Teat preference (paper III) was tested with analysis of variance using the general linear models (GLM) procedure (SAS, 1999).

The treatment effects on weight gain, milk intake, serum cortisol concentration and body temperature (paper II) were tested with analysis of variance using the GLM procedure (SAS, 1999). The number of steps taken and heart beats (paper II) were analysed by the Friedman two-way analysis of variance (Siegel and Castellan, 1988).

Treatment effects on milk yield and fat, protein and lactose content of the machine-milk and the fore-milk (paper I) and time to milk let-down (paper III) were evaluated with analysis of variance using the GLM procedure (SAS, 1999). The CMT scores of cows (paper III) were analysed by a Chi-square test (SAS, 1999).

## Results

Of the initial 24 cow-calf pairs in paper I, one of the RS dams became dry due to mastitis and the data from the following period, weeks 6 to 9, were excluded from the study. One AR calf died at four weeks of age due to dehydration as a result of diarrhoea and both cow and calf were excluded completely. Of the initial 27 cow-calf pairs in paper III, five calves and five cows were excluded due to various problems such as diarrhoea and subsequent death in the calves and abomasal dislocation and mastitis in the cows.

### **Behaviour during milking and during suckling/milk feeding**

#### *Zebu crossbred cattle (paper I)*

The Zebu crossbred AR calves performed more 'lick and sniff interior' than the RS calves during milking ( $p < 0.05$ ). 'Cow social with calf' showed a tendency to be performed more frequently by RS cows compared to AR cows ( $p < 0.08$ ) and also showed a decrease with calf age. 'Cross-suck' was displayed a few times in AR calves but never in RS calves.

During milk feeding, the AR calves performed significantly more ( $p < 0.01$ ) 'calf social with calf' and 'calf lick and sniff interior' than the RS calves did during suckling. All but one of the AR calves displayed cross-sucking, while there was only one observation in RS calves ( $p < 0.001$ ). Cross-sucking was most frequently recorded during week 3.

#### *Holstein dairy cattle (paper III)*

The Holstein AR calves exhibited 'cross-suck' ( $p < 0.001$ ) and 'lick interior' ( $p < 0.01$ ) much more than the RS calves. The AR calves showed 'cross-suck' mainly during weeks 3 and 5, while it was almost absent in the RS calves regardless of age. The behaviour was mainly directed towards another calf's ear. Among the social interactions between RS cow-calf pairs, 'cow sniff calf' and 'calf sniff cow' were observed more often than 'lick' and 'rub against' each other. The AR calves performed much more 'calf sniff calf', 'calf lick calf' and 'calf rub calf' than the RS calves during suckling. 'Cow sniff calf' showed a decrease with increasing calf age.

The calves spent a significantly higher per cent of the suckling time suckling on the front teats than the rear teats ( $p < 0.01$ ). This preference was particularly pronounced during the first week of life.

## **Observations of general behaviour in calves**

### *Zebu crossbred cattle (paper I)*

The RS calves were more often recorded to 'lick self' ( $p < 0.05$ ) and 'walk' ( $p < 0.01$ ), whereas the AR calves were more frequently ( $p < 0.05$ ) recorded to 'cross-suck' and 'eat concentrate' than the RS calves. In both treatments the calves were recorded to 'walk', 'stand' and 'graze' more with increasing age, whereas 'lie' decreased. 'Lick self' differed between calf ages, but there was no apparent trend of increase or decrease.

### *Holstein dairy cattle (paper III)*

The AR calves displayed significantly more ( $p < 0.05$ ) 'cross-suck' and 'lick interior' than the RS calves. Cross-sucking was mainly directed at another calf's ear, but also to the belly and mouth. There was a positive correlation between 'cross-suck' and 'social behaviour', 'lick interior' and 'social behaviour', 'lick interior' and 'sniff interior' and a tendency of a positive correlation between 'cross-suck' and 'lick interior'. 'Eat concentrate', 'eat hay' ( $p < 0.05$ ) and 'ruminant' ( $p < 0.01$ ) were more frequently observed in the AR calves and were more common in both groups week 7 compared to week 1. The RS calves showed a tendency to perform more 'drink water' than the AR calves ( $p = 0.07$ ). With increasing calf age there was an increase of recordings of 'stand', 'move', 'lick self', 'social behaviour' in both treatments, whereas 'lie' decreased. 'Sniff interior' and 'lick calf' were more frequently recorded week 7 compared to week 1.

## **Calves' weight gain and milk and concentrate intake**

### *Zebu crossbred cattle (paper I)*

After the change from whole milk to milk substitute for the AR calves in the middle of the study some of the calves refused to drink and many of them suffered from diarrhoea. The average daily live weight gain (LWG) during the experimental period was similar for the RS ( $0.28 \pm 0.03$  kg; mean  $\pm$  standard error) and AR ( $0.24 \pm 0.04$  kg) calves. Due to the problems encountered at the abrupt change of diet this treatment difference was not tested for significance.

