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Effects of three colostrum-feeding methods on the behaviour and management of dairy calves

Tre olika råmjölkutfodringsmetoders påverkan på mjölkkraskalvars beteende och skötsel

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Summary

Saving time and money are always important considerations in the agricultural sector and in dairy farming the calves' management plays a central role. One of the most important aspects of rearing calves is the colostrum-feeding routines and several studies have come to the same conclusion that the volume fed, quality of milk fed and when the calves are fed after parturition, are the three most important aspects. This master thesis was done as a part of a larger research study that evaluated the effects of 3 different ways of feeding colostrum to calves during the first feeding; being fed by a bottle compared to with an oesophageal tube (OT) or if the calves first were fed with a bottle and thereafter could suckle the dam during the first 24 hours after parturition. The aim of this master thesis was to investigate the calves behaviour and management compared between the different feeding routines. The study took place at the Swedish Livestock Research Centre Lövsta in Uppsala Sweden. The study was made during February- September 2015, and 21 calves were included in the study. Both heifer calves (12) and bull calves (9) were used and 13 of the calves were of Swedish Holstein (SLB) breed and 8 were of Swedish Red (SRB) breed and the distribution between sex, breed and treatments were random. Before calving the dam was moved into a separate calving pen and within 4 hours after parturition the calf was allocated to one of the 3 treatments and moved into a separate treatment pen. Criteria's for the calves to be included in the study were: the dams had to be healthy primi- or multiparous cows whom did not have any dry-period treatment, the dams should not have suffered from dystocia, have enough colostrum for the calves (8.5% of the calves body weight), the colostrum had to be of good quality (at least 20% Brix) and the calves birthweight had to be 30 kilos or more.

During the first 4 feedings behaviours, time spent on feeding and amount fed were observed and analyzed. Behaviours were analyzed from video-recordings during the treatment (first colostrum feeding) but also 30 minutes after treatment, when the calf was left alone. During the 2nd-4th feeding the behaviours were analyzed from both direct observations and video-recordings, the same person did all the behavioral observations. The behavioural data, time spent on feeding and amount fed were analyzed with the Kruskal-Wallis test and birthweight were tested with the Shapiro-Wilk W test and analyzed with one-way ANOVA. Number of 'attempts to feed' during the first feeding were significantly ($p=0.004$) lower for calves fed with an OT compared to both the other groups. There was a tendency ($p=0.07$) that calves fed with an OT vocalized more during the first feeding. Calves from both the bottle- and the suckling group lied down earlier than calves from the OT group. Calves from the suckling group lied down during a longer duration compared to the other groups. No clear pattern was found for cross-sucking was found, however calves from the suckling group did not cross-suck at all during the 4th feeding. During the 2nd to the 4th feedings the calves that had been fed with an OT during the first feeding were in average more resistant to feeding compared to the other groups. It took significantly less ($p=0.05$) time to feed the OT group during the first feeding and significantly less ($p=0.05$) time to feed the bottle group during the 2nd feeding. It took least time in average for the first 4 feeding to feed calves from the suckling group and the most for the OT group. This was partly due to the fact that calves in the suckling group were not fed on the 2nd feeding as they suckled the dam. Calves from the OT group fed in average less during the first 4 feedings, however were the differences were not significant. There are both positive (control quantity fed, quality fed and time when being fed) and negative aspects (risk of injuring the oesophagus, the groove reflex is not triggered and stress) of using an OT. The conclusion drawn was that no advantages of using an OT during the colostrum feeding were found in regard to ease of feeding, time spent on feeding and amount fed. Due to the low number of animals included in this study, further research in the area is required to confirm the findings presented on this thesis.

Sammanfattning

Att spara tid och pengar är alltid viktigt inom lantbrukssektorn och inom mjölkproduktionen har kalvskötseln en central roll. En av de viktigaste aspekterna inom kalvuppfödning är utfodringsrutinerna av råmjölk och ett flertal studier har kommit till samma slutsats att volym mjölk, kvalitén på mjölken och när kalvarna är utfodrade efter födseln, är de tre viktigaste aspekterna. Detta examensarbete gjordes i samband med en större forskningsstudie som undersökte huruvida kalvar påverkades av 3 olika sätt att bli utfodrade råmjölk på under det 1:a målet; med en flaska jämfört med en sond eller om de först blev utfodrade med en flaska och sen fick dia modern de första 24 timmarna efter födseln. Målet med detta examensarbete var att undersöka kalvarnas beteende och skötsel jämfört mellan de olika utfodringsrutinerna. Studien utfördes på Lövsta forskningscentrum i Uppsala Sverige. Studien genomfördes mellan februari-september 2015 och 21 kalvar deltog i studien. Både kvigkalvar (12) och tjurkalvar (9) användes och 13 av kalvarna var av rasen Svensk Holstein (SLB) och 8 av Svensk Röd boskap (SRB) och fördelningen mellan kön, ras och behandling var slumpmässig. Innan kalvning flyttades kon till en separat kalvningsbox och inom 4 timmar efter födseln blev kalven tilldelad en av de 3 olika behandlingarna och flyttades till en separat behandlingsbox. Kriterierna för att kalvarna skulle kunna medverka i studien var: korna var tvungen att vara friska primi- eller multipara kor som inte var behandlade under sinperioden, inte ha lidit av kalvnings svårigheter, ha tillräckligt med råmjölk till sina kalvar (8.5% av kalvens kroppsvikt), råmjölken skulle vara av god kvalitet (minst 20 % BRIX) och kalvarnas födelsevikt behövde vara 30 kilogram eller mer.

Under de 4 första målen var beteenden, tid för utfodring och hur mycket kalvarna åt observerat och analyserat. Beteendena analyserades från videoinspelningar under behandlingarna (1:a råmjölksutfodringen) och under de nästkommande 30 minuterna, när kalven var lämnad ifred, 2:a till 4:e utfodringen analyserades både från direktobservationer samt videoinspelningar. Alla beteendeobservationer gjordes av samma person. Datan för beteenden, tid för utfodring samt födomängd testades med Kruskal-Wallis test och födelsevikt testades med Shapiro-Wilk W testet och analyserades med envägs ANOVA. Antal 'försök till att äta' under 1:a utfodringen var signifikant ($p=0.004$) lägre för kalvar matade med en sond jämfört med de andra grupperna. Det fanns en tendens ($p=0.07$) att kalvarna matade med en sond åt mer under 1:a utfodringen än de andra kalvarna. Kalvarna både från flask- och digivningsgruppen låg ned tidigare än kalvarna från sondgruppen. De kalvar som var i digivningsgruppen låg ned under en längre tid än de andra grupperna. Inget klart samband var funnit kring korsdi mellan grupperna, dock så korsdiade inga från digivningsgruppen under det 4:e födotillfället. Under det 2:a till 4:e födotillfället så var de kalvar som hade utfodrats med en sond under 1:a födotillfället mer motsträviga till att äta, jämfört med de andra kalvarna. Det tog signifikant ($p=0.05$) mindre tid att utfodra kalvarna i sondgruppen under 1:a utfodringen och signifikant ($p=0.05$) mindre tid att utfodra kalvarna i flaskgruppen under 2:a utfodringen. Det tog minst tid i genomsnitt för de 4 första födotillfällena att utfodra kalvarna från digivningsgruppen, och mest tid för de i sondgruppen. Detta berodde delvis på att kalvarna i digivningsgruppen ej blev utfodrade under det 2:a födotillfället eftersom de kunde dia modern. Kalvarna från sondgruppen åt mindre i genomsnitt under de 4 första utfodringsarna, dock var ej skillnaderna signifikanta. Det finns både för- (kan kontrollera given mängd, kvalitén given och tiden given) och nackdelar (risk att skada strupen, bollrännan kommer inte upp och stress) med att använda sondmatning. Slutsatsen dragen var att det observerades inte fördelar med att använda en sond under råmjölkutfodringen när svårighetsgraden av att utfodra, tid spenderad på utfodring och konsumerad mängd, var taget i åtanke. På grund av det låga antalet djur i denna studie, så bör fler studier inom området genomföras för att bekräfta resultaten presenterade i detta examensarbete.

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1. Introduction

Sweden's milk production today has not a strong economic stability (LRF Konsult, 2016) and it is important to save money every way possible. Being sufficient and if possible to save time would also many times save money, so it is understandable that the branch is searching for new ways to save time. A general labor cost per hour for animal production in Sweden is currently around 232 SEK (SLA, 2016). Today many of the dairy calves are separated from the dam early after parturition and are fed by hand, often through a bottle, and this has been suggested by some to be time-consuming and insufficient (Persson Waller *et al.*, 2013). Since the calves are born agammaglobulinemic it is very important that they have a sufficient transfer of immunoglobulins (antibodies) from the colostrum to get protection against different pathogens and the environment that they might need (Weaver *et al.*, 2000). As problems have been reported with the current routines other ways to feed the calves which is time-efficient and that provides a sufficient transfer of immunoglobulins have been investigated. Some would argue that a solution to this problem would be to feed with an oesophageal tube (OT) during the first feeding. Feeding with an oesophageal tube gives the opportunity to control the quantity fed, the quality of the milk fed and when the calf is feed, which is, according to some, the most important aspects in regard to failure of passive transfer (FPT) (Davis and Drackley, 1998; Weaver *et al.*, 2000; Morin *et al.*, 2001). All these aspects would be good to be able to control, but there are also disadvantages with using an oesophageal tube as well as advantages. Some of the reported disadvantages with using an oesophageal tube is that there is a risk of feeding the calf too much and too fast, hurting the mouth, throat and esophagus and a risk of getting fluid in the calf's lungs (Quigley, 2002; Kaske *et al.*, 2005; Persson Waller *et al.*, 2013). Furthermore, feeding with an oesophageal tube is an invasive procedure that requires significant restraint of the calf in order to avoid injury, which could be quite stressful for the calves. At present, to my knowledge, there are not studies made evaluating the behavioural response of dairy calves to the intubation procedure and this warrants further studies.

It is quite common that Sweden is described to have good animal welfare, but what is welfare? There are several different definitions and one is *the five freedoms* (Webster, 2001). These contains as follows; freedom from thirst, hunger and malnutrition, freedom from discomfort, freedom from pain, injury and disease, freedom to express normal behaviour and freedom from fear and distress. Another definition is that the welfare is good if the animals could sustain fitness and avoid suffering (Webster, 2001). In the Swedish organic organization KRAV there is legislations that states that the calf should suckle at least for 24 hours after parturition (KRAV, 2015). It is not allowed to feed with an oesophageal tube in organic production if the calf is not sick and cannot feed otherwise, since feeding with an oesophageal tube is classified as forced feeding, and thus not allowed (EG: 20.5 889/2008, 2016).

This master thesis was done as a part of a larger project, with the aim to evaluate the transfer of passive immunity, growth, health, behaviour and welfare of dairy calves fed colostrum with a bottle, via an oesophageal tube or a bottle then suckling the dam. The project is a 2-year project and this master thesis was done during the first stage of the study. This master thesis focused on evaluating the behavioural response, time spent on feeding and amount fed using the three different colostrum-feeding routines.

