Optimal cleaning of equipment

Effectiveness of optimised teat cup cleaning in the prevention of mastitis pathogens transfer

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Effectiveness of optimised teat cup cleaning in the prevention of mastitis pathogens transfer

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Abstract

Flushing of the teatcup liners between the milkings of two cows is a common procedure in AM-systems.
In this investigation the standard procedure, flushing with cold water, is compared with flushing with a disinfecting fluid, composed of cold water and per acetic acid. Liners were contaminated with a suspension of *Streptococcus agalactiae* in milk, comparable with a severe inflammation of the udder. The log reduction in number of pathogens was 1,80 and 2,26 for respectively cold water and the disinfectant. The extra log reduction with a disinfectant was small and did not compensate for the negative effects of using disinfectants in a milking system (extra rinse necessary, higher risk of contamination of milk).
Further experiments to show the effect of cluster flush on transfer of pathogens through liners are done with a cold water rinse only.

46 Cows were challenged with pathogens by milking with deliberately contaminated liners with a high dose of *Streptococcus agalactiae* prior to the cluster flush. Two liners (cross positioned front and rear) were flushed after contamination, the other two liners were not flushed. The udder health of the cows was measured during one week after the challenge. In none of the foremilk samples *Streptococcus agalactiae* could be recovered and none of quarters showed signs of an inflammation (increase in SCC or conductivity). The effect of a cluster flush on the rate of new infections could not be established in this experiment.

However, based on the results of the reduction of pathogens in liners, it is recommended to perform a cluster flush to reduce the number of pathogens in the liner.

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1. Introduction

Mastitis is an important disease in animal production. Large numbers of cows are infected each year, each infected cow is a potential source for spreading the pathogen. The milking machine is known as a possible vector for spreading of pathogens (Dodd et al., 1963, Miltenburg et al., 1997), especially the udder associated pathogens. The milking process offers multiple opportunities for bacteria to be transferred between cows and quarters. After a infected cow has been milked, the liner surfaces carry bacteria originating from the teat surface and milk of that cow. These bacteria are transferred to the next cow. In traditional milking parlours it is advised to milk infected or suspected cows last, just before the cleaning of the installation. In automatic milking systems it is hardly possible to milk cows in an order corresponding to their udder health status. Besides that, not all infected cows are known on forehand because new infections will occur and some minor infections will not be recognised. This is true for traditional and automatic milking systems as well. To minimise the role of the milking machine as a vector, the teatcups can be cleaned after each milking by flushing with water. In this way most of the pathogens, present in the liner after milking an infected cow, can be removed. The effect of the cluster flush in AM-systems on new infection rate however is unknown.
It should be mentioned that mastitis infections with environmental bacteria will occur between milking times. The infection rate with these type of bacteria is not likely to be influenced by a cluster flush.

In this investigation the effect of a cluster flush on the removal of pathogens is studied. In a challenge test the effect of a cluster flush on the rate of new infections will be tested .

2. Material and Methods

The effect of the cluster flush is measured as the removal of pathogens from an contaminated liner and the risk on new infections when milking with contaminated liners, comparing flushed liners with liners which are not flushed.

2.1 Removal of pathogens

The removal of pathogens is tested on an Automatic Milking System of Gascoigne Melotte, installed on the Waiboerhoeve in Lelystad, The Netherlands. Before the test run, a system cleaning of the AMS was performed.

Streptococcus agalactiae was chosen as the challenge pathogen in this experiment. Although *Streptococcus agalactiae* can live outside the udder for short periods of time in the right conditions, it is considered to be an obligate pathogen of the udder. This pathogen was not found in the experimental dairy herd.