### *Holstein dairy cattle (paper III)*

The average total milk consumption happened to be equal in both treatments, 286 kg. The AR calves consumed all the milk given and thus no standard error (SE) is presented. The amount of suckled milk (RS) varied greatly, resulting in an SE of 2.7 kg. Due to a higher fat- and dry matter content in the suckled milk the average intake of ME from milk for the eight weeks was higher in the RS calves compared to the AR calves. The AR calves consumed more than four-fold as much concentrate compared to the RS calves, which resulted in a similar total intake of ME in the RS (1051 MJ) and AR treatment (1092 MJ).

The average birth weight was similar in calves in the RS ( $37.3 \pm 6.3$  kg; mean  $\pm$  standard deviation) and AR treatments ( $36.1 \pm 6.0$  kg). There was no difference between the RS ( $0.48 \pm 0.22$  kg; mean  $\pm$  standard error) and the AR ( $0.47 \pm 0.09$  kg) calves in daily LWG from birth to weaning. However, the individual variation in LWG was greater in the RS calves than in AR calves ( $p < 0.01$ ).

## **Stress measurements in relation to early cow-calf separation**

### *Zebu crossbred cattle (paper II)*

There was a higher heart rate in RS calves before milking compared to AR calves, while the cortisol concentration after suckling/milk-feeding was higher in the AR calves ( $p < 0.05$ ). AR induced a significantly higher level of serum cortisol concentration in cows than RS ( $p < 0.05$ ). No difference was found in calves' average milk consumption, weight gain, number of steps counted, body temperature or respiratory rate or in cows' body temperature, heart and respiratory rates.

## **Milk composition and udder health**

### *Zebu crossbred cattle (paper I)*

The RS cows produced 14% more saleable milk than the AR cows ( $p < 0.01$ ). However, the milk fat content was significantly lower in the RS cows. The amount of energy corrected milk (ECM, Sjaunja *et al.*, 1990) was calculated and the saleable daily ECM yield was similar in the RS (5.8 kg) and AR (5.5 kg) treatments. There was no difference between treatments in the protein content of the saleable milk, while the AR cows had a much lower lactose content compared to RS cows. The RS cows had lower fat content in the fore-milk than AR cows, while the AR cows had lower lactose content.

In the RS cows, 84% of the udder quarter samples had a CMT score 1, compared to 68% of the quarters in the AR cows. The RS cows also had 7% of the quarters registering a CMT score 5 while the AR cows had 14%. A further indication of an improved udder health in RS cows was the considerably lower lactose content both in machine-milk and fore-milk of the AR cows compared to the RS cows.

### *Holstein dairy cattle (paper III)*

There was a tendency to improved udder health in RS cows compared to AR cows. Nevertheless, the udder health was deteriorated in both treatments. In week 2, 13 % of the udder quarters in both groups were classified with CMT scores between 3 and 5. In week 5, 22% of the quarters in RS cows and 21% in the AR cows were classified with scores between 3 and 5. Six RS cows and five AR cows were treated for mastitis during the eight experimental weeks. Despite the

preference for front teats showed among the suckling calves there was no difference in udder health between the front and rear quarters in suckled cows. On the other hand, the majority of the AR cows' CMT scores between 3 and 5 were from the rear udder quarters.

## **Milk let-down**

### *Holstein dairy cattle (paper III)*

The time to milk let-down was similar in RS cows (68 sec) and AR cows (61 sec) both for the morning and afternoon milkings. The time to milk let-down increased with increased time after parturition, with the exception of week 8.

## **Discussion**

The main findings in this thesis are that RS reduces the frequency of abnormal sucking such as cross-sucking and licking of interior in the calves. The weight gain of the calves was similar irrespective of treatment in both Zebu crossbred and dairy cattle. There were indications of improved udder health both in Zebu crossbred cows and dairy cows, though there was only a tendency to lower CMT scores in the udder quarter of the latter breed. The Zebu RS cows' lower CMT scores taken together with the AR cows lower lactose content indicated a better udder health in the RS cows. There was a substantial effect of RS on the total milk production in the Zebu cattle.

### **Social and suckling behaviour**

The most important sense for the cow in recognition of her calf appears to be olfaction and the function of licking is to strengthen the bond between mother and young (Lidfors, 1994). This may explain why sniffing was the most common social behaviour between the cow and calf pairs in our studies on Zebu crossbred (paper I) and Holstein cattle (paper III). Sniffing is the most frequently performed social behaviour up to three months of age with a peak at seven days of age according to Kiley-Worthington & de la Plain (1983). In our study on dairy calves, 'cow sniff calf' showed a decrease with calf age as early as the third week after parturition, which may be explained by the cow-calf bonding being established at that time.