2. Literature review

Since the fetal blood supply is separated from the cows blood supply by the placenta, transmission of protective immunoglobulins is prevented in utero (Davis and Drackley, 1998; Weaver *et al.*, 2000; McGuirk and Collins, 2004). The consequence of this is that the calf is born agammaglobulinemic, which means that the calf is born with very low levels of antibodies. In regard to this condition, the calves are depending on the absorption of Ig (antibodies) from the colostrum they are fed after birth and the colostrum provides the calf with Ig from the mother. Passive transfer, which means the absorption of Ig from the colostrum through the calves' small intestine the first 24 h after birth, is protecting the calf from several diseases until the calf's own immune system is working (Weaver *et al.*, 2000). With an adequate passive transfer of Ig there is a lot to gain such as; a lower risk of pre-weaning morbidity and mortality, lower mortality in the post-weaning period, higher feed efficiency, lower age at first calving, enhanced milk production for both 1st and 2nd lactation and also a decreased risk of culling in the 1st lactation (Robison *et al.*, 1988; DeNise *et al.*, 1989; Wells *et al.*, 1996; Godden, 2008).

Therefore, having good colostrum feeding routines that ensure adequate transfer of passive immunity shortly after parturition are essential both for the health and the survival of the calf. Today the most commonly used colostrum feeding routine for dairy calves in Sweden is feeding with a bottle, but in for example the USA and in Denmark it is common to use an oesophageal tube instead (OT) (Persson Waller *et al.*, 2013). Feeding with an oesophageal tube could be time saving and it is possible to control the amount fed, which is positive aspects (Persson Waller *et al.*, 2013). Nonetheless, there are also negative aspects to this feeding routine, such as that the groove reflex is not triggered and the colostrum is going into the rumen instead of the abdomen (Lateur-Rowet, 1983), there is a risk of getting fluid in the lungs that could cause pneumonia (Quigley, 2002; Kaske *et al.*, 2005; Persson Waller *et al.*, 2013) and there is also a risk of hurting the calf's larynx and esophagus (Stull and Reynolds, 2008). Since feeding with an oesophageal tube has begun to be used as a routine as well in Sweden (Persson Waller *et al.*, 2013) this thesis was made to evaluate the behavioural response of the calves when comparing being fed with a bottle to being fed with an oesophageal tube during the colostrum feeding.

2.1 Behaviours

2.1.1 Suckling and cross-sucking

It is natural for the calf to suckle the dam and it has been showed that calves has a high motivation for sucking and by giving them the chance to suckle it increases their satiety, the time they sleep, secretion of hormones that have a central role in digestive function and it also decreases the nonnutritive sucking (de Passillé, 2001; Vasseur *et al.*, 2010). Calves fed milk with a bucket are not able to express their natural sucking behaviour, but when fed milk by a teat-based system they are able to express such a behavior (de Passillé, 2001; von Keyserlingk *et al.*, 2009; Vasseur *et al.*, 2010).

Flower and Weary (2001) stated that a separation between calf and dam that was postponed after parturition (after 14 days) would induce a better health, weight gain, social behaviour development for the calves when compared to calves that were separated 1 day after birth

from their mother. Although, Flower and Weary (2001) acknowledged that the bond between the dam and her calf is likely to get stronger with increasing time spent together which would make the distress higher when separation was delayed. Wagenaar and Langhout (2007) found no negative effect of suckling on the animal health. They rather found lower incidence of diarrhea in calves that suckled compared to bucket-fed calves. However, when housing calves that are fed milk together there is a possibility for cross-sucking (Wagenaar and Langhout, 2007). De Passillé and Ruschen (2006) suggested that cross-sucking could be decreased or even eliminated if the calves are given their milk with free access from an artificial teat. They also thought that the performance of sucking behaviour is responsible for decreasing the sucking motivation more than the ingestion of milk (de Passillé and Ruschen, 2006). Cross-sucking that occurs between calves that are raised away from the dam is considered to be detrimental for the calves' welfare (de Passillé *et al.*, 1992). Jensen (2003) also defines cross-sucking as a non-nutritive sucking and a detrimental behaviour. The author claims that cross-sucking is only observed in calves that are artificially reared and not by calves that are nursed by their dam. Loberg and Lidfors (2001) also proposed that to be able to prevent or decrease abnormal sucking the calves should be able to suck the milk and the flow of the milk should be slow because then hopefully the calf's motivation to suck end by the same time the milk supply is finished. Both de Passillé (2001) and Jensen (2003) agreed with Loberg and Lidfors (2001) results.

In the study by Krohn *et al.* (1999) calves that were housed individually had higher duration of non-nutritive sucking. Calves that were housed with the dam and were allowed to suckle had lower frequency and duration of non-nutritive sucking after the treatment period, than the other groups in the study. Time spent feeding from a bucket did not differ between the individually housed calves and those who were housed with the dam but not allowed to suckle. Even so, the suckling-calves spent 8 times longer on feeding. Licking on surroundings were lower for calves that suckled and higher for those in single pens, but the difference was not significant. No difference was found between sucking on other calves between the different groups (Krohn *et al.*, 1999). Fröberg *et al.* (2008) findings were similar in regard to cross-sucking but also showed that artificially reared calves ate more concentrate, hay and ruminated more than calves that suckled. Fröberg and Lidfors (2009) concluded that calves that were allowed to suckle freely showed less non-nutritive behaviours, they also rested more and ate less solid feed compared to calves that were fed from an automatic feeder with milk substitute.

2.1.2 Distress behaviours

Vocalizing, sudden stop to play behaviour and pacing could be interpreted as distress behaviours (Enríques *et al.*, 2010). Acute pain in animals can lead to that they show behaviours such as a loss of mobility, vocalization, loss of appetite and less mobility according to Stull and Reynolds (2008). Broom (1983) wrote that if a stereotype, irrespectively of which, is displayed for 10 % or more on the animals' life (awake time), it is a sign of bad welfare. The onset of different stereotypes can be stimulated by feeding frustration, for example like suppressed feeding (quantitatively and qualitatively) and artificial feeding (Sato *et al.*, 1994).

Kiley-Worthington and de la Plain (1983) studied the vocalization of new-born calves, and they established that a new-born calf vocalize very rarely. When the calves did vocalize Kiley-Worthington and de la Plain (1983) thought that it was a response to a situation that was uncomfortable, but they pointed out that it was hard to be certain that they had interpreted this correctly. Even though they thought it hard to get a satisfactory explanation to why new-

born calves vocalize, they did conclude that the calf vocalized only seldom as an answer to the dam calling (Kiley-Worthington and de la Plain, 1983). Thomas *et al.* (2001) proposed that providing a calf with a higher quantity of milk would be beneficial for the calf by making the calf less hungry. Thomas *et al.* (2001) also showed that calves usually vocalize when they are hungry and by giving them a greater supply with colostrum and milk, vocalizing could be minimized.

2.1.3 Behaviours indicating good welfare

Stull and Reynolds (2008) suggest that when calves are showing play behaviour it is a sign of good welfare. This because the young in different species is motivated to express play behaviour when and if the primary needs is met (Stull and Reynolds, 2008). Grøndahl *et al.* (2007) described locomotor play behaviour as kicking, bucking, turning and galloping. According to Kiley-Worthington and de la Plain (1983) self-grooming and scratching only occurs occasionally in the first 3 hours after parturition in (beef) calves that stayed with their dam.

2.2 Methods for feeding colostrum

There are several ways to feed calves colostrum, for example through a bottle, through an oesophageal tube or letting the calf suckle the dam. All of these different ways to feed colostrum could affect when the first feeding takes place, how much (volume) is fed and how efficient the Ig absorption is (Brignole and Stott, 1980; Besser *et al.*, 1991).

2.2.1. Bottle-feeding

Bottle-feeding in this thesis refers to that the colostrum is poured into a bottle (with a rubber-teat) and manually fed to the calf and is, according to Persson Waller *et al.* (2013), the most common colostrum feeding routine to healthy dairy calves in Sweden.

2.2.2 Oesophageal tube

This feeding method refers to that the farmer place the colostrum in a bag that is connected to a tube and then the tube is moved through the calf's mouth and down through the oesophagus to the rumen. Feeding with an oesophageal tube as a routine is quite common in for example Denmark and the USA (Persson Waller *et al.*, 2013). There are some known problems associated with feeding with an oesophageal tube. One is when providing fluid through an oesophageal tube the oesophageal groove reflex is not triggered, and this results in that the fluid is going into the rumen instead of the abomasum (Lateur- Rowet and Breukink, 1983). In new-born calves the rumen has a very small volume compared to the abomasum (Kaske *et al.*, 2005). Therefore, when milk is transferred to the rumen instead of the abomasum it could lead to a development of rumen acidosis or anaerobic conditions and become a risk factor for development of diarrhea (Persson Waller *et al.*, 2013). However, one of the advantages Persson Waller *et al.* (2013) point out are that it is a relatively quick procedure and that it is possible to give the calf quite a large volume of colostrum or milk. It is possible to control volume ingested, time for ingestion and quality of feed ingested by using an oesophageal tube, which are all positive aspects.

When feeding with an oesophageal tube there are risks of feeding the calf too much milk and also too quickly, which could be detrimental for the calf. There is also a risk to harm the calves mouth, throat and esophagus if there is some defect on the tube or if the person conducting the feeding uses excessive force or if the calf struggles. Furthermore, there is also risk of fluid getting in the lungs, if the tube is inserted to the trachea instead of the oesophagus or the calf is fed a too large volume and this could for example cause pneumonia (Quigley, 2002; Kaske *et al.*, 2005; Persson Waller *et al.*, 2013). Stull and Reynolds (2008) also put

emphasis on that it is important that, if intubating a calf, it is done carefully since there is a risk for hurting the calf's larynx and esophagus. It is also important to consider the risk of infection if the equipment (the tube) is not thoroughly cleaned and bacteria could grow in the tube and could be transferred directly into the calf's gastrointestinal tract during the time when the calf is most sensitive towards new infections (Quigley, 2002; McGuirk and Collins, 2004; Godden, 2008). However, this risk also occurs when using rubber teat bottles for feeding if they are not thoroughly cleaned and disinfected (Quigley, 2002; McGuirk and Collins, 2004; Godden, 2008).

Lateur-Rowet and Breukink (1983) and Persson Waller *et al.* (2013) also mentions that if the colostrum is transferred to the rumen instead of the abomasum there would be a delayed uptake of antibodies. According to Quigley (2002) feeding with an oesophageal tube could result in a lower serum concentration of antibodies and have less efficiency in regard to absorption, if compared to feeding with a bottle. However, several studies have presented results that indicate that this delay will not matter if the volume fed is large and the quality is of good measure (Adams *et al.*, 1985; Godden *et al.*, 2009; Elizondo- Salazar, 2011). Lateur-Rowet and Breukink (1983) found that the delayed uptake of antibodies was approximately 3 hours. However, only three calves were used in this experiment (Lateur- Rowet and Breukink, 1983). It is suggested by Persson Waller *et al.* (2013) that to be able to be sure of an adequate and fast uptake of antibodies the volume of colostrum fed should probably not be lower than 3 liters, no matter the content of antibodies. Persson Waller *et al.* (2013) also mentions that there is a higher likelihood that calves fed by an oesophageal tube the first meal, will not want to feed the second meal, this irrespectively of which volume that was fed. Even so, Persson Waller *et al.* (2013) also stated that if there are sick or weak calves it could be lifesaving to feed them by an oesophageal tube and Vasseur *et al.* (2010) agrees that it could be a suitable method to feed neonatal calves with complications of voluntarily intake of colostrum.

