



# system on teat tissues, intramammary infections and somatic cell counts

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#### **ABSTRACT**

To assess the impact of automatic milking systems (AMS) on the different aspects of milk production a research project involving both commercial and experimental dairy farms with different AMS and different management was started. This paper reports the results of a follow-up study on primiparous cows focused on assessing some markers to be used to monitor udder and teat health. Heifers were included after calving and sampled for at least 12 months. Quarter milk samples and teat measurements were taken to assess: intramammary infection (IMI) frequencies, somatic cell counts (SCC), teat thickness changes, teat skin and apex conditions. The study included 28 cows in herd A and 27 in herd B for a total number of 2344 samples. Overall, teat apex and skin conditions were maintained along the lactation. Teat skin conditions tended to decrease because of the accumulated number of milkings while lactation proceeded, but at a largely acceptable level in both herds. Teat apex conditions showed a decrease. Teat thickness changes displayed different patterns in the two herds, probably because of the different type of AMS, but in both cases a trend to decrease in thickness could be observed. The application of AMS in herd B, free from contagious pathogens, did not influence the frequency of IMI and the SCC. In herd A, characterized by the presence of Staphylococcus aureus IMI, the frequency of IMI showed a progressive increase, very likely as a consequence of the spread of infections during milking. Teat skin had no association with the frequency of IMI. Teat thickness changes outside values considered as physiological proved to be associated with decreased conditions in the teat apex score in herd A, but not in herd B. However, a decrease in teat apex score proved to be associated with an increase in IMI frequency in both herds. The results of this field trial confirm that AMS have no negative impact on IMI incidence, SCC and teat tissue conditions when the initial cow health status and overall herd management are good. In the presence of contagious pathogens, and when cows have more than 300 days in milk (DIM) the frequency of negative outcomes significantly increases and the proper control measures should be taken.

Key words: Automatic milking system, Teat tissues, Intramammary infection, Somatic cell counts

#### Introduction

The introduction of automatic milking systems (AMS) in Italian dairy herds has raised some concerns regarding mainly milk quality and cow welfare and health. These are some of

the aspects still under discussion and that need more extended investigations, most of the data being contradictory.

To assess the impact of this new technology on the different aspects of milk production a research project has been financed by the Lombardy Regional Government, in cooperation with the Animal Production Research Institute in Cremona. The project involved both commercial and experimental dairy farms with different AMS and different management. This paper reports the results of a follow-up study on primiparous cows focused on assessing some markers to be used to monitor udder and teat health and to prevent the outcome of intramammary infections and clinical mastitis.

### Material and methods

Herds

The herds considered in the study were a commercial dairy herd (herd A) and an experimental herd (herd B). Herd A, located in the province of Milan, has been milking 120 cows with 2 Lely Astronaut AMS since year 2000. Herd B is located in Cremona and includes 50 cows selected among the original herd of 80 cows previously milked in a herringbone milking parlour. The AMS machine is a single-stall robot (VMS<sup>TM</sup>, DeLaval).

# Cows and sampling

Heifers were included after calving and sampled for at least 12 months. In the commercial dairy herd, samples were taken 1 week after calving from all the freshening heifers with weekly frequency in the first month and then monthly. In the experimental dairy herd, cows were sampled twice in the first month and then monthly.

Teat thickness and skin and orifice conditions

Teat thickness was assessed by a cutimeter as described by (Hamann and Mein, 1990). Teat thickness was measured by a modified cutimeter immediately before and after milking and the changes were calculated using the formula [(thickness post-milking minus thickness pre-milking)/thickness pre-milking]. Teat conditions were assessed by the method proposed by (Casirani *et al.*, 2002a). In practical terms, a digital camera picture was taken, then transferred on to a PC and scored by a panel of 3 people using reference images. The scores ranged from 1 to 4: scores 1-2 defined a teat in good condition and scores 3-4 defined a teat in bad condition.

#### Milk samples

Quarter milk samples (QMS) were collected before milking by an aseptic procedure. Teat ends were cleaned using gauze pledgets moistened with alcohol (700ml/l). From each quarter, about 10 ml milk was collected in sterile plastic bottles. Samples were placed on ice and delivered immediately to the laboratory.