According to our observations of teat preference, the dairy calves (paper III) suckled more on the front teats compared to the rear teats, particularly during the first week of life. Previous studies have found that young beef calves (Kiley-Worthington & de la Plain, 1983) and dairy calves (Jung, 1994) suckled mainly on front teats. In high-yielding cattle, the front teats are easier to reach but still have enough milk to satisfy the calves (Jung, 1994). Important factors for the calf's choice of teat are milk yield, udder form and calf age (Jung, 1994; Lidfors, 1994).

A preference for the front teat may risk udder health in the rear quarters in beef cattle according to Kiley-Worthington & de la Plain (1983).

### **Abnormal behaviour**

The higher frequency of cross-sucking in the AR calves in the two studies presented in papers I and III, may point to an unsatisfied need for sucking in this treatment as suggested by de Passillé *et al.* (1992). Even though the dairy AR calves (paper III) obtained the same amount of milk as the RS calves and were fed from buckets with a rubber nipple, their innate need for sucking was not satisfied. The average duration of milk intake, less than 2 min, was almost 10 min shorter compared to the average suckling time in the RS calves. Maybe the cross-sucking could have been reduced by leaving the teat bucket until the AR calves stopped suckling. It has been reported that both the performance of sucking and the time taken to ingest milk are important factors to reduce the frequency of cross-sucking (Loberg & Lidfors, 2001). Cross-sucking seemed to be most common at earlier ages both in the Zebu crossbred and dairy calves (papers I and III). A decrease in cross-sucking with calf age could partly be explained by a reduced dependency on the milk as diet and partly due to the increase in other behaviours such as eating solid feed, social behaviour and playing (Das, 1999).

‘Lick interior’ occurred in both treatments in Zebu crossbred (paper I) and dairy calves (paper III), even though the behaviour was most frequently recorded in the AR calves. This is in line with Das (1999), who found that artificially reared calves up to three months of age tended to show frequent licking and nibbling of objects compared to restrictedly suckling calves. Veissier *et al.* (1998) suggested that nibbling of objects by young calves is a step in developing their feeding behaviour. As the calves grow older, only edible objects, if available, should be nibbled. It has also been proposed that deprivation of sucking can constitute a conflict and that this stress could result in abnormal biting and licking (Wiepkema *et al.*, 1987). These reports together with the findings from the present study, show that licking of interior occurs even though the calves are provided access to suckle its dam. It appears that licking of interior is not necessarily a harmful behaviour. Tongue-rolling was not recorded in any calf.

### **Eating, ruminating and other behaviours**

Irrespective of treatment, the Zebu crossbred and dairy calves (papers I and III) were observed to begin eating solid feed and ruminating after one week of age, which is in agreement with the literature (Roy, 1980; Margerison *et al.*, 2003). The foraging behaviours increased with calf age, which agrees with Margerison *et al.* (2003), and this corresponds well with the rumen development of calves. The findings that calves were lying less and standing more with increasing age were also expected and have been documented in previous research (Ylipekkala, 1990).

The Zebu crossbred calves (paper I) in the RS treatment seemed to be more active during observations of general behaviour, as they walked about and licked

themselves more, but no other differences were found between treatments. Das (1999) found that the restrictedly suckling calves walked more, but also grazed more. In the dairy calves (paper III) the calves in the AR treatment displayed much more foraging behaviour as eating hay and concentrate and ruminating. The Zebu crossbred calves in the AR treatment were also recorded to have a higher frequency of 'eat concentrate' compared to the RS treatment. It has been suggested that food ingestion could serve as a replacement stimulus to suckling (Margerison *et al.*, 2003). Also, it is probable that the AR calves were more motivated to consume food than RS calves because of their lower energy intake from milk in comparison to RS calves.

### **Stress measurements in relation to early cow-calf separation**

The physiological and behavioural indices of distress responses in ruminants include: blood hormone concentrations (*e.g.* cortisol); heart rate; breathing; body temperature; and locomotion (Mellor *et al.*, 2000). However, changes in cortisol concentration appear to be a particularly useful index of acute distress, related to the activity of the hypothalamic-pituitary-adrenocortical system (HPA). The cow-calf pairs in the AR treatment had a larger increase in cortisol concentration after milking than the RS cows and calves (paper II). This suggests that the suckling sessions resulted in less stress in the animals during the separation process. Lupoli *et al.* (2000) observed an increase in cortisol during milking but not during suckling in cows, and in calves a decrease in cortisol after suckling but not after bucket drinking.