2.2.3 Suckling

Quigley *et al.* (1995) showed that calves that were allowed to suckle the dam had higher IgG and IgM concentrations at 24 hours of age, compared to bottle fed calves. However, Franklin *et al.* (2003) found that calves that only suckle the dam do not always get colostrum with good quality, enough volume of colostrum or at the right time. All of these aspects increase the risk for FPT (Franklin *et al.*, 2003). Besser *et al.* (1991) also showed that suckling calves had higher prevalence of FPT than calves fed by bottle or with an oesophageal tube. Mee (2008) showed different results and concluded that if the risk for infectious diseases is acceptable, it could be advantageous for the calf to stay with its dam to be able to increase the opportunity to suckle in a natural way and also increase the absorption of colostrum immunoglobulins compared to when not staying with its dam.

According to Krohn *et al.* (1999) calves that were allowed to stay together with their dam (both suckling and no suckling allowed) for the 4 first days after parturition had a much higher growth (about 100 %) per day compared to calves that were housed individually without any contact with the dam. Fröberg *et al.* (2008) found that calves that suckled (restrictedly twice/day for 30 minutes) or were artificially reared had similar live weight gain, although there were more differences between different individuals than between calves that suckled. However, a study by Roth *et al.* (2009) showed that before weaning the growth was higher in calves that could suckle the dam.

When calves get the possibility to suckle on the dam, they can consume significantly more milk and grow faster than if they were fed conventionally according to Flower and Weary

(2003). Diaz *et al.* (2001) and Shamay *et al.* (2005) showed similar results without keeping the calf with the dam. They draw the conclusion that allowing higher intakes of milk leads to higher body weight gains, better feed conversion and a lower age at first breeding. There are reports that many calves suffered from FPT when they had suckled the dam (McGuirk and Collins, 2004; Godden *et al.*, 2008). These findings could have different explanations, which could be mastitis or other illness in the dam, the calf could be weak or injured or the dam could have too large udder for the calf to suckle (McGuirk and Collins, 2004; Godden *et al.*, 2008). Since acceptable concentrations of serum IgG could be accomplished without housing the cow and calf together, and also since there is a risk of exposing the calf to the cows environment, it is recommend of some today that the calf is moved from the cow 1 hour postpartum, or even as early as 30 minutes postpartum, and be fed colostrum by hand (McGuirk and Collins, 2004).

2.2.4 Ease of feeding and time spent feeding

A study by Metz (1987) indicated that calves that are allowed to suckle their dam for some period (10 days) had not more difficulties to learn how to feed from a bucket than those that were separated from the dam immediately after birth. Vitale *et al.* (1986) showed that with increasing age of the calf the duration of the suckling bouts increased, even though the daily number of suckling's and the total time per day spent suckling decreased.

2.3 Colostrum management

Since it is established that a sufficient passive transfer of Ig is essential for a good management of calves it is important to know how to achieve this. First the calf needs to consume an adequate amount of Ig through colostrum intake. After that the calf need to absorb a satisfactory amount of these molecules into their circulation. Factors that are affecting the quantity of Ig the calf can consume is for instance the volume and quality of the colostrum fed (Godden, 2008). Regarding the absorption of Ig molecules into the circulation, there are other factors that affect this as well. The greatest factor is how quickly the calf is fed the first colostrum after birth according to Godden (2008). It has been recommended that calves should be fed at least 100 grams of IgG during the first feeding by Davis and Drackley (1998). However, others recommend that in average a calf would need to ingest at least 123 grams of colostral IgG 2 hours after parturition and 164-226 grams of colostral IgG if fed 6 hours after parturition (Chigerwe *et al.*, 2008).

An analysis made by Wells *et al.* (1996) showed that during the first 3 weeks of the calves' lives there was a 31% mortality rate amongst heifers in the USA. One of the acknowledged reasons for this was poor management in regard to colostrum handling. The study estimated pre-weaning mortality before 3 weeks of age of the calf to be connected to failure of passive transfer (Wells *et al.*, 1996). Mee (2013) did a review of perinatal calf mortality rates in different countries during the years of 2000-2011. In the USA calves that were born by primiparous cows had a mortality of 12.1% but when combining the mortality of the calves born both from primiparous and multiparous cows, it was 8.0%. This could be compared to the mortality of calves born from primiparous cows in Sweden, which was 3.6% and the mortality of calves born by multiparous cows that was 2.5%. The difference between the countries could be influenced of that in Sweden the perinatal calf mortality is death within 24h of parturition, but in the USA it is described as death at birth (Mee, 2013). In Sweden there was a mortality of 2.4 % the first month after birth for calves born in 2013 and there were indications that bull calves seemed to have higher mortality than heifer calves (Jordbruksverket, 2014a).

2.3.1 Quality of colostrum

When referring to the colostrum quality in this thesis it is referred to the Ig content, if not otherwise stated. The content of IgG is approximately 85-90%, IgA 5% and IgM 7% in colostrum of the total Ig. Transfer of Ig goes from the bloodstream through the mammary barrier into the colostrum (Larson *et al.*, 1980). Things that can affect the quality (Ig content) of the colostrum are for example breed (Muller and Ellinger, 1981; Guy *et al.*, 1994), age of the dam (Muller and Ellinger, 1981; Pritchett *et al.*, 1991), season of calving (Nardone *et al.*, 1997; Morin *et al.*, 2001) and volume of colostrum produced (Pritchett *et al.*, 1991). Some studies have shown that older cows seem to produce colostrum of higher quality (Muller and Ellinger, 1981; Pritchett *et al.*, 1991). Nardone *et al.* (1997) stated that cows that are exposed to high temperature at the end of the gestation have poorer colostrum quality. Pritchett *et al.* (1991) concluded that cows that produced less than 8.5 kilograms colostrum the first milking were likely to have better colostrum quality than cows that produced more than 8.5 kilograms.

2.3.2 Volume of colostrum

Unfortunately the majority of producers today do not know how much IgG their cow's colostrum contains and due to this it is common to give recommendations based on volume fed to the calf rather than grams of IgG fed to the calf. According to Godden (2008) it is recommended to give an amount of milk that is equal to 10-12% of the calf's body weight. Conneely *et al.* (2014) found on the other hand that calves fed 8.5% of their birthweight in colostrum had higher IgG serum content than calves fed 7% or 10% of their birthweight. Nilsson (2015) claimed that the volume colostrum fed do not matter for the uptake of immunoglobulins but it is the content of IgG in the colostrum fed that has a big impact.

Hopkins and Quigley (1997) investigated whether or not calves were affected by being provided with colostrum one time with the whole volume or divided into two times. They found that there was no effect of providing the calf colostrum two compared to one time. In the study they did not find differences in IgG concentration in serum concentration or in the calves' growth, and even if the calves were fed colostrum supplement to increase their intake of IgG this did not affect the serum concentration. Hopkins and Quigley (1997) draw the conclusion that if calves are fed colostrum of good quality it does not seem to be any additional advantages by giving supplements. Sakai *et al.* (2012) made the conclusion that calves do not have higher serum concentration when fed 4 liters of colostrum compared to 3 liters, at least not when fed by an oesophageal tube. Jasper and Weary (2002) also conducted a study on this, and they concluded that calves that were fed through a bucket two times per day ingested 89 % less milk than those fed *ad libitum* from a rubber teat. De Paula Viera *et al.* (2008) and Borderas *et al.* (2009) suggest that there is evidence that calves are suffering of hunger when they are fed a restrictive diet.

The maximum volumes of colostrum possible to apply to calves differ between the sizes of the calves. There is a recommendation from McQuirk and Collins (2004) to give a maximum of 4 liters to calves of Holstein breed, but a maximum of 3 liters to calves of a lesser size, such as Jersey or Ayrshire bred.

2.3.3 When to feed colostrum

For optimal Ig transfer through the gut epithelium, the calf should be fed within 4 hours postpartum. After 12 hours postpartum the efficiency of Ig absorption is gradually decreasing (Weaver *et al.*, 2000).

2.4 Colostrum composition

Colostrum contains a lot of different constituents, such as Ig, cytokines, growth factors, maternal leukocytes and different nutrients (Godden, 2008). Many of these constituents have greater volume in the first milked colostrum and are steadily decreasing for the next six milkings (transition milk). The concentration has by then decreased to the quantities that are commonly measured in saleable whole milk (Foley and Otterby, 1978).

An easy way to measure the concentration of antibodies in the colostrum on the farms is by using a colostrometer. If using a colostrometer it would be easy to control that the colostrum fed to the calves is of good quality. To measure the passive transfer of immunity has to be made through a blood test though (Vasseur *et al.*, 2010).

3. Aim

The aim of this thesis was to evaluate the behavioural response to different colostrum feeding methods on dairy calves, 4 h after birth, when the calves either were fed with a bottle, via an oesophageal tube or bottle-fed and suckled its dam. Also the management routines for the different colostrum feeding methods were evaluated.

This master thesis aimed to answer the following questions:

- 1) Do calves fed colostrum with an oesophageal tube during the first colostrum feeding show different behavioural responses during the feeding compared to bottle-fed calves? Do the calves fed with an oesophageal tube vocalize more, move hind legs more often, slip more or falling down more compared to calves fed with a bottle? Or do calves fed with an oesophageal tube need more attempts to feed during the first feeding, try to lie down more often or push forward more than calves fed with a bottle?
- 2) Do calves fed colostrum with an oesophageal tube during the first colostrum feeding lie down earlier and spend more time lying down after the first feeding than calves fed with a bottle?
- 3) Does the calves fed with an oesophageal tube during the first feeding cross-suck more than calves fed with a bottle during the 30 minutes post first colostrum feeding and/or during the 2nd-4th feedings?
- 4) Are there any differences in ease of feeding during the 2nd to the 4th meal, when comparing bottle-fed calves to calves fed with an oesophageal tube during the first colostrum feeding? Or are there differences in ease of feeding during the 3rd to 4th feeding when all three treatment methods are included?
- 5) Does it take less time spent on feeding in total if feeding with an oesophageal tube during the first colostrum feeding compared to feeding with a bottle, when taking feeding number 1-4 in consideration?
- 6) Does calves fed with an oesophageal tube during the first colostrum feeding feed more in average compared to bottle-fed or calves that first were bottle-fed and then could suckle, when taking feeding 1-4 into consideration?

4. Materials and Methods

This study was conducted at the Swedish Livestock Research Centre Lövsta, which belongs to the Swedish University of Agricultural Sciences (SLU), in Uppsala Sweden. At Lövsta there is space for 300 milking cows. The study was made during February- September 2015.

4.1 Animals and housing

In the study 21 dairy calves of the breed Swedish Holstein (SLB) (n=13) and Swedish Red (SRB) (n=8) were included. Both heifer (n=12) and bull (n=9) calves were used in the study and the distribution between the breeds, sex and treatments were random.