## Bacteriological analysis

At the laboratory, samples (0.01 ml) of each QMS were spread on blood agar plate (50 g/l, Difco Laboratories, Detroit MI, 48232 USA), and incubated at 37°C for 24 h. Colonies were isolated and identified by appropriate methods according to the National Mastitis Council (N.M.C., 1999). Somatic cells were counted on a Bentley Somacount 150 (Bentley USA).

#### Statistical analysis

Values were collected in a database and analysed by the means of Student's t test, Mantel-Haenszel  $\chi^2$  test on SPSS 11.5 (SPSS, 2002), while IMI prevalence was calculated by software specifically developed to analyze IMI epidemiology (Sala and Zecconi, 2002).

#### Results and discussion

The study included 28 cows in herd A and 27 in herd B with an overall number of samples of 1528 and 816 respectively in herd A and herd B, giving a total number of 2344 samples. Cows were sampled from the beginning of the lactation up to drying-off, so the number of samples was different for each cow, depending on the length of lactation.

#### Intramammary Infections

The two herds considered showed different epidemiological patterns. Herd B (experimental herd) was free from contagious pathogens, while herd A had *Staph.aureus* intramammary infections (IMI). Table 1 shows the overall bacteriological and cytological results. As expected, negative quarters had a significantly lower geometric mean level of somatic cell counts (SCC). Within positive quarters, the one infected by environmental Streptococci showed the highest level of SCC, but the differences among bac-

IMI aetiology	Herd A Cases (n.)	Herd B Cases (n.)	Total Cases (n.)
Environmental Streptococci	28	25	53
Gram Negative	8	9	17
Others	18	5	23
Staphylococcus aureus	166	0	166
Coagulase Negative Staphylococci	89	62	151
Contaminated	11	16	27
Negative	1208	698	1906
Missing	0	1	1

teria were not significant. Figure 1 reports the pattern of IMI in the 2 herds, by days in milk (DIM). The data showed that IMI in herd B were below 20% during the whole lactation, except for the period between 210-270 DIM. In herd A, a progressive increase of IMI frequency was observed as lactation proceeded. Indeed, the overall frequency was 12% when DIM was <30 d and rose to 33% at the end of the follow-up period. At calving 3.4% of the cows had at least 1 quarter infected with *Staph.aureus* and at the end 66.7% were infected.

#### Somatic cell counts

Figure 2 reports the pattern for SCC in the 2 herds by bacteriological status. The data showed that in both herds the lowest SCC levels were in negative quarters. In herd B, except for the period 0-30 DIM and 241-270 DIM the geometric mean level was below 32,000 cells/ml (log<sub>10</sub> 4.51), while in herd A, negative quarters were in the range between 30,000 cells/ml (log<sub>10</sub> 4.47) and 100,000 cells/ml (log<sub>10</sub> 5.00). Positive quarters showed a progressive increase of SCC in herd A,

Figure 1. Distribution of bacteriological positive quarters (%) in the two herds by days in milk.

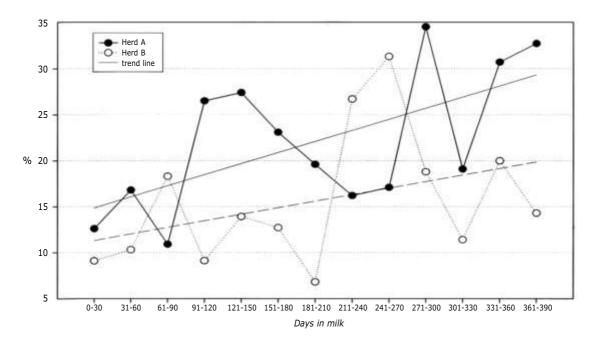


Figure 2. Somatic cell counts (geometric mean) in bacteriological positive and negative quarters by days in milk.