The larger increase in serum cortisol found in the calves compared to cows, could indicate that calves are more affected by the AR than multiparous cows. Nevertheless, it is essential to keep in mind that quantitative age-related differences in cortisol responses have been found (Kent *et al.*, 1993). Moreover, according to Mellor & Stafford (1997) the relative slow response time of the HPA axis may make it insensitive when discriminating levels of stress elicited within the first few min of a negative stimulus such as separation. Heart rate may be more accurate in assessing the early stages of distress response (Mellor *et al.*, 2000). The faster heart rate in the RS calves could be due to frolicking observed but not measured during suckling. The number of steps taken was similar, probably because calves in both treatments were isolated from their mothers between the meals. Different results could be expected when calves can see, smell and/or hear their dams during calf-cow separation (Price *et al.*, 2003).

The experiment in paper II was designed to use data from day 4 in both treatments as internal controls. However, these values, although lower than days 5 to 7, were higher than data recorded day 9 (minimum values). This could be due to the stress caused by the initial exposure to the data collection procedure. Furthermore, the lowest values recorded day 9 were generally similar to those of the previous day, suggesting that the effect of treatment and handling procedures on these days had been reduced to a minimum (Andrade *et al.*, 2001).

The duration of stress in relation to separation is variable. Multiparous dairy cows abruptly separated from their calves after calving responded only mildly and immediately (Hopster *et al.*, 1995). Other studies have shown that distress responses of calves and cows were greatest 24 h after the separation (Lefcourt & Elsasser, 1995), that cows remained highly responsive to their calves throughout day 2 (Price *et al.*, 2003) and that separation after 4 days affects some behaviour (Lidfors, 1996). These reports, together with the findings from the present study in paper II, indicate that signs of stress in cows and calves do not last for more than three to four days after separation.

In paper II, the long-term affect of suckling was not measured but could be expected in both cow and calf. Lupoli *et al.* (2000) reported that cows had higher oxytocin levels during suckling compared to milking whereas the cortisol levels were increased during milking compared to suckling. In the suckling calf, oxytocin and insulin release was higher compared to calves drinking milk from a bucket and after the suckling there was a marked decrease in plasma cortisol levels compared to levels following bucket drinking. It has been found that oxytocin has anti-stress effects (Uvnäs-Moberg *et al.*, 2001) and if oxytocin is injected in rats the growth is stimulated (Björkstrand & Uvnäs-Moberg, 1996; Uvnäs-Moberg *et al.*, 1996).

### **Calves' weight gain and milk and concentrate intake**

The average LWG in both Zebu crossbred (paper I) and dairy calves (paper III) were relatively low in comparison to previous studies comprising Zebu crossbred (Knowles & Edwards, 1983; Mejia *et al.*, 1998; Hernández *et al.*, 1999), and dairy calves (Jonasen & Krohn, 1991; Fallon & Harte, 1980). However, some studies on Zebu crossbred calves covered a longer time period than the period of 8 weeks of life in this thesis. One study (Mai Van *et al.*, 1995) reported a low daily LWG in Zebu crossbred calves during the first weeks after calving, but that the daily LWG increased as the calves became older.

In the dairy calves (paper III) the amount of milk consumed in the two treatments happened to be the same. Due to a higher fat content in suckled milk, compared to the milk given to the AR calves, the RS calves received a higher level of metabolisable energy (ME) from milk. Conversely, the RS calves' concentrate intake was only 22% of that of the AR calves', which resulted in comparable daily intakes of ME and LWG in the two treatments. Earlier studies also found a higher concentrate intake in artificially reared calves; two-fold (Margerison *et al.*, 2003) and five-fold (Jonasen & Krohn, 1991) as much as restrictedly suckling calves.

In our study on Zebu crossbred calves (paper I), the average daily LWG from calving to eight weeks of age was the similar for the two treatments. In contrast, a number of studies have reported higher LWG in restrictedly suckling calves, both in dairy (Fallon & Harte, 1980; Jonasen & Krohn, 1991; Bar-Peled *et al.*, 1997) and Zebu crossbred calves (Knowles & Edwards, 1983; Little *et al.*, 1991; Mai Van *et al.*, 1997). Better growth in suckling calves has been attributed to a higher



fat content in the residual milk (Mai Van *et al.*, 1997; Mejia *et al.*, 1998) and the amount of milk (Jonasen & Krohn, 1991). Obviously, differences in LWG must be highly correlated to the consumption of milk and concentrate and forage. Nevertheless, information on feed intake is lacking in many reports.