Each experiment consisted of 6 runs, in the first three runs the cluster was flushed with cold water, the last three runs a disinfectant was used (2 % peracetic acid solution).
In each run the liners were contaminated with a *Streptococcus agalactiae* suspension in pasteurised milk (approx. 1,5 10^6 cfu/ml). The pathogen was derived from a severe clinical mastitis infection. 10 ml of this suspension is flooded at the top end of the barrel of the liner in one circular movement, to provide an equal film of contaminated milk over the entire interior of the liner. Each time after contamination two cross positioned liners were flushed in the standard way of the AMS, the other two liners were not flushed.
The number of pathogens in the liner is measured by swabbing a part of the interior surface of the liner, 1 to 2 minutes after flushing the liners. Swabs are taken from the liners by making two circular movements with the swab, approx. 10 cm below the liner mouth piece. After sampling the swabs were moistened with a peptone-buffer -, to inactivate the disinfectant when present. The swabs are analysed for number of *Streptococcus agalactiae* by the Dutch Animal Health Service.
At the end of each run the cluster was disinfected and flushed with cold water.

This procedure was repeated once.

2.2 New infection rate

The effect of a unit flush on the rate of new infections was tested on an Automatic Milking System of Gascoigne Melotte, installed on the Waiboerhoeve in Lelystad, The Netherlands. This system used quarter take off and no post-milking teat disinfection.

The cows used for the experiments were accustomed to automatic milking. Cows were selected from a herd of about 50 cows on the basis of good udder health, established by SCC, conductivity and bacteriological examination of foremilk samples. 10 to 12 cows were used in a each challenge test. These cows were separated from the rest of the herd in a part of the cubical housing system. Twice a day this group of 10 to 12 cows was brought to the AMS to be milked. See table 1 for parity, days in milk and milk production of the cows used in the experiment.
In the first run cows were selected based on SCC and pathogens in foremilk samples, collected in the cubicle housing system during the day. Due to the voluntary milking system, the time lap between milking and sampling differed largely. This effected in a large extend SCC, which was very often raised compared to values in samples taken during milking for recording milk production. In the second, third and forth run cows were selected on the SCC in samples taken for milk recording. In the evening milking prior to the challenge test, udder health was checked based on fore milk samples. Cows were selected when the SCC was below 200 * 10^3, the SCC in none of the quarter milk samples exceeded 250 * 10^3 and no major pathogens were found in fore milk samples.

Streptococcus agalactiae was used in this experiment as the infecting pathogen. Normal *S. agalactiae* infection most often occurs during milking and is passed from cow to cow on contaminated milking equipment, hands and common use towels used between cows in pre-milking udder preparation. This pathogen was not found in the experimental herd as a mastitis pathogen and mastitis due to *Streptocuccus agalactiae* is very responsive to intra-mammary antibiotic therapy.

The contamination of the liners was done during one evening milking (day 0). All the liners of the AMS were contaminated with a *Streptococcus agalactiae* suspension in pasteurised milk before milking the next cow. 10 ml of the suspension was flooded at the top end of the barrel of the liner in one circular movement, to provide an equal film of contaminated milk over the entire interior of the liner. Two cross positioned liners were flushed in the standard way of the AMS, the other two liners were not flushed. The cluster was attached by hand to be sure that attachment was conducted without delay and retrials. Teat cleaning, as it is usually performed in this AMS, was inactivated, to prevent a flushing of the liners and teats at the start of milking and so prevent removal of pathogens. The air bleed in the short milk tube was blocked during the milking session with contaminated liners, to create sub optimal milking and circumstances which will lead to a higher risk on infection.
The cows in the experiment were milked in the AMS twice a day, from two days before the start of the contamination of the liners until 8 days afterwards. During the evening milking of day –1, 1, 2, 3, 5 and 7 foremilk samples of all quarters of the cows were taken and analysed for SCC and pathogens by the Dutch Animal Health Service. Conductivity measured during milking was recorded by the AMS. Foremilk samples of the day before contamination of the liners (day –1) were used to have a final check on the cows for udder health.

Because the effect of this type of challenging cows with pathogens is not known, it was decided to have a go-no-go moment after 50 and after 100 challenged cows.