Before calving the cows were moved from a group pen with cows in gestation into separate calving pens (length: 143 cm, width: 143 cm and height of the walls: 150 cm), which were opposite the group pen. The calving pen had sawdust on the floor, lockable feed gates in front of the manger and a water bowl. The calf was moved into a separate area within the same building to a treatment pen where calves could not hear or see their dam (figure 1). The measurements for the treatment pen was: length: 150 cm, width: 114 cm and height of the walls: 77 cm. During the first 7 days of life, the calves were kept indoors in individual calf pens (length: 123 cm, width: 101 cm and height of the walls: 95 cm), see figure 2. The calves had free access to water and access to concentrate and hay from day 1. The calves had sawdust bedding in the calving pen, the treatment pen and in the tiestall. The calves had always at least two companion-calves at all times.

Criteria's for the calves to be included in the study were: the dams had to be healthy primi- or multiparous cows, with no dry-period treatment, free from dystocia, with enough colostrum to feed the calves (8.5% of the calves birthweight), with colostrum of good quality (at least 20% Brix) and the calves birthweight had to be 30 kilos or more.



Figure 1: Treatment pen. Ida Lundmark. 2015.

Figure 2: Tiestall pen. Ida Lundmark. 2015.

4.2 General management

In the calving pens the cows had on an udder net which prohibited that a new-born calf would suckle. After parturition the calf and dam were kept together and undisturbed in the pen for 2 hours, so the dam had the opportunity to lick the calf. Two different persons applied the treatments to the calves.

In the 2nd to 4th meal fed in the tiestall the calves were fed 3 liters twice a day of the dam's colostrum/transitionmilk in a nipple-bucket. The milks temperature was measured (it was heated up to 40°C) and the milk was weighed (Scout: Pro, Vetek, Sweden). The reading range was in grams with one decimal. If there were any leftovers by the end of the feeding these were recorded as well, also the ease of feeding was recorded. The staff at the research farm carried through the 2nd to 4th feedings while an observer was present to measure the time the feedings took to and measure the amounts fed and, if any, the leftovers. The observer also recorded the behaviours. The timekeeper was a 1/100Chronograph with a reading scale of min:sec. A list of routines was written so that the feedings followed the same procedures (Appendix 1).

4.3 Treatments during colostrum feeding

The calves were randomly allocated to one of three treatments: bottle-feeding, feeding via an oesophageal tube or bottle-feeding plus suckling on the dam until 24 hours after parturition, and the treatments were applied within 4 h after birth. All calves were fed colostrum from the dam. For treatment calves were moved to the treatment pen where they were fed according to their treatment. Calves were kept in the treatment pen for 30 minutes after treatment was done to video-record their post-treatment behaviour. Thereafter, bottle and OT calves were moved to individual pens in a separate room in the tiestall. Suckling calves were moved back to their dam and kept with their mother until 24h after calving where they could suckle freely. After this time with the dam the calves were moved into a single pen in the tiestall with the other calves and followed the same procedures as for the other treatments. The management procedures for the different treatment methods are displayed in figure 3. The tube used in the study was a colouquick.

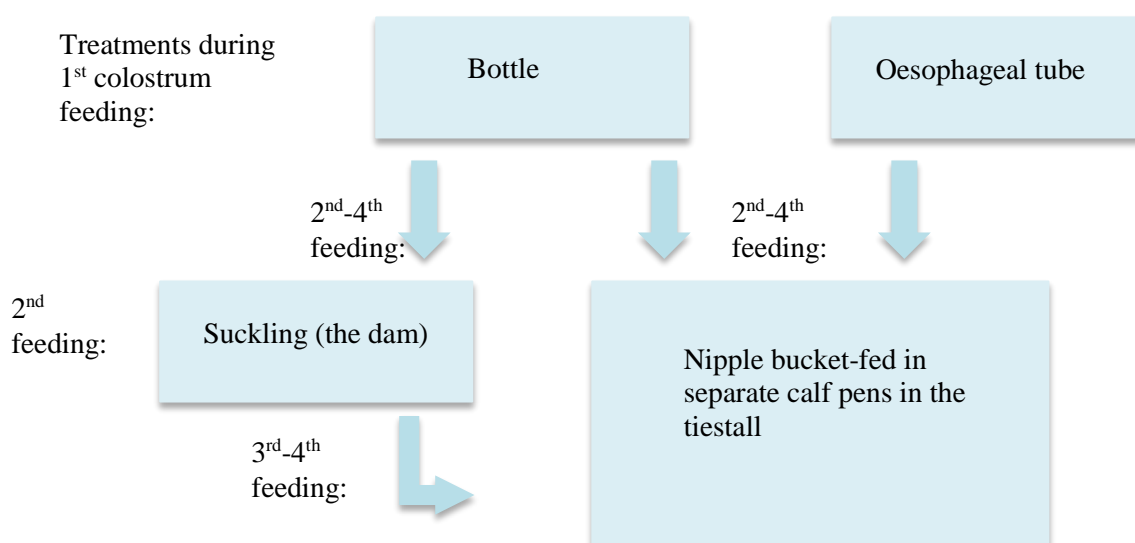


Figure 3: Management procedures for the different treatment methods during the 1st-4th feeding.

4.4 Data collection

4.4.1 Behavioural observations

The calving pen, treatment pen and the pens in the tiestall were continuously video-recorded. Treatments were video-recorded (Sony Handycam HDD 30GB, model DCR-SR35E) and the recording continued for 30 minutes after the colostrum feeding was finished. During the 30 minutes observation after the treatments, the calves were left alone. Behaviours during and after the treatment were evaluated from video-recordings and the videos of the treatments and “the 30 minutes after treatment” were recorded in two separate files. Therefore, it was possible for the observer of the behaviours to be blind to the treatments since the videos of “30 minutes after” were always analyzed before the videos of the treatments. The videos were always analyzed by the same person. The video program used was VLC media player version 2.2.1 Terry Pratchett (Weatherwax) (Intel 64bit), and the data was transferred into excel sheets for further statistical analyze. The observer collected direct observations of the behaviours during the 2nd to the 4th feeding on most occasions. If the usual observer could not attend for any reason, the behaviours were collected from video-recordings (Avtech, USA) of the feedings. Therefore the observer of the behaviours during the 2nd-4th feedings was also always the same person. More than behavioural observations, the amount fed and how long each feeding took were also measured from the 1st-4th feeding.

Behaviours recorded were divided into three parts: Part 1A (table 1) was behaviours during the colostrum feeding (1st feeding/treatment), part 1B (table 2) was behaviours recorded during the 30 minutes after the colostrum feeding and part 2 (table 3) was behaviours recorded during the 2nd to 4th feedings.

Table 1: Behavioural definitions of the behaviours recorded in dairy calves during the first colostrum feeding either with a bottle or an oesophageal tube

Behaviours – Part 1A	Definitions
Attempts to feed	Times the rubber teat or tube was put into mouth
Hind leg movements	Lifting legs up from floor and then put them down again, not slipping. Each leg was counted separately. Was measured from standing position of the calf
Slipping	Both slips from front and hind legs were recorded. When the calf slipped with one or more legs in any direction while putting pressure on it/them. From standing position
Vocalizations	Any vocalization. Each separate time calf opened mouth and vocalized were counted as one time
Attempts to lie down	When the calf was down on both front knees but not the hind legs, and had started from a standing position, but did not lie down and got the front legs up again
Lying down, voluntarily	All four legs were down on the floor. Starting from a standing position. Not lying down due to falling or slipping
Falling down	When the calf due to struggle or slipping fell down with all four legs on floor or to a sitting position. Starting from a standing position
Make the calf stand	When the human had to help the calf to a standing position after the calf had lost support of two or more legs or had lied down voluntarily
Calf is pushing forward	When calf tried to push, leap or jump away from feeding device and human in a forward direction

Table 2: Behaviours that were recorded after completed colostrum feeding up to 30 minutes after completed colostrum feeding to dairy calves by a bottle or an oesophageal tube

Behaviours - Part 1B	Definitions
Number of times lying down, and duration of time lying down (seconds)	With legs folded into the body, and head resting on the legs or floor or with holding head up. Or with legs and head stretched out. Lied down voluntary or dropped down
Latency to first lying down (seconds)	At what time the calf first lied down on all four legs
Attempts to lie down	When the calf was down on both front knees but not the hind legs, and had started from a standing position, but did not lie down and got the front legs up again
Self-grooming (yes/no)	Licked and scratched own body
Cross-sucking (yes/no)	Sucked and licked on interior and/or humans
Locomotor play (yes/no)	Running and jumping
Locomotor activity	The activity of the hind legs was measured. Each time one claw crossed the line of one square in a grid, it was counted as one activity
Escape attempts	Putted the head outside of the pen while pushing on the gates
Attempts to stand up	When the calf was starting from a lying position and got the hind legs up but not the front legs and lied down with the hind legs again
Standing up	When the calf was starting from a lying position and got up on all four legs
Vocalizations	Any vocalization. Each separate time calf opened mouth and vocalized were counted as one time

Table 3: Behaviours that were recorded during the 2nd to 4th feeding of dairy calves, which were fed either with a bottle or via an oesophageal tube during the first colostrum feeding

Behaviours – Part 2	Definitions
Help to start feeding (yes/no)	Help to find rubber teat, holding the head by the rubber teat, putting fingers with milk on in the mouth to get to begin to suckle. For the first feeding bout
Cross-sucking (yes/no)	Sucked or licked on interior or human. Measured as one time during the whole feeding period
Number of feeding bouts	How many times. Also was the ease of the feeding for each time/bout the calf took the rubber teat again recorded (Resistant to feeding, actively looking to feed or passive to feeding)
Ease of feeding for each feeding bout	
Resistant to feeding	Stopped to feed and needed help to continue feeding if it had stopped, tried to lie down, tried to back away. (Except for the first time the calf began to feed)
Actively trying to feed	Searching for rubber teat, bumping the bucket, feeding without help. (Except for the first time the calf began to feed)
Passive to feeding	Neither resistant to feeding, or actively looking to feed. (Except for the first time the calf began to feed)

4.4.2 Locomotor activity

The voluntary locomotor activity of the calves during the 30 minutes post treatment were accessed from videos by dividing the computer screen with a grid. Every time one hind claw crossed one line, it was counted as one activity. If the claw was put down in the middle of a line, it was not counted as an activity until the claw had crossed the line. Due to some different camera angles in the videos there were 6 grids used but all were divided into 50 equal sized areas. Many of them had the same exact measurement but some differed a little. Vertically the grids were divided into ten equal sized pieces, and in five equal sized pieces horizontally. This resulted in that the grids contained 50 equal sized areas.

4.4.3 Data analysis

Since the thesis data was unbalanced due to the low number of animals used, it was decided to show the data descriptively. The data was transferred into excel-sheets after video analysis, and then transferred into SAS and Minitab for basic descriptive data analysis and the data except birthweight, cross-sucking, self-grooming, play behaviour, help to start feed first bout and ease of feeding, were tested with the non-parametric test Kruskal-Wallis. Birthweight of the calves were tested with the Shapiro-Wilk W test for normality and analyzed with one-way ANOVA. Cross-sucking, self-grooming, play behaviour, help to start feed first bout and ease of feeding were measured in percentage occurrence for each treatment method.