According to calculations based on Olsson (1981) and unpublished data, the LWG in the dairy calves (paper III) in both treatments was less than expected given the ME intake from milk. The low daily LWG may partly be explained by the fact that all suffered from diarrhoea. There were also extreme weather changes and large paddock areas, resulting in more energy being spent for maintenance and movements and less for weight gain. Although the average LWG was similar, the variation in LWG between calves was much higher among the RS calves than in the AR calves. A more uniform LWG is a desirable effect that relates both to well-being and management of the calves. It may be more difficult to ensure a satisfactory nutrient supply for the individual calf in a RS system.

### **Milk yield and milk quality**

Although the Zebu crossbred cows (paper I) in the RS treatment were milked once and suckled two times daily they produced 14% more saleable kg milk than the AR cows which were milked only once daily. When including the quantity of suckled milk, the RS cows had an even higher total milk production. The increased milk yield in RS cows is probably an effect of more frequent udder emptying, a finding which agrees with other studies (Allen *et al.*, 1986; Bar-Peled *et al.*, 1995). The biological explanation is mainly a more frequent removal of a milk protein, FIL (feed back inhibition of lactation), that has a negative feedback control of the milk synthesis (Wilde *et al.*, 1995). It has also been proposed that milk production is increased due to an enhanced teat stimulation performed by the suckling calf (Bar-Peled *et al.*, 1995) and to a beneficial effect on udder health from suckling (Preston, 1984; Mejia *et al.*, 1998).

In spite of a lower milk fat content in saleable milk in the RS cows (paper I) the amount of saleable milk calculated as ECM was similar in the two treatments. The lower fat content in saleable milk in RS cows was possibly a carry-over effect of calves suckling the residual milk after milking. Removal of residual milk by suckling reduces the milk fat content (Boden & Leaver, 1994; Tesorero *et al.*, 2001), which is an undesired effect for the processing industry. The composition of saleable milk can be manipulated by employing different restricted suckling systems (Sandoval-Castro *et al.*, 2000). If calves suckle before milking, the fat content of saleable milk can be elevated (Tesorero *et al.*, 2001), since milk fat content is lower in the beginning of udder emptying (Johansson *et al.*, 1952).

In Zebu cattle it has been reported that the calf presence makes it easier to achieve a sustained lactation (Everitt & Philips, 1971; Alvarez *et al.*, 1980). In Holstein cows it was found that increased milking frequency in early lactation *e.g.* three milkings instead of two, resulted in a more sustained lactation (Österman & Bertilsson, 2005). If the milk ejection is not disturbed when the cow is machine-

milked during the suckling period as described in a review by Krohn (2001), an improved teat stimulation and frequent udder emptying performed by the calf might improve subsequent milk production.

### **Udder health**

In the study on Zebu crossbred cows (paper I), it was observed that the RS cows had a better udder health compared to AR cows, indicated both by the higher frequency of udder quarters with low CMT scores and that AR cows had lower milk lactose content both in the machine-milked milk and quarter fore-milk. However, it has to be noted that the milk samples were not analysed for SCC and bacteriology either in the study in paper I or III, whereby the findings should be taken as indications for improved udder health. In agreement with our findings, there is a number of studies on lower incidence of mastitis in *Bos taurus* × *Bos indicus* following suckling (Rigby *et al.*, 1976; Knowles & Edwards, 1983; Mai Van, *et al.*, 1997; Mejia *et al.*, 1998). Improved udder health due to suckling has been explained as relating to mechanical factors (Rigby *et al.*, 1976), a better udder emptying and inhibitors in the saliva (Rigby *et al.*, 1976; Mejia *et al.*, 1998). In contrast, in the study on dairy cows (paper III), regardless of treatment, the udder health was deteriorated with time after parturition and half of the cows in the experiment were treated for mastitis within three weeks after parturition. There are other studies with dairy cows that found no difference between restricted suckled and unsuckled cows, but in contrast to our findings, the overall incidence of mastitis was low, which was attributed to an excellent milking hygiene (Fulkerson *et al.*, 1978; Thomas *et al.*, 1981; Bar-Peled *et al.*, 1995).

It has also been observed that the teat skin condition deteriorates after suckling compared with machine-milking (Rasmussen & Larsen, 1998). Rough skin is more likely to crack and bacteria might colonise such cracks. In the study on dairy cows (paper III), the level of hygiene was poor in the housing that contributed to an environment favourable for bacteria and possibly can explain deterioration of udder health.

The RS calves in the study on dairy cattle (paper III) showed a front teat preference. Jung (1994) found that calves in high-yielding cattle mostly suckled on the front teats, which were easiest to reach and had enough milk to satisfy the calves. Kiley-Worthington & de la Plain (1983) assert that udder health in the rear quarters may be at risk if calves suckle less on rear teats. In low-yielding cattle, the calves suckle also from the rear teats, which explains the beneficial effect of suckling in Zebu crosses (Jung, 2001). However, in our RS Holstein dairy cows, the CMT scores indicated no difference in udder health between front and rear quarters despite of the calves' front teat preference. On the other hand, the majority of the AR cows' CMT scores between 3 and 5 were from the rear udder quarters.