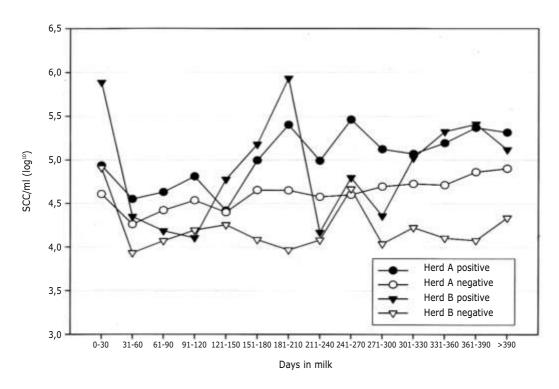
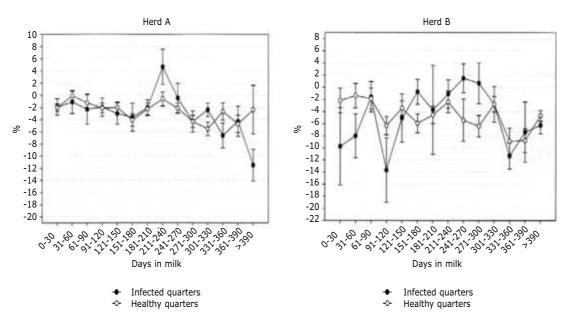


Figure 3. Mean teat thickness changes (%) before and after milking in negative e positive quarters by days in milk



from 100,000 cells/ml ( $\log_{10} 5.00$ ) up to 200,000 cells/ml ( $\log_{10} 5.30$ ) at the end of lactation, while in herd B it fluctuated with peaks of up to 1,000,000 cells/ml ( $\log_{10} 6.00$ ) at 0-30 DIM and at 180-210 DIM.

#### Teat thickness changes

The teat thickness changes before and after milking by IMI status are reported in Figure 3. Teat thickness in herd A had very small variations up to 240 DIM in both IMI positive and negative samples, then infected quarters showed

larger variations in thickness in comparison with negative quarters. The latter showed changes in thickness in the range 0, -5%, values that can be considered fully physiological. In herd B, IMI positive and negative quarters showed a different pattern from the beginning. Indeed, negative quarters showed a progressive decrease in thickness from -2% at the beginning of lactation to values in the range -3%, -9% at the end of the lactation, while infected quarters showed large and unpredictable variations in thickness along the whole lactation.

Figure 4. Distribution of teat skin scores in the two herds by days in milk

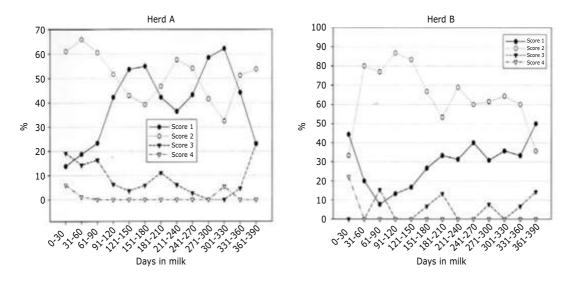
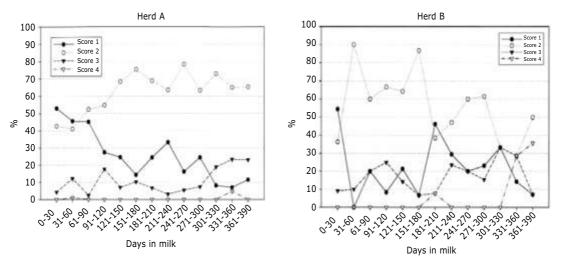


Figure 5. Distribution of teat apex scores in the two herds by days in milk



#### Teat skin score

The distribution of teat skin scores in the two herds considered is reported in Figure 4. The two patterns differed: in herd A scores 3 and 4, decreased their frequency as long as lactation proceeded, while scores 1 and 2 were the most frequent ones even if large variations could be observed between their frequencies. In herd B, scores 3 and 4 were also infrequent, while the frequency of score 2 proved to be higher in the first 210 DIM in comparison with the last part of lactation, and score 1 frequency had the opposite pattern.

#### Teat apex score

Teat apex scores patterns were also different in the two herds (Fig 5). Herd A showed an increasing frequency of score 2 as the lactation progressed up to 150 DIM, when it reached a level of about 70%, which was maintained for the remaining part of the lactation. Score 1 declined from a frequency higher than 50% in the first 30 DIM to 10% at the end of lactation. Score 3 frequency was in the range 5-10% during most of the lactation, increasing to values higher than 20% after 300 DIM.