5. Results

Distribution of the data was unbalanced between the different treatment methods but also between the different feedings. The distribution was unbalanced for example due to not as many born calves as anticipated, cows that did not have enough colostrum and technical difficulties with the video-recordings. During the colostrum feeding (1st feeding/treatment) there were 8 calves fed with a bottle, 6 were fed with an OT and 7 calves were fed with a bottle and then was moved back to the dam for suckling. During the 2nd feeding there was no data collection for calves from the suckling group (since they were still with the dam), so the suckling group had data for 3 feedings (including the colostrum feeding) whereas the bottle and OT groups had data for 4 feedings. Due to low numbers of calves used, females and males were considered equal for statistical analysis.

Birthweight of the calves were recorded and it was the only parameter measured during the thesis that was normally distributed. Calves from the bottle group weighed in average 43 ± 1.3 kilos at birth, calves from the OT group weighed 43.3 ± 2.5 and calves from the suckling group weighed in average 41.8 ± 2.2 kilos at birth.

5.1 Behaviours during colostrum feeding

Calves fed with an oesophageal tube during colostrum feeding required significantly less ‘attempts to feed’ than calves from both the bottle and the suckling group ($H=11.06$, $d.f.=2$, $p=0.004$), see figure 4. There was also a tendency ($p=0.07$) for calves fed with an OT to vocalize more during colostrum feeding, than the calves from the other two treatment method groups (table 4). There were no significant differences between the treatment methods in the number of ‘hind leg movements’ (table 4), ‘slips’ (figure 5), ‘falling down’ and ‘make calf stand’ (table 4).

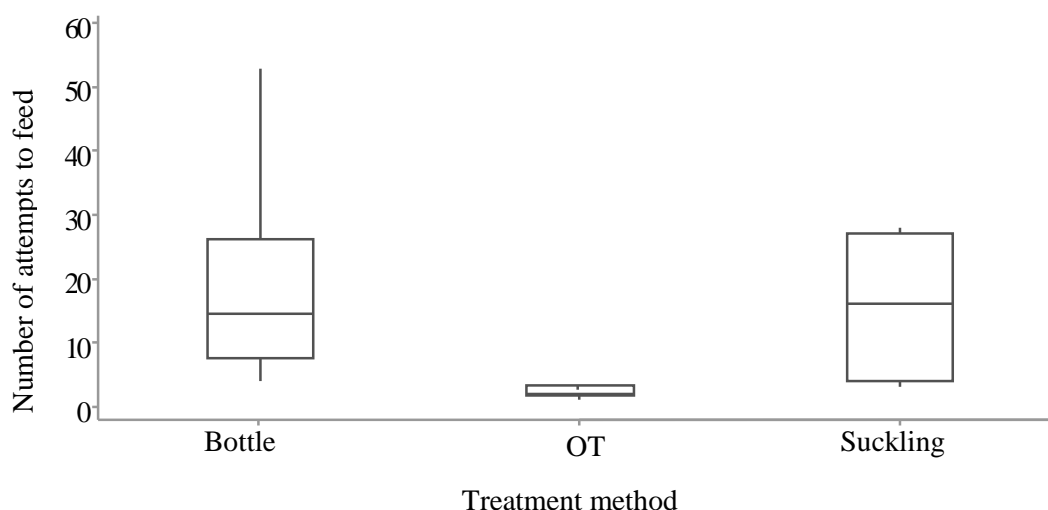
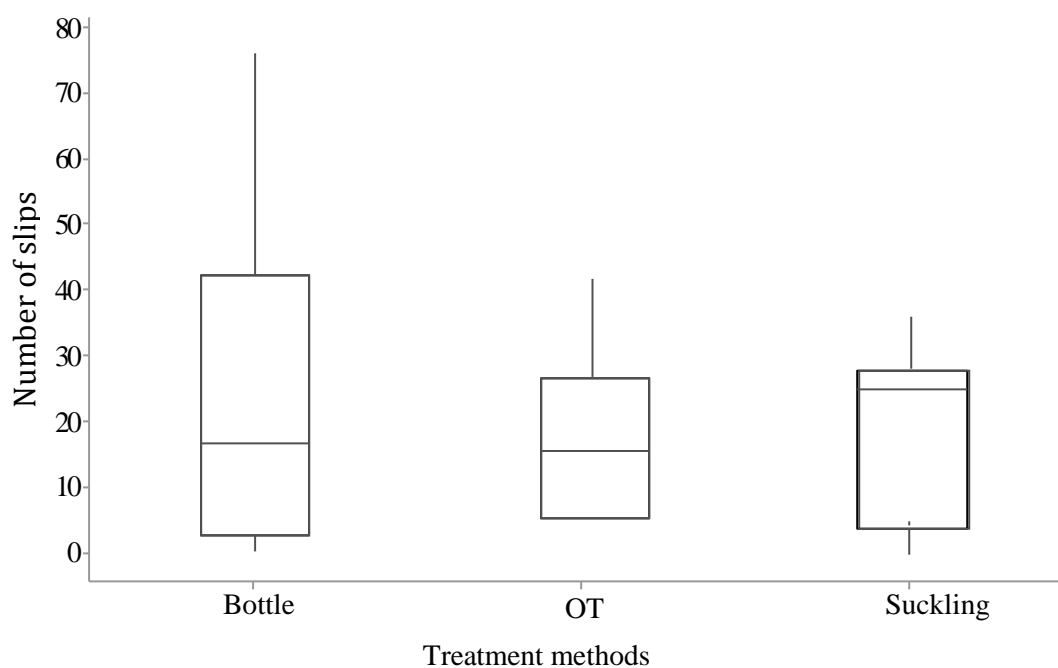


Figure 4: Median, Q1, Q3, min and max occurrence of attempts to feed in dairy calves during the colostrum feeding (bottle $n=8$, OT $n=6$ and suckling $n=7$).

Table 4: Behavioural response to different feeding methods during colostrum feeding in dairy calves

Behaviour	Treatment method	n	Mean	SE	Median	Q1	Q3	Min	Max
Hind leg movements	Bottle	8	85.88	78.17	47.5	23	164	13	205
	OT	6	59.67	14.24	60.5	54	66	37	80
	Suckling	7	124.71	89.4	108	49	165	43	43
Vocalizations	Bottle	8	0	0	0	0	0	0	0
	OT	6	1	1.26	0.5	0	2	0	3
	Suckling	7	0.29	0.76	0	0	0	0	2
Falling down	Bottle	8	0.5	1.07	0	0	0.5	0	3
	OT	6	0.67	0.82	0.5	0	1	0	2
	Suckling	7	0.14	0.38	0	0	0	0	1
Make the calf stand	Bottle	8	0.25	0.71	0	0	0	0	2
	OT	6	0.5	0.55	0.5	0	1	0	1
	Suckling	7	0.43	0.79	0	0	1	0	2

None of the calves showed the behaviour ‘lying down (voluntarily)’ and only 1 (suckling group) showed the behaviour ‘attempts to lie down’ 1 time. Only 3 calves in total displayed the behaviour ‘pushing forward’. In figure 4 the occurrence of the behaviour ‘slips’ compared between the different treatment methods is displayed and there were no significant differences between the groups.

**Figure 5:** Median, Q1, Q3, min and max occurrence of slips in dairy calves during the colostrum feeding (bottle n=8, OT n=6 and suckling n=7).

5.2 Behaviours after completed colostrum feeding up to 30 minutes after completed colostrum feeding

None of the behaviours during the following 30 minutes after colostrum feeding differed significantly between the treatment methods (table 5, figure 5-7). Calves that were fed with a bottle during the colostrum feeding did not display the behaviour ‘escape attempts’ but calves from the other two treatment methods did, though the difference between the treatment method groups were not significant (table 5). There were almost no differences between the treatment groups in regard to how many times they vocalized the first 30 minutes after first colostrum feeding. Even though the suckling group had the highest frequency of ‘vocalizations’ the differences were not significant (table 5).

In total there were 8 calves that did ‘attempt to lie down’ and 2 of them were fed with a bottle during the colostrum feeding and 2 with an OT. There were also 4 calves from the suckling group that did ‘attempt to lie down’. There were only 1 calf that showed the behaviour ‘attempts to stand up’ (1 time) and it was a calf from the suckling group. In total there were 5 calves that were ‘standing up’ and 4 of the calves that were ‘standing up’ were fed with a bottle during colostrum feeding and 1 with an OT. The individual variation of the behaviour only varied from 1-2 times.

Table 5: Behavioural response to different feeding methods, during colostrum feeding, 30 minutes after colostrum feeding in dairy calves

Behaviour	Treatment method	n	Mean	SE	Median	Q1	Q3	Min	Max
Escape attempts	Bottle	8	0	0	0	0	0	0	0
	OT	6	1.5	3.21	0	0	1	0	8
	Suckling	7	4.71	9.3	0	0	7	0	25
Vocalizations	Bottle	8	2.88	5.96	0	0	3	0	17
	OT	6	2	2.68	1	0	3	0	7
	Suckling	7	3.14	3.76	2	0	5	0	10

During the 30 minutes after colostrum feeding the ‘first time lying down’, ‘duration lying down’ and ‘activity’ had some individual variations, although the differences between the treatment methods were not significant. Calves from the bottle group lied down earlier (‘first time lying down’) than calves from the other groups and calves from the OT group lied down for the first time the latest, according to the median values (figure 6). However, the maximum values were the lowest for the OT group, which meant that there were calves from the other treatment groups that had lied down later for the first time than the calves from the OT group (figure 6).

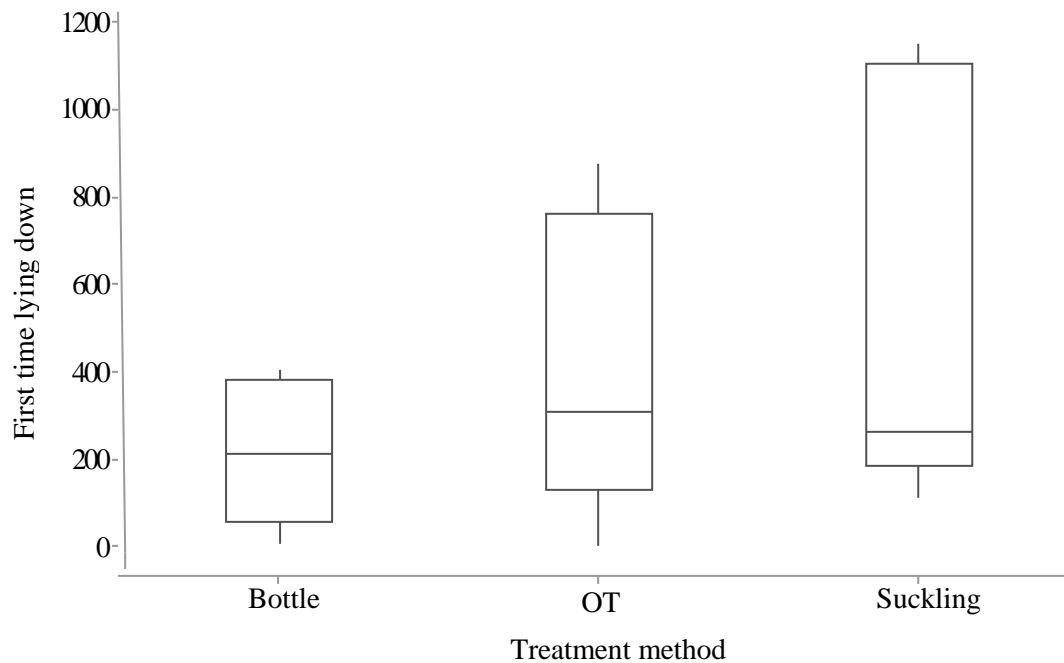


Figure 6: Median, Q1, Q3, min and max time for the first time dairy calves lied down 30 minutes after colostrum feeding, measured in seconds (bottle n=8, OT n=6 and suckling n=7).