## **Milk let-down**

In the two studies in papers I and III, there was a different milking management. In the Zebu crossbred cattle (paper I) the calves were present during milking and directly after milking suckled the residual milk and one un milked quarter. In the study of dairy cattle (paper III), the calves were not present during milking and suckling occurred two hours after milking. Although *Bos taurus* cows in general do not need the calf presence for milk ejection (Preston & Vaccaro, 1989), it has been found that up to 20% of restricted suckled cows may withhold most of their milk during milking saving it for the calf (Boden & Leaver, 1994). In another study (Fröberg *et al.*, 2005) it was found that milk let-down in cows suckled one hour before milking was seriously impaired compared to cows where calves suckled two hours after milking. Therefore it was decided to let the calves suckle two hours after milking.

The time to milk let-down was similar between the RS and AR cows (paper III), which may indicate that the RS cows did not withhold the milk during milking. In contrast, in previous studies on dairy cattle, it has been observed that the milk ejection can be disturbed when the cows are machine-milked during the suckling period (Bar-Peled *et al.*, 1995; Sandoval-Castro *et al.*, 1999; Krohn, 2001). Milk let-down occurs about 30 sec to 2 min after stimulation depending on degree of udder fill, measured from the start of teat stimulation until udder pressure increases (for a review see Bruckmaier & Blum, 1998). Considering that in our study the time to milk ejection was measured from the start of stimulation till a milk flow of 300 g/min, the finding of milk let-down time of 64.5 sec does not indicate any major disturbance of milk ejection. However, the slower milk let-down with advancing week after parturition, may imply a reduced milk ejection derived from the oxytocin unsystematically injected at milking. Bruckmaier (2003) asserts that a permanent injection of oxytocin could result in addiction as well as in reduced milk ejection.

## **Implications of the results**

### **Implications to benefit the farmer**

In industrialized countries, applied research in animal behaviour is often focused on animal welfare. On the other hand, in developing countries, animal welfare is often regarded as an unnecessary luxury for rich countries. In many countries the food supply for the population with affordable products is not sufficient. It is difficult to discuss housing conditions for farm animals from a welfare point of view when the housing conditions of the farmer is inadequate. Especially in cases where economy and animal welfare are in conflict a balance is not easy to reach. Although there are such conflicts, the understanding of the animal's needs may contribute to improve farming systems and production, and thereby also increase farming income.

In some tropical areas of Mexico the cows are only milked once daily due to lack of refrigeration facilities. Milking once a day results in lower milk production compared to twice daily milking (Davis *et al.*, 1999). Our study on Zebu crossbred cows reveals that by employing a restricted suckling system with twice daily suckling the saleable and especially the total milk production can be considerably improved. In the study presented here, a RS system indicated a simpler management procedure when feeding calves in the afternoon. Instead of storing milk from the morning milking for the AR calves, the RS dams supply the milk to their calves. Also, the RS system pointed to a better udder health in the Zebu crossbred cows according to the CMT scores and lactose content of the saleable milk compared to AR cows. Increased milk yield can also be attributed to a beneficial effect on udder health from suckling (Preston, 1984; Mejia *et al.*, 1998).

Although the concentrate consumption of the Zebu crossbred calves was not recorded in the present study the study on Holstein dairy cattle and previous studies have found considerably higher concentrate consumption in the artificially reared calves (Jonasen & Krohn, 1991; Margerison *et al.*, 2003). A high concentrate consumption might be unfavourable where ingredients of concentrate are expensive. Mexico produces only about half of its total requirements for feed grain (Peel, 2002).

### **Benefits of the RS system for cattle**

Our studies on Zebu crossbred cows showed that the RS system improved animal welfare because it allowed the cow and calf to maintain social contact, resulting in lower cortisol concentrations in the cow and calf and less abnormal behaviours in the calves. The abnormal behaviours were also reduced in the dairy RS calves compared to the AR calves. The RS dams offer their calves milk at an optimal temperature with very little possibility of becoming contaminated (Fulkerson *et al.*, 1978), which is of importance where possibilities for good hygiene may be restricted. Also, the RS system indicated improved udder health according to the higher CMT scores in RS cows and the lower lactose content in the AR cows.

Although the LWG between treatments in the dairy calves was similar, the variation in milk intake and LWG between calves was much higher among the RS calves than in the AR calves. A more uniform LWG is desirable and relates both to welfare and management of the calves. It may be more difficult to ensure a satisfactory nutrient supply and hence a satisfactory weight gain for the individual calf in a RS system. In the study on dairy calves the variation in milk intake between individual RS dairy calves was very large. This study and previous studies (Walsh, 1974; Fröberg *et al.*, 2005) show that it might be necessary to take action on restrictedly suckling calves having a redundancy of milk. In our study the suckling session was shortened, whereas Walsh increased the number of suckling calves to eight instead of the previous four on each of the high-yielding cows.