In herd B the distribution of scores was more irregular with a higher frequency of score 2 in the first 180 DIM (range 40-90%), and frequencies in the range 30-60% in the remaining part of lactation. Score 1 frequency was very high at the beginning of lactation, but with few exceptions was

between 20 and 30% during the lactation. Score 3 frequency was in the range 10-25% for most of the lactation, while score 4 was practically observed in cows with more than 300 DIM.

Relationship between teat tissue parameters and IMI

Table 2 reports the analysis of the association between changes in teat thickness outside physiological values (defining physiological values as changes in the range -10% - +5%) and teat score 3-4 (bad condition) and 1-2 (optimal condition), by means of odds ratio estimation (OR). The analysis showed that in herd A a change outside physiological values had odds about 2 times higher of being associated with a teat apex score 3-4 than teats with changes within physiological values. In herd B that association proved not to be significant, as it was the association between teat skin score and teat thickness changes.

In table 3, the associations between each of the three different teat parameters (thickness, skin score and apex score) and IMI are assessed. Teat thickness and teat skin score did not show any significant association with IMI occurrence, while teat apex score 3-4 proved to have a 3-times higher chance of having an IMI in comparison with teat with an apex score 1-2.

Considerations on AMS impact on teat tissues and udder health

The application of AMS in dairy herds induces changes in cow management, behavior, nutrition

Table 2. Association between teat thickness changes outside physiological values and teat skin and apex scores (1-2 optimal condition; 3-4 bad condition) estimated by Odds Ratio.

Risk factor	Herd	OR	Lower confidence limits	Higher confidence limits	P=(1)
Apex Score	А	1.902	1.183	3.058	0.007
	В	1.086	0.611	1.929	ns
	Overall	1.509	1.048	2.174	0.033
Skin Score	Α	1.325	0.850	2.064	ns
	В	1.488	0.594	3.733	ns
	Overall	1.355	0.909	2.019	ns

<sup>(1)</sup> Assessed by Pearson's  $\chi^2$ 

ns = not significant

and in the interaction between the milking machine and the udder. The individual quarter milking applied by AMS could decrease some of the possible negative effects of conventional milking such as overmilking. However, the increased milking frequency and therefore the more frequent exposure of the teat to the liner and to chemicals (for cleaning and disinfection) could have negative effects on teat tissues and skin.

In this study, teat apex and skin conditions were maintained along the lactation, overall. Teat skin conditions tended to decrease due to the accumulated number of milkings while lactation proceeded, but at a largely acceptable level (scores 1 and 2) in both herds. Teat apex scores showed an increase (worse status) as days in milk increased with scores 3 and 4 more frequently observed when DIM exceeded 300.

Teat thickness changes showed different patterns in the two herds, probably because of the different type of AMS. In general terms the decrease in thickness after milking was relatively small in herd A and in the range -2% and -9% in herd B, when bacteriological negative quarters were considered.

Changes in teat tissues are known to influence the risk of infections (Zecconi et al., 1992; Hamann et al., 1994), therefore the epidemiological pattern of IMI could be influenced by the teat conditions. Overall the introduction of AMS in herd B, free from contagious pathogens, did not influence the frequency of IMI and the SCC, as already shown in

similar experiences (Svennersten-Sjaunja et al., 2000; Casirani et al., 2002b). In herd A, characterized by the presence of Staph.aureus IMI, the frequency of IMI showed a progressive increase, very likely as a consequence of the spread of infections during milking, as reported in similar studies (Hamann and Reinecke, 2002; Petermann et al., 2002). These results suggest the role of liner material and the disinfection procedures were inadequate or not properly performed by the system, and their influence should not be underestimated. The presence of a significant number of herds with Staph.aureus IMI in different countries emphasizes the importance of addressing this aspect of AMS milking.

The results of the study showed that teat skin had no association with the frequency of IMI. Teat thickness changes outside values considered as physiological proved to be associated with decreased conditions in teat apex score in herd A, but not in herd B. Meanwhile, a decrease in teat apex score proved to be associated with an increase in IMI frequency in both herds.