In figure 7 the ‘duration lying down’ is displayed for each treatment method and the suckling group had the highest median average and the bottle group had the lowest, even if the OT group differed only very little from the bottle group. However, the difference from the highest to the lowest value was less than a minute. Furthermore, had the OT group the highest maximum value and the suckling group had the lowest (figure 7).

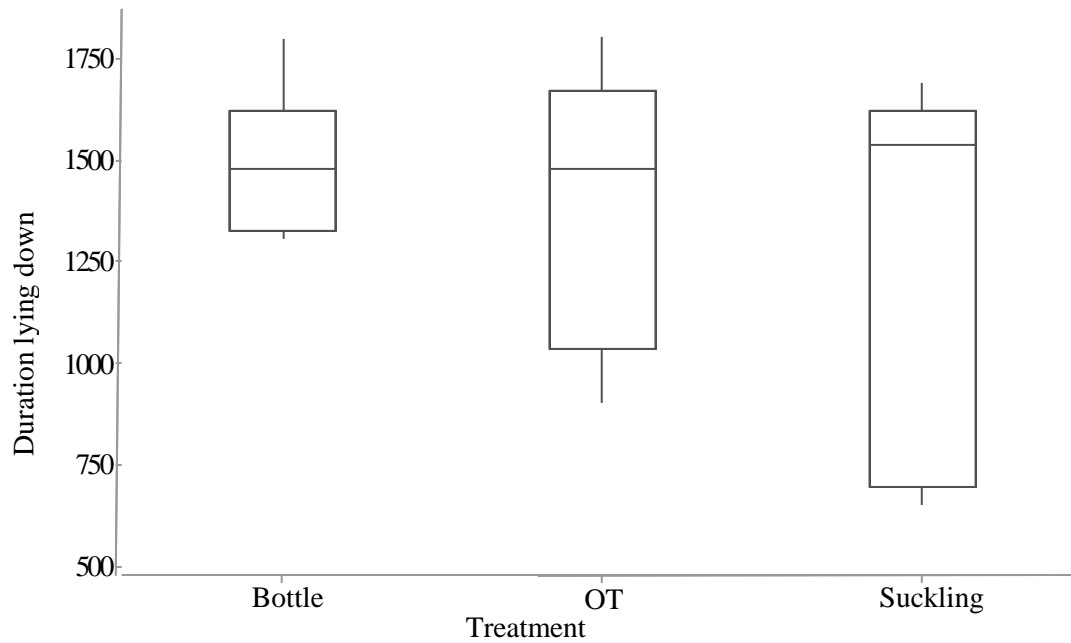


Figure 7: Median, Q1, Q3, min and max duration dairy calves spent lying down, measured in seconds, 30 minutes after colostrum feeding (bottle n=8, OT n=6 and suckling n=7).

Highest frequency for ‘activity’ had the calves from the bottle group and the calves from the suckling group had lowest median score, even though the suckling group had the highest recorded maximum score and the bottle group had the lowest maximum score. Calves from the suckling group had also the highest minimum score (figure 8).

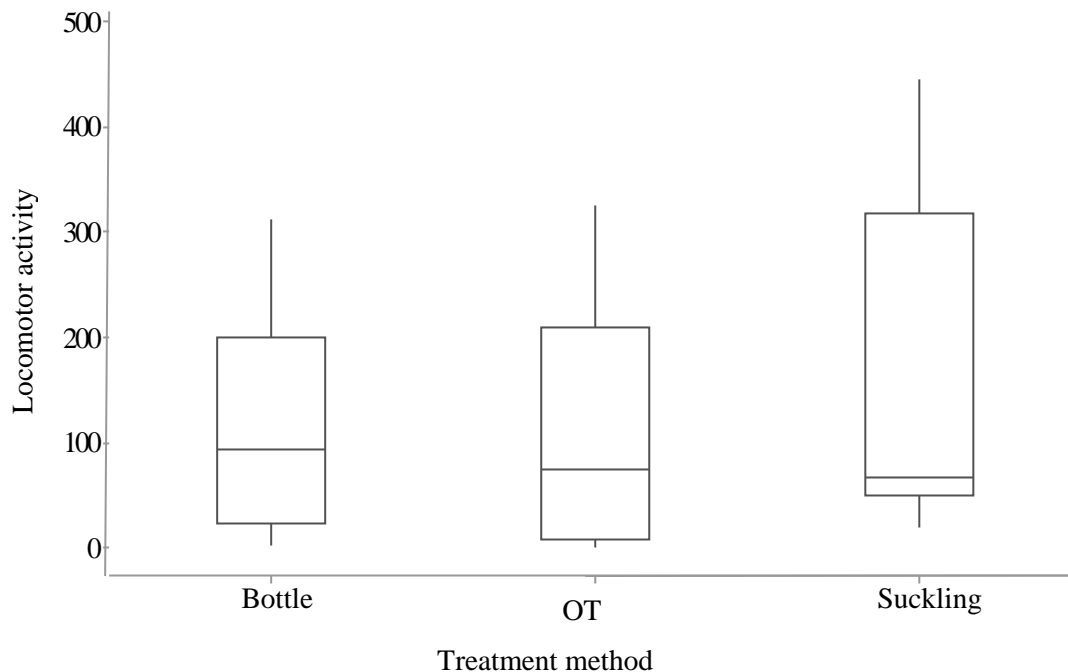


Figure 8: Median, Q1, Q3, min and max measured locomotor activity in dairy calves 30 minutes after colostrum feeding (bottle n=8, OT n=6 and suckling n=7).

5.2.1 Cross-sucking, self-grooming and play behaviour

There were quite high differences between the occurrences of cross-sucking for the different treatment methods during the 30 minutes after the colostrum feeding, the bottle group had an average occurrence of 50%, the OT group 33.33% and the suckling group 42.86%. Of the calves that did cross suckle 5 were heifers and 4 bulls. None of the calves in the OT treatment group showed the behaviour ‘self-grooming’, the bottle group had an average occurrence of 25% and the suckling group 14.29%. All the calves that were self-grooming were bull calves. None of the calves in the OT group or the suckling group displayed ‘play-behaviour’ but one calf in the bottle group did (average 12.50%).

5.3 Behaviours during 2nd to 4th feeding

5.3.1 Help to start feeding & cross-sucking

During the 2nd to the 4th feeding ‘help to start feeding- first bout’ and ‘cross-sucking’ were recorded for each feeding and compared between the different treatment methods and during the 3rd and 4th feeding there were differences between the treatment methods (table 6). During the 2nd feeding data were not collected for the suckling group since those calves were with their dam. Under the same feeding (2nd) the recordings did not differ between calves that were fed with a bottle or an OT during the colostrum feeding (table 6).

Table 6: Percentage occurrence of ‘help to start feeding-first bout’ and cross-sucking during the 2nd-4th feeding in dairy calves compared between different colostrum feeding methods

Behaviour	Treatment method	2 nd feeding	3 rd feeding	4 th feeding
Help to start feeding	Bottle	100	67	50
	OT	100	100	80
	Suckling	N/A [⌘]	75	50
Cross-sucking	Bottle	50	50	50
	OT	50	25	80
	Suckling	N/A [⌘]	75	0

[⌘]Not applicable

5.3.2 Ease of feeding

Ease of feeding from the 2nd – 4th feeding is displayed in figure 9, for the different colostrum feeding methods. There were not recorded ease of feeding for the suckling group during the 2nd feeding and for the other two groups the ease of feedings during the 2nd feeding differed quite much. During the 3rd feeding there were not a difference between calves from the bottle group or the suckling group, however calves from these two groups displayed much more ‘actively looking to feed’ then calves from the OT group. There were not as much differences in ease of feeding during the 4th feeding between the treatment methods, compared to earlier feedings (figure 9).

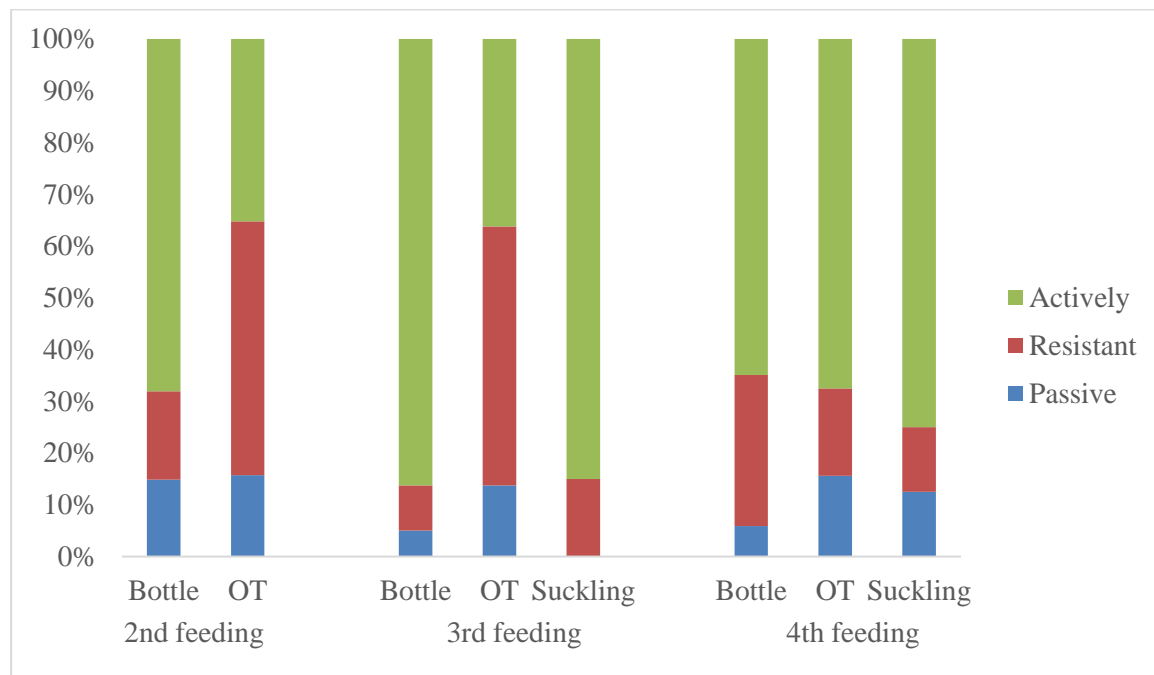


Figure 9: Ease of feeding shown in percentage of recorded behaviour in regard to number of feeding bouts during the 2nd-4th feeding for the three different feeding methods. The calves could either be actively trying to feed (actively), be resistant to feeding (resistant) or passive to feeding (passive). During the 2nd feeding there were not collected any data for the suckling group, since those calves were still with their dam.