## **Implications to benefit the dairy industry**

In Mexico there is an increasing demand for more milk. The study on Zebu crossbred cows showed that the RS system maintained the saleable and increased the total milk yield. Moreover, the finding that udder health was improved may, apart from increasing milk production (Preston, 1984; Mejia *et al.*, 1998), also have a positive effect on milk constituents. Mastitis reduces milk quality, where the proportion of whey protein is increased and casein is decreased (Walstra & Jenness, 1984). A high casein content is favourable for cheese production.

The fat content of the saleable milk from the RS Zebu crossbred cows was lower compared to the AR cows. Sometimes, the amount of fat decreases to levels undesired by the processing industry. However, Tesorero *et al.* (2001) showed that by practising a restricted suckling system where calves suckles before milking, the fat content of saleable milk was elevated, since milk fat content is lower at the beginning of udder emptying. Employment of different restricted suckling system makes it possible to manipulate the composition of saleable milk (Sandoval-Castro *et al.*, 2000).

## **Conclusions**

- RS reduced the abnormal behaviours of the calves compared to AR. RS seemed to result in a higher level of activity in Zebu crossbred calves, as calves in this treatment performed more 'walk' and 'lick self', whereas they performed less 'eat concentrate'. RS resulted in less foraging behaviour in the dairy Holstein calves, compared to AR.
- Irrespective of type of animals RS resulted in similar weight gain in comparison to AR.
- RS of Zebu crossbred cattle induced fewer indications of physiological stress for both calves and their dams than AR after separation.
- RS of Holstein dairy calves resulted in a lower concentrate intake whereas the amount of milk ingested was similar to AR. Due to a higher fat content from the milk ingested by RS calves compared to the whole milk given to AR calves, the total ME intake was similar in RS and AR treatment.
- RS of Zebu crossbred cows increased the total milk yield (saleable and suckled milk) and maintained saleable milk yield, decreased the fat content in saleable milk and improved the udder health according to the elevated CMT scores and the lower lactose content in AR compared to RS

- RS of Holstein dairy cows did not impair milk let-down. There was a tendency to improved udder health in RS compared to AR, according to the lower CMT scores of RS cows compared to AR cows. RS calves preferred to suckle on the front teats compared to the rear teats, which did not impair udder health.

## Future studies – problems to be solved

In the study on dairy calves some calves ingested great amounts of milk that subsequently endangered their health. The amount of milk ingested was up to 10 kg per day in some calves. However, this amount has not been reported to be unusually big or harmful for calves. Calves may well be given such milk quantities without ensuing complications such as scouring (Roy, 1980) which was encountered in paper III. It is likely that calves having free access to their dams may distribute their suckling sessions over the day and obtain smaller amounts of milk at each suckling. Calves having free access to their dams suckle five to eight times per day (Hafez & Bouissou, 1975). If calves are kept together with the cows in a loose house system with free access to suckle the dam during the milk-feeding period the calves natural suckling pattern could be expressed. In a highly mechanised system, for instance an automatic milking system, it would be possible to control the concentrate intake of the individual calves to ensure a satisfactory nutrient supply. Probably, the milk consumption of the calves may not be possible to estimate in such a system. However, the effects on the cow's yield and milk let-down from the continuous presence of the calves should be investigated.

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## Populärvetenskaplig sammanfattning

Den här avhandlingen baseras på tre olika studier genomförda i Mexiko. Resultaten från studierna är rapporterade i tre olika vetenskapliga artiklar. Första och andra studien genomfördes i tropiskt lågländ på en försöksgård med Zebukorsningskor (Zebu × Holstein), vilka användes i ett system för kombinerad mjölk- och köttproduktion. Zebukor och zebukorsningskor är vanliga i många utvecklingsländer eftersom de är bättre anpassade till det tropiska klimatet och dess sjukdomar än Europeiska och högproducerande mjölkkor. Den andra studien genomfördes i en privatägd mjölkbesättning med Holsteinkor, belägen i en halvtorr klimatzon i högländerna. I studierna undersöktes effekten av att låta kalven få dia kon, dels kalvens beteende och tillväxt, dels kons mjölkproduktion och juverhälsa.