These results suggest that changes in teat tissues and in teat skin conditions induced by AMS are unable to increase the risk of IMI in the two herds. Otherwise, the effect on teat apex proved to have a strong association with increased frequency of IMI. The decrease in teat apex conditions is more frequent as lactation progresses, suggesting that cows in late lactation should be prevented from being milked too often.

4.917

Risk factor	Herd	OR	Lower confidence limits	Higher confidence limits	P=(1)
Teat Thickness	Α	0.828	0.640	1.072	ns
	В	0.864	0.581	1.284	ns
	Overall	0.838	0.675	1.041	ns
Skin Score	Α	0.954	0.544	1.673	ns
	В	0.813	0.179	3.695	ns
	Overall	0.934	0.552	1.582	ns
Apex score	Α	3.376	2.048	5.564	0.000
	В	2.905	1.327	6.362	0.006

2.114

Association between teat tissues-related risk factors and IMI outcome

Table 3.

Overall

3.224

0.000

 $<sup>^{(1)}</sup>$  Assessed by Pearson's  $\chi^2$  ns = not significant

#### Conclusions

The results of this field trial confirm that AMS have no negative impact on IMI incidence, SCC and teat tissue conditions, when the initial cow health status and overall herd management are good. In presence of contagious pathogens, and when cows have more than 300 DIM, the frequency of negative outcomes deriving from AMS application significantly increases and the proper control measures should be taken.

#### REFERENCES

- CASIRANI, G., BINDA, E., PICCININI, R., ZECCONI, A., 2002a. Sviluppo ed applicazione di un sistema basato sulla valutazione dello stato del capezzolo. Obiettivi Doc.Vet. 23(10): 21-27.
- CASIRANI, G., PICCININI, R., MIGLIORATI, L., PIRLO, G., ZECCONI, A., 2002b. The effects of voluntary milking system on teat tissues, intramammary infections and somatic cell counts. Proc. 1<sup>st</sup> North Am. Conf. on Robot Milking, Toronto, Canada. Wageningen Pers. IV: 49-54.
- HAMANN, J., BURVENICH, C., BRAMLEY, A. J., OSTERAS, O., WOOLFORD, M., WOYKE, M., HAIDER, W., MAYNTZ, M., LEDU, J., 1994. Teat tissue reactions to machine milking and new infection risk. Bull. Int. Dairy Fed. 297: 1-43.
- HAMANN, J., Mein, G. A., 1990. Measurement of machine-induced changes in thickness of the bovine teat. J Dairy Res. 57: 495-505.

- Hamann, J., Reinecke, F., 2002. Machine milking effects on udder health comparison of a conventional with a robotic milking system. Proc. 1st North Am. Conf. on Robot Milking, Toronto, Canada. Wageningen Pers. IV: 17-27.
- NATIONAL MASTITIS COUNCIL., 1999. Laboratory handbook on bovine mastitis. National Mastitis Council Inc., Madison WI, USA.
- Petermann, M., Wolter, W., Rittershaus, C., Kloppert, B., Seufert, H., Zschock, M., 2002. Automatic milking systems: udder health and milk flow profiles. Proc. 1st North Am. Conf. on Robot Milking, Toronto, Canada. Wageningen Pers. IV: 67-70.
- SALA, S., ZECCONI, A., 2002. Development of an epidemiological software for intramammary infection diagnostic data recording and analysis. In: Proc. Ann. Meet. National Mastitis Council, Orlando Fl, USA, 41:156-157.
- SPSS STATISTICAL PACKAGE FOR SOCIAL SCIENCE. 2002. SPSS Advanced Statistics 11.5. SPSS Inc., Chicago, IL, USA.
- Svennersten-Sjaunja, K., Berglund, I., Pettersson, G., 2000. The milking process in an automatic milking system, evaluation of milk yield, teat conditions and udder health. pp 277-288 in Proc. Int. Symp. on Robot Milking, Lelystad NL. Wageningen Pers, Wageningen, Netherlands.
- ZECCONI, A., HAMANN, J., BRONZO, V., RUFFO, G., 1992. Machine-induced teat tissue reaction and infection risk in a dairy herd from contagious mastitis pathogens. J Dairy Res. 59: 265-271.