5.4 Overview of colostrum feeding to 4th feeding

5.4.1 Time spent on feeding

It took significantly ($p=0.05$) less time to feed calves from the OT group during the first colostrum feeding compared to the other groups (table 7). However, it took significantly ($p=0.05$) less time to feed calves from the bottle group compared to the OT group during the 2nd feeding (table 7). The differences were not significant for the 3rd or 4th feeding (table 7). When comparing the ‘time spent on feedings’ from the colostrum feeding (1st feeding/treatment) to the 4th feeding, the average time spent on the feedings in total for the bottle group were 1588 ± 369 seconds, for the OT group 1661 ± 361 seconds and for the suckling group 1117 ± 208 seconds. For the 4 first feedings this showed a difference between the OT- (highest average) and suckling group (lowest average) of 9 minutes and 4 seconds spent on feedings.

Table 7: How much time (seconds) that was spent on each feeding, from 1st feeding (colostrum feeding/treatment) to the 4th feeding

		Treatment method	n	Mean	SE	Median	Q1	Q3	Min	Max
Time spent on feeding (sec)	1 st feeding *	Bottle	8	757	133	603	452	1058	387	1428
		OT	6	370.8	49.7	341	286.8	449.5	247	595
		Suckling	7	738	133	843	281	956	260	1190
	2 nd feeding *	Bottle	4	336	173	117	0	814	0	1211
		OT	4	530	180	626	0	919	0	1053
		Suckling	N/A \square	N/A \square	N/A \square	N/A \square	N/A \square	N/A \square	N/A \square	N/A \square
	3 rd feeding	Bottle	6	360	118	422	0	581	0	888
		OT	4	384	131	463	0	670	0	731
		Suckling	4	178.3	71.1	178	0	321	0	474
	4 th feeding	Bottle	6	255.5	84.6	266.5	0	499.3	0	540
		OT	5	376.5	93.3	415	215.3	507.5	0	698
		Suckling	4	201.3	79.6	189	0	363	0	515

\square Not applicable

* $p=0.05$

5.4.2 Amount fed

There were no statistically significant differences between the treatment methods and feedings in amount fed (table 8). During the colostrum feeding the amount fed was highest in the OT group (table 8), however the amount fed was correlated to the calf's birthweight during the colostrum feeding. The total amount of colostrum/milk fed for the first 4 feedings was not significantly different between the treatment methods (bottle: 7496±1302 grams versus OT: 7021±754 grams versus suckling: 6391±1150 grams). However, the amount that the calves suckled from their dam was not accounted for. If comparing the amount fed per feeding from colostrum feeding (1st feeding/treatment) to the 4th feeding, the OT treatment group fed the least, 1755.3 grams/feeding, the bottle group fed 1874 grams/feeding and the suckling group fed 2130.33 grams/feeding.

Table 8: Amount fed to dairy calves during the 1st feeding (colostrum feeding/treatment) to the 4th feeding, for each feeding

		Treatment method	n	Mean	SE	Median	Q1	Q3	Min	Max
Amount fed (g)	1 st feeding	Bottle	8	3318	185	3426	2761	3601	2583	4150
		OT	6	3610	231	3604	3173	3998	2835	4500
		Suckling	7	3138	362	3500	2547	3793	1376	4198
	2 nd feeding	Bottle	4	956	421	481	0	2093	0	3009
		OT	4	1238	496	1302	0	2374	0	2487
		Suckling	N/A [⌘]	N/A [⌘]	N/A [⌘]	N/A [⌘]	N/A [⌘]	N/A [⌘]	N/A [⌘]	N/A [⌘]
	3 rd feeding	Bottle	6	1652	502	2126	0	2992	0	3007
		OT	4	471	239	212	0	1128	0	1345
		Suckling	4	1647	586	2516	0	3003	0	3009
	4 th feeding	Bottle	6	1570	548	1758	0	3015	0	3021
		OT	5	1703	503	1848	486	2905	0	3000
		Suckling	4	1606	577	2232	0	3001	0	3006

[⌘]Not applicable

6. Discussion

The aim of this thesis was to evaluate the effects of 3 different colostrum-feeding methods (bottle versus oesophageal tube versus suckling) on the behavioural response during the first 4 feedings after parturition in dairy calves. During the first feeding, there were no differences in the calves' behavioral response to the method of colostrum feeding, except for a tendency for calves fed with an OT to vocalized more and that OT-fed calves required significantly less attempts to feed than bottle-fed calves. Feeding colostrum during the first feeding with an OT was faster than using a bottle. However, during the next 3 feedings calves that was fed with an OT during their first feeding appeared more resistant to feeding and it took longer time to feed during the first 4 feedings. In addition, OT calves fed less in average during the first 4 feedings, even though the differences were not significant.

Swedish farms have traditionally used bottle-feeding to feed their calves colostrum, however there has been an increased interest of using an oesophageal tube as colostrum feeding routine instead (Persson Waller *et al.*, 2013). There is a discussion about how and why the oesophageal tube would be used as a routine and some would argue that the time spent on feeding is one big part of why the usage of an oesophageal tube could be attractive (Persson Waller *et al.*, 2013). Many would agree that time is equal to money and not the least for the farmers today were many already struggles financially (LRF Konsult, 2016) and every bit of money counts. Although, the question is also that *if* the choice of feeding method used affects the wellbeing and welfare of the calves, is the time more important? Maybe there is a line when the time is more important? And in that case, how much time would be more important than the welfare?

The results of this thesis showed that it took less time in average spent on feeding during the first 4 feedings when first supplying the calf with colostrum from a bottle and then letting it suckle the dam, than by feeding it colostrum through an OT, the difference were a little more than 9 minutes. It took also less time to feed the calves from the bottle group than the OT group during the first 4 feedings. That the calves from the suckling group fed faster during the 3rd-4th feeding could both be due to that they had learned to suckle after being with the dam or that they were hungry after being with the dam. However, according to Herskin *et al.* (2010) drinking rate were not related to level of hunger. The general labor cost for animal production in Sweden is around 232 SEK/hour (SLA, 2016) and then 1 minute would be worth 3.87 SEK. This means that 9 minutes would be equal to 34.83 SEK, which are the money that would be saved per calf during the first 2 days after parturition if letting the calf suckle the dam instead of feeding it with an OT during the first colostrum feeding.

A problem when comparing and discussing this thesis results is the unbalanced data. There was not only a difference in how many calves that had the different treatments, but also for how many feedings/calf data were collected. An added complication was the high individual variation in some of the behaviours and other parameters observed. With so few calves included in this study one should be careful when interpreting the findings presented in this thesis. The results could also possibly been affected by the calves sex or breed, even though these parameters were not considered during the statistical analysis in this thesis, due to the uneven distribution of the data. According to Lidfors and Jensen (1988) heifer calves are more active after birth compared to bull calves and that they also has a longer suckling time. Dufty (1973) showed that heifer calves generally has a lower birthweight than bull calves, this could possibly be connected to higher activity level. In 2013 2.6% of SRB calves in Sweden died within a month after parturition and 2.9% of SLB calves died (Jordbruksverket, 2014b). Even if the total mortality rate did not differ so much, it did differ more between the bull

calves, 2.8% (SRB) versus 3.4% (SLB). It did not differ between the heifers though (2.5%) (Jordbruksverket, 2014b). Health and mortality rate could possibly affect the behaviours.

6.1 Feeding with an oesophageal tube or a bottle

There were a significant difference in 'attempts to feed' between treatment methods and the highest frequency had the suckling group at 16 times and the lowest had the OT group at 2 times. Although, it is important to have in mind that the OT group could not have very frequency (number of times that the behaviour occurred) in this category, since it was decided that it would put too much distress on the calves if a feeding would proceed after four feeding attempts with that feeding routine. It was decided in the later part of the project that it was only allowed to try to feed with the oesophageal tube twice and then the feeding was terminated. This made that the variations were difficult to draw conclusions from. One possibility could although be that the person who is feeding the calves would perceive it easier to feed when not having so many feeding attempts. Maybe even arguing that if a calf has feeding attempts of almost 20 times it would be forced feeding as is argued that feeding with an oesophageal tube is (Lorenz *et al.* 2011; EG: 20.5 889/2008, 2016). And since it was thought that more than 2 times for feeding with an OT would be too many times, how many times is too many for when feeding with a bottle? It should be noted that the calves that had more than 2 'attempts to feed' when being fed with an OT in this thesis, were only bull calves (2) and those calves were participating in another study as well were is was allowed to intubate them more than twice.

A difference in average was also found in 'hind leg movements', with the suckling group that had the highest average and the OT group had the lowest and the difference were approximately 65 times. One thing that seemed possible to be related to number of 'hind leg movements' were the time spent on the treatment and that seems quite logical that the more time spent on the feeding there is a longer time for the calf to express a behaviour. Even if there were many 'hind leg movements' recorded, it was not a behaviour that was easy to evaluate. From the beginning the behaviour was thought to reflect if the calves were resistant to feeding or not, but as the analysis started it became clear that no such conclusion could be made, that more 'hind leg movements' would be equal to a more resistant calf. This was clear since calves sometimes were just standing and moving their legs up and down without showing any discomfort and also could the person feeding affect the calf with for example by moving the calf, and by doing so it would bring the frequency of 'hind leg movements' up. So unfortunately, this behaviour did not really give much information about the differences between the feeding methods. There were no big variation between the feeding methods in regard to the behaviour 'slips', which could have been assumed that it would be affected by if the calves were resistant to feeding or not, but no such connection were found. There were 50% of the calves from the OT group that fell down during the colostrum feeding, although in average it did not differ much between treatments. But when calves fell down due to slipping it seemed to be related to how resistant they were to feeding, since those calves were very resistant to feeding. To my knowledge there have not been earlier studies analyzing behaviours in regard to feeding when taking 'hind leg movements' and 'slips' into account, although with the results from this thesis other behaviours measured were more interesting due to the fact that conclusions could be drawn from them.

Regarding vocalizations, there were some variations. During the colostrum feeding 3 of the 4 calves that vocalized belonged to the OT group, but during the 30 minutes after the colostrum feeding there were more calves expressing the behaviour. Kiley-Worthington and de la Plain (1983) studied vocalizations in young calves and believed that vocalizing could be a sign of discomfort, but they found it hard to find a precise conclusion to the behaviour. At least they concluded that new-born calves do vocalize rarely. Thomas *et al.* (2001) on the other had concluded that calves usually vocalize due to hunger. If a conclusion could be drawn from this, it was that calves fed with an oesophageal tube during the treatment could have expressed discomfort, which is not good. During the 30 minutes after the colostrum feeding the calves could have been hungry, although it seems unlikely due to the high amount fed, or also showing discomfort, which is of course not good either way. There is also a possibility that the calves reacted on other animals that were quite close by them and were vocalizing, the calves might have only have answered the other animals.