Table 1: Parity and days in milk (DIM) of cows used in the 4 runs

Run	Cow	Parity	DIM	Milk yield (kg/day)
1	1302	4	146	37.3
1	1783	2	233	24.2
1	1907	2	142	27.8
1	2068	1	196	32.5
1	2126	1	54	30
1	2164	1	227	20.5
1	2191	1	259	25
1	2207	1	193	33.7
1	2216	1	208	27.2
1	2406	1	42	30.6
2	1302	4	172	35.8
2	1783	2	259	22.4
2	1907	2	168	23.5
2	2068	1	222	32.6
2	2126	1	80	31.3
2	2139	1	67	24.8
2	2164	1	253	20
2	2207	1	219	30.3
2	2216	1	7	30.1
2	2380	1	50	34.4
2	2406	1	68	32.3
2	2407	1	55	30.1
3	719	5	292	30.1
3	975	3	450	26
3	1146	1	86	28.3
3	1302	4	228	27.9
3	1425	4	95	37.3
3	1679	3	106	30.6
3	1691	2	276	25.8
3	1757	3	108	31.8
3	1794	2	231	34
3	2068	1	278	31.4
3	2199	1	320	28.5
3	2216	1	57	24.6
4	102	8	86	40
4	397	6	83	41.1
4	423	6	79	34.5
4	686	6	265	36.4
4	719	5	418	27.6
4	2169	2	48	46.7
4	2171	2	95	35.3
4	2184	2	83	38.7
4	2360	3	203	25.1
4	2459	1	102	28.7
4	2473	1	61	23.6
4	2680	1	47	23.7

3. Results

3.1 Removal of pathogens

The number of pathogens in the suspension to contaminate the liners was 1,34*10^6 cfu/ml. This is a dose comparable to a highly infected quarter. Flushing of the liners resulted in a reduction in the number of *Streptococcus agalactiae* found in the swabs, compared with no flushing: 98,4 % and 98.9 % of the bacteria were removed after flushing with respectively water and the disinfecting fluid. The logreduction was respectively 1,80 and 2,26.

3.2 New infection rate

46 cows have been challenged in 4 runs, with respectively 10, 12, 12 and 12 cows. All cows have been checked before the start of the experiment using foremilk samples. None of the cows showed signs of an infection, based on SCC and presence of pathogens. In some cases minor pathogens were found in foremilk samples (coagulase negative Staphylococci), but no signs of inflammation was found based on SCC. In none of the cases Streptococci were found in the foremilk samples before challenge.

After the first run the number of pathogens (Str.ag.) in the suspension used to contaminate the liners, was increased from 0,5*10^6 to 3*10^6 to increase the risk of infection.

In none of the cases *Streptococcus agalactiae* could be found in the quarter foremilk samples on day 1, 2, 3, 5 or 7. None of the cows showed a significant increase in SCC. See figure 1 for average SCC in fore milk samples prior and after the challenge. Also none of the cows were signalled by the AMS to have a change in conductivity.
After challenging 46 cows it was concluded that is was not possible to create new infections with *Streptococcus agalactiae* by milking with deliberately contaminated liners, whether the liners were flushed or not flushed after contamination.