I de allra flesta industrialiserade länder är det brukligt att separera kalven från kon antingen direkt efter kalvningen eller efter några dagar (råmjölksperioden). Därefter utfodras kalven med mjölk eller mjölkersättning s.k. artificiell uppfödning. I flertalet utvecklingsländer är det däremot mer vanligt att hålla ko och kalv tillsammans under den tid som kalven utfodras med mjölk. Ett vanligt system är att låta kalven dia ett par gånger per dag, oftast i samband med mjölkningen. Kalven binds vanligen bredvid kon under mjölkningen för att stimulera mjölknedsläppet. Efter avslutad mjölkning får kalven dia mjölken som finns kvar i juvret, den s.k. residualmjölken och ibland även en juverfjärdedel som lämnats omjölkad i syfte att dias av kalven.

Betydelsen av att hålla ko och kalv tillsammans har börjat diskuteras även i mer intensiva mjölkproduktionssystem. I en del besättningar har man infört amko-system, där korna dias av tre till fyra kalvar vardera. I andra besättningar låter man kalven dia restriktivt ett par gånger per dag. Ytterligare ett exempel finns i Finland, där man låter kalvarna gå tillsammans med korna i ett automatiskt mjölkningssystem och har fri tillgång till att dia sin mamma de första tre veckorna.

Det finns en del forskningsresultat som indikerar att system med digivning har en positiv inverkan både på kalven och kon. För kalvarnas del har försöksresultat visat att digivning ger en god tillväxt. Även sådana problem som onormala beteenden, t.ex. att kalvarna suger på varandra, har minskat. Hos kon har man funnit att mjölkproduktionen ökat eller legat på samma nivå jämfört med kor som enbart maskinmjölkats. Dessutom har man funnit tendenser till förbättrad juverhälsa hos diade kor. Många studier har behandlat en eller några enstaka aspekter på effekten av restriktiv digivning jämfört med artificiell uppfödning. Det är dock angeläget att man i jämförelser mellan dessa två system tar hänsyn till påverkan på både ko och kalv med avseende på kons mjölkproduktion och mjölkens kvalitet och kalvens beteende, foderintag och tillväxt.

Syftet med våra studier var att undersöka effekterna av restriktiv digivning jämfört med artificiell uppfödning ur ett helhetsperspektiv. Dessutom utvärderades möjligheter och begränsningar med restriktiv digivning och artificiell uppfödning både i en kombinerad mjölk- och köttproducerande besättning med mjölkning en

gång per dag och i en ren mjölkobesättning med mjölkning tre gånger per dag. Följande uppgifter registrerades de första åtta veckorna efter kalvning: socialt, onormalt och generellt beteende och tillväxt hos kalvarna och mjölkproduktion, mjölksammansättning och juverhälsa hos Zebukorsningskorna och juverhälsa och mjölknedsläpp hos mjölkorna. I anslutning till separationen mellan zebukorsningskon och kalven fem dagar efter kalvning, registrerades uppgifter på stress såsom kortisolkoncentration i blodet, hjärt- och andningsfrekvens och kroppstemperatur.

Våra resultat visade att restriktiv digivning minskade frekvensen av onormala sugbeteenden och resulterade i en minskning av ätbeteenden t.ex. 'äta kraftfoder' hos båda raserna av kalvar. De artificiellt uppfödda mjölkkraskalvarna åt fyra gånger så mycket kraftfoder som de restriktivt diade kalvarna. Eftersom den mjölk som mjölkkraskalvarna diade var betydligt fetare än den mjölk som de artificiellt uppfödda kalvarna utfodrades med, blev det totala energiintaget lika för de båda behandlingarna. Oavsett ras var kalvarnas tillväxt lika hos de restriktivt och de artificiellt uppfödda kalvarna. Restriktiv digivning framkallade färre indikationer på fysiologisk stress hos både kalv och ko vid separation av Zebukorsningsdjuren, i jämförelse med artificiell uppfödning. Mjölproduktionen var högre både mätt i säljbar och total (maskinmjölkad plus diad mjölk) mjölmängd hos Zebukorsningskorna. Dessutom förbättrades juverhälsan hos de restriktivt diade korna, bedömt i förhållande till sämre CMT-värden och lägre laktoshalt i mjölken hos de kor som inte diades. Restriktiv digivning av mjölkorna medförde inte att mjölknedsläppet försämrades och juverhälsan tenderade att vara förbättrad i jämförelse med de kor som enbart maskinmjölkats.

Slutsatsen är att restriktiv digivning av Zebukorsningskor kan vara mycket lönsam för mjölkproducenten, då system med mjölkning en gång om dagen kombinerat med digivning två gånger om dagen förbättrade produktionen av säljbar och total mjölmängd och ökade djurens välmående. Restriktiv digivning kan tillföra positiva effekter också för kor och kalvar av mjölkkras i konventionell mjölkproduktion, men det krävs fler studier för att bättre definiera under vilka förutsättningar det är lönsamt.

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