Calves from the bottle group lied down earlier compared to the OT group, and so did the calves from the suckling group. Calves from the suckling group lied down during a longer duration, compared to both the other groups. Even if calves from the OT group lied down during a longer duration than calves from the bottle group, the difference were only 1 second in average. According to Eicher and Dailey (2002) behavioural responses to acute pain were increased moving and decreased lying. Which is somewhat agreeing with Enríques *et al.* (2010) conclusion that pacing could be a sign of distress, but so could also vocalization and sudden stop to play behaviour according to Enríques *et al.* (2010). However, according to Stull and Reynolds (2008) loss or less mobility could be an indicator of acute pain. So depending on which study to rely on, the results could be interpret differently, although the difference between the treatment method groups were not significant so a conclusion can not be drawn of the differences between the groups. However, the calves need to rest but it may be more interesting to analyze how quickly after the calf was fed (for example when the rubber nipple of the bottle or the oesophageal tube is taken out of the calf's mouth) until they lay down than the duration they lay down during the 30 minutes after first colostrum feeding. The behaviour 'first time lying down' is close to account for this, but since the person feeding the calves had to get out of the pen and leave the calf alone, there were a few moments delay until next video-recording started and thus also when the behaviour 'first time lying down' were recorded. There were only few signs of behaviours that are thought to promote good welfare in this thesis, one calf from the bottle group displayed for example play behaviour and play behaviour is thought to promote good welfare (Stull and Reynolds, 2008). There might have been to few observations of behaviours of that is thought to be a sign of good welfare in this thesis to say if either of the different treatments methods gives better or worse welfare. However, it might have been too early for the calves to express for example play behaviour when they were only a couple of hours old. Although, the ease of feeding could be argued that it shows an indication of better or worse welfare, for example if the calves were very resistant to feeding, it would be difficult to argue that it would be good welfare.

Occurrence of cross-sucking differed between the treatment method groups where the calves from the OT group had a highest average occurrence during the 4th feeding, even if they did not have a higher average during the 30 minutes after the colostrum feeding or the 2nd and 3rd feeding. During the 3rd feeding calves from the OT group had the lowest average of occurrence and the suckling group the highest. Jensen (2003) defined cross-sucking as a detrimental behaviour and that it is a form of non-nutritive suckling. Jensen (2003) also stated that cross-sucking is only observed by calves that are artificially reared, although the calves that could suckle in this thesis cross-sucked also. Even though, this was observed after the

calves had been moved from the dam, so it could not be ruled out that the behaviour only occurred in those calves since they were moved away from the dam, even if they could suckle for some time before being reared artificially. This thesis results however agreed with both Krohn *et al.* (1999) and Fröberg *et al.* (2008) results, which were that calves that could suckle displayed lower average of cross-sucking. Fröberg and Lidfors (2009) concluded both that calves that could suckle freely cross-sucked less and that they rested more. Occurrence of cross-sucking is negative and could indicate that calves fed with an oesophageal tube during this thesis might not have fulfilled their sucking needs during the 4th feeding. It could also show that the calves fed with a bottle might not have fulfilled their needs during the colostrum feeding or that the calves from the suckling group had not during the 3rd feeding. Herskin *et al.* (2010) came to the conclusion that the occurrence of cross-sucking were correlated to level of hunger, that hungry calves cross-suck more. If this would be the explanation to why the calves cross-sucked during this study, it would suggest that by providing the calves with more milk the cross-sucking would have decreased or been eliminated. However, even if it might have been a factor in some calves, it seems unlikely that this would be to only explanation due to the large amounts fed.

When evaluating the behaviours from the 2nd-4th feeding, calves from the OT group had higher percentage of 'resistant to feeding' behaviours, than calves fed with a bottle. And calves fed with a bottle during the colostrum feeding had a higher percentage of 'actively trying to feed' behaviours, than calves fed with an OT. Calves from the suckling group had the highest average of 'actively trying to feed' (90 %) though and the lowest of 'resistant to feeding' (2 %), during the 3rd to the 4th feeding. This results agreed with Metz (1987) results that calves that can suckle the dam has not more difficulties to learn how to feed from a bucket, than calves that are separated from the dam shortly after parturition and reared artificially. This thesis showed that it was more difficult to feed calves during the 2nd-4th feeding if they were fed with an OT during the colostrum feeding compared being fed with a bottle. This results agreed with Persson Waller *et al.* (2013) conclusion that it is likely that calves fed with an OT is resistant to feeding the next feeding. And with this in mind it could be argued that feeding with an OT actually is worse both for the calf, since it so resistant to feed the following feedings, but also for the person trying to feed the calf, since it takes longer time to feed and the calf struggles more.

One aspect that could be argued to be positive in regard to feeding with an oesophageal tube is that it could be easier to control that the calves actually has been feeding (Persson Waller *et al.*, 2013), which could make the occurrence of FPT lower. During this thesis even though calves that were fed with an OT during the colostrum feeding, fed a larger amount during the colostrum feeding, they fed less in average during the 2nd-4th feeding, compared to calves fed with a bottle during the colostrum feeding. Birthweights of the calves could have had an impact of the results during the colostrum feeding, since how much they were fed were correlated to their birthweight. Laestander (2016) came to the conclusion that using on OT on calves that do not feed voluntarily could be a good way to ensure that they have fed a sufficient amount of colostrum. Although, if the calves are healthy and are motivated to feed, feeding with an OT did not gain any advantages compared to bottle-feeding or suckling when taking health, growth and transfer of passive immunity into account (Laestander, 2016).

Several studies concluded that calves that suckled had higher occurrence of FPT (Besser, 1991; McGuirk and Collins, 2004; Godden *et al.*, 2008). Quigley *et al.* (1995) results showed on the other hand that calves that could suckle had higher concentrations of IgG than calves fed by a bottle. It would have been interesting to measure occurrence of FPT in all of the calves used in this thesis and compare between the different feeding methods and evaluate if FPT and behaviours in regard to feeding could have any relation. For example that when measuring 'ease of feeding' during the 2nd-4th feeding also measure FPT and see if the calves that were more resistant to feeding had higher occurrence of FPT or not. Laestander (2016) measured serum IgG levels in some of the calves used in this thesis, and also in some more calves and all the calves used in Laestander (2016) thesis were managed the same way as the calves in this thesis and same treatment methods were used. Laestander (2016) results showed that there were no significant differences in serum IgG levels between the different treatment methods and neither were differences in the calves' health found. These findings could strengthen the argument for not feeding healthy calves with an OT during the first colostrum feeding, since added with the other disadvantages found with the routine, it does not supply a higher serum IgG level.

7. Conclusions

The results of this study suggest that there are no clear advantages with using an oesophageal tube during the first colostrum feeding when considering the ease of feeding, time spent on feeding and amount fed during the first 4 feedings. There are many risk factors associated with using an OT, such as feeding the calves too much and too fast, getting fluid in the lungs, hurting the mouth, throat and esophagus, the calves also gets more resistant to feeding the following feedings, in average it do not save time during the first 4 feedings, the amount fed in average during the first 4 feedings is lower and due to all of theses factors it could be a risk for worse welfare for the calves. Feeding with an oesophageal tube could have advantages as well, if used correctly, but it could also promote a false security. It could promote a false security since even that it could be easier to control the amount fed it is still important to feed at the right time (shortly after parturition) and that the colostrum is of good quality. It is not enough to only make sure to feed a sufficient volume of colostrum. The results from this thesis should be interpreted with caution due to the low number of calves included. Further research is needed and there should also breed and sex be considered.

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Appendix 1:

List of routines during the 2nd to 4th feeding

- Change into stable clothes, boots and wash your hands.
- Put on gloves.
- Go and put on the scale in the tiestall.
- Collect protocol, pen and timekeeper in the tiestall and put it on the table by the scale.
- Go and get the thermometer in the room with the video-equipment (second door to the left from the door where you collect the boots, in direction to the tiestall). Also see that the video-recording is up and running.
- Go to the room with the milk, see if the staff is there, otherwise go and find the person who is working and ask if it is okay to start with the feeding.
If "yes" →
- Check the temperature on the milk (should be about 40°C, but at least range 38-41°C).
- Bring the milk (in the bucket it was milked in), one nipple-bucket to feed the calf in and a pitcher to measure the milk in and go to the tiestall.
- Put the empty pitcher on the scale and tare it.
- Pour the milk in the pitcher (3 liters) and weigh it so it is the right amount (range 3000-3010g).
- Write up the amount milk fed in the protocols, both in the stable protocol and in the one for the behaviours.
- Pour the milk in the nipple-bucket it should be fed in.
- Be ready with protocol and timekeeper.
- The staff could start to feed the calf.
- Start to measure time when the calf has the rubber teat in its mouth.
- Stop the time when the milk is finished or when the calf does not want to feed any more. (At least try to give the calf milk for 15 minutes. The staff can go into the pen and help the calf if it needs help to suck on the rubber teat. But if the calf does not need help to feed, the calf should be left in peace to feed. Do not walk away from the calf, but stay and observe if the situation changes. The calf should not be held stuck at the feeding. The maximum time of feeding the calf is 30 minutes, then stop the time and weigh the amount leftovers of the milk). If the calf refuses to suck the teat bucket during the feeding, the staff is allowed to feed it in a bottle or in a tube if necessary, but first *after* the data collection is done, if they think it is the best option for the calf. If this would occur, make a note in the protocol.
- When putting the teat bucket on the gate to the pen give the calf at least one minute to get started to feed by itself. (When feeding session is starting, not during feeding).
- If the calf does not feed by itself, help it to find the rubber teat, don't just stand and look at it for several minutes.
- After the calf has finished the feeding be sure to fill in everything in the protocols and clean up after, turn off the scale and wash the equipment used. BUT if the staff offers to do the dishes the person who is observing should at least wash the thermometer and put it back in the video-equipment room. Put off the lights in the video-equipment room.

- NOTE! Do not forget to ask the staff if the milk sample from the dam is taken. If it is not taken, take the sample. (From the dam/dams used in the project).
- Wash your boots and hands on your way out. Throw away your gloves.

Some repetition and things that is good to know

- If the calf is very resistant against feeding and you have tried to feed it for 15 minutes, end the feeding session. Ask the staff if they think the calf will feed more, otherwise stop the feeding. If there is a calf that is very much resistant (this is calves of exception) you can even stop the feeding before 15 minutes. This is for example a calf that you have not been able to get to feed anything and it is so resistant and struggling so much that it is falling down. Talk with the staff. *You can never and should never try to force the calves to feed.*
- If the calf has not been feeding during the data collection, the staff is allowed to feed it with a bottle or a tube, after the data collection is done, if they think it is the best solution. But in that case, remember to make a note.
- The staff is allowed to go in the pen to the calves during feeding.
- The staff should try to go through with the feeding as they usually do. But if the calf has not started to feed by itself or has a long break after it has let go of the rubber teat (after ≈ 30 second) after it has started to feed, they should help the calf to suck again, even if this would differ from the normal routine for the staff. This is since it needs to be as similar as possible for all the calves in the study, otherwise the time it takes to feed would be misleading.