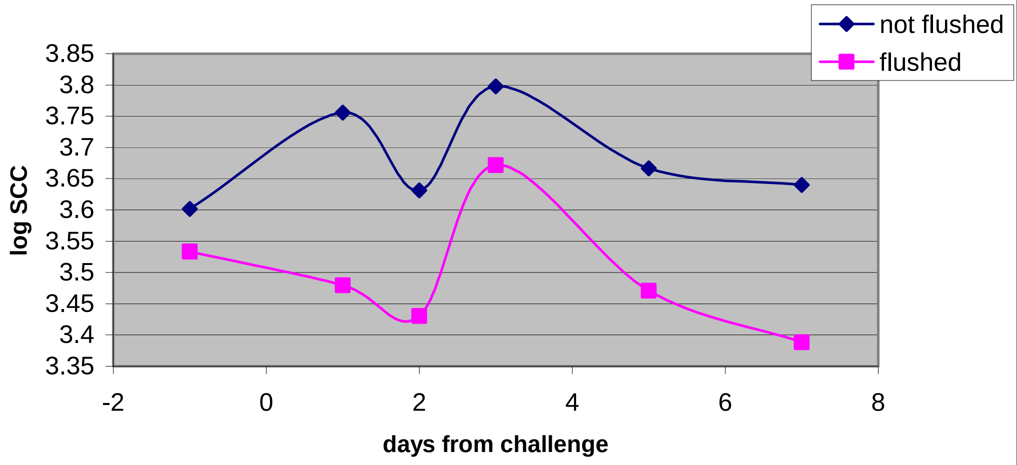

Figure 1. Average SCC of quarter foremilk samples

4. Discussion and recommendations

It is obvious that a cluster flush reduces the number of bacteria in liners. In the test conducted with a high dose of a pathogen (*Streptococcus agalactiae*) 98,4 to 98,9 % of the pathogens could be removed, respectively when flushing with cold water or a disinfectant. Logreductions were 1,8 and 2,26 respectively. Adding a disinfectant increases the reduction of bacteria, but due to the short exposure time which is available, the liners are not fully disinfected. Using a disinfectant has some disadvantages, because of the risk for residues in milk. So the system has to be flushed with water after disinfecting.
In this view the small increase in reduction of pathogens does not compensate for extra risk on contamination of food products and/or the extra costs for the more complex flushing system to avoid contamination when using a disinfectant.
Smith et al (1985) also found a comparable reduction in numbers of pathogens, ranging from 98,5 % to 99,5 % depending on the type of pathogen. Shearn et al (1994) found a smaller reduction in TBC: a logreduction of 0,41 and 0,63 using water and disinfectant respectively.
Rasmussen (1995) found in a field experiment using a Airwash system a reduction in TBC of 88 % and a reduction of *Staphylococcus aureus* of 48 % in general and 75 % after milking infected cows.

During this experiment, teat washing during milking was induced by blocking the air bleeds in the short milk tube to increase the risk for infection. None of the cows responded in one or more quarters of the udder to the challenge: no pathogens could be found in foremilk samples during one week after challenging and no major increase in SCC or conductivity was detected during this time.
The risk for a new infection when milking with contaminated liners was to small to establish a difference in flushing and not flushing of liners, even in a sub optimal milking situation as created in this experiment. Pathogens in the liner at the start of milking will normally be flushed away during milking. The highest risk for a new infection will occur at the end of milking or just after milking, when the teat canal is not fully closed yet and the milk flow has stopped. Although DeHart et al. (1975) found that new infection rate was not related to the time of infection with *Escherichia coli.* However *E. coli* is an environmental pathogen and *Streptococcus agalactiae* an udder pathogen. Smith et al (1985) showed that a cluster flush using a disinfectant reduced new infections with *Corynebacterium bovis*, but not for Streptococci and Staphylococci.

So the effect of a cluster flush on preventing new infections should not be overestimated. Also Shearn et al (1994) state that cluster flush with the Airwash system only leads a marginal reduction of bacterial load, whether using water or a disinfectant. In both cases a cluster flush could not be considered cost effective.

On the other hand it is obvious that reducing the number of pathogens in the liner by flushing the liner will contribute in the decrease of the risk for new infections. As the cluster flush can be performed during changing of cows in the AMS, there will be no effect on capacity of the AMS. Also a cluster flush will contribute to cleaner liners, because sometimes dirt is transferred from the teat to the liner during milking and will be washed away during the cluster flush. The amount of water to flush the cluster should be optimised, to prevent unnecessary waste of water.

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