# Comparison of different diagnostic measures to identify bovine mastitic quarters at milking time

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## Summary

A precise diagnosis of bovine mastitis is only possible by examination on quarter level. The pre-selection of quarters with changes in quarter milk yield and electrical conductivity depending on inflammatory processes, will be useful under economical aspects, particularly concerning the following cyto-bacteriological examination. These parameters are already available, especially in automatic milking systems.

Sensitivity, specificity and probability of misclassification to predict the diagnosis of clinical and sub-clinical mastitis were calculated, using a retrospective analysis of both parameters in comparison to a cyto-bacteriological reference based on quarter foremilk samples. Ninty-six cows on two farms were part of the survey, over a period of at least one lactation per cow.

Using analyzing techniques, based on consecutively or momentary orientated surveys of the parameters, up to 70 % of all sub-clinical mastitis can be identified, with a specificity of approximately 80 %.

The results indicate that these techniques can be used as early automatic warning systems, although they are not sufficiently precise to replace the cyto-bacteriological examination.

***Key words****: Bovine, mastitis, diagnosis, milking, quarter yield, electrical conductivity.*

## Introduction

The early identification of mastitic quarters is essential for all dairy farmers, to ensure animal welfare, milk quality and farm productivity. The most precise diagnostic categorization in this respect can be obtained by cyto-bacteriological analysis of quarter milk samples (Table 1). Nevertheless, this procedure is not economic for a continuous screening of udder health. For this reason, alternative parameters, which however indicate an inflammation only, are used for an assessing categorization of the udder health. In this context, most systems employed in milking devices up to today (especially automatic milking systems), measure the electrical milk conductivity and milk yield at quarter level. This study offers the possibility for a retrospective assessment of the diagnostic quality of the parameters mentioned above, by using the cyto-bacteriological and clinical examinations parallel.

## Material and methods

Ninty-six German Holstein cows, originated in two different production systems (loose house system and fixed stanchion barn) were sampled over at least one lactation. During lactation, sampling was carried out 18 times. The sampling pattern included weekly survey for the first eight and the last three weeks of lactation, and monthly intervals for the period in between. At morning milking, cows were milked with a quarter milking machine, after foremilk sampling had occurred for cyto-bacteriological analysis. In addition to this, all udders were clinically examined. Bacteriological examination was performed according to IDF recommendations (IDF, 1981). Somatic cell count (SCC) was determined by fluorescent microscopy (Fossomatic®, Foss Electric™, Denmark, precision: Cv < 5%; Schmidt-Madsen, 1975). The quarter milk yield was determined by weighing electronically (DNP15 SNR 2116076262, Ohaus™; precision: Cv 0,5 %). Furthermore, electrical conductivity was measured in quarter machine samples (LF 539, WTW, precision: Cv < 0,5 %). Udder health was determined according to the categorization scheme shown in table 1. Physiological ranges on quarter level were determined (Figure 1), using data from cows, healthy on all four quarters throughout the whole lactation. Milk yield data could be corrected lactation-related, with these ranges (Grabowski, 2000). The statistic processing of diagnostic data was carried out by means of the procedure Freq (SAS, 1987). The thresholds

*Table 1. Mastitis definition* (DVG, 1994).*

| Cell content | Pathogens | |
|---|---|---|
| [cells / ml milk] | Not identified | Identified |
| < 100.000 | Normal secretion (NS) | Latent infection (LI) |
| > 100.000 | Unspecific mastitis (UM) | Mastitis (M) |

*based on cyto-bacteriological examination of udder quarter foremilk samples

were calculated using the principle of least probability of misclassification (PM) (Krömker et al., 1998). The diagnostic test criteria sensitivity, specificity, and PM were determined with Win Episcope 2.0® (Epidecon).

## Results

The data of 96 cows with 382 quarters was used in the present study. Udder health of animals and quarters involved was determined at the beginning of the survey on both farms, with constant monitoring during the trial. Table 2 shows the prevalence of mastitis at the onset of the investigation for both production systems.

Udder health was defined by SCC less than 100,000 cells / ml and the absence of pathogenic bacteria in foremilk samples (DVG, 1994). By this definition merely six animals were considered as udder-healthy on all quarters throughout the whole lactation. The physiological development of quarter milk yield and electrical conductivity during lactation is depicted in figure 1.

During the survey, 684 new cases of sub-clinical mastitis (NS becoming M or UM) and 44 clinical cases of mastitis were recorded. In order to assure comparability, a similar number of non-disease cases (NS remaining NS) was used along with the new cases.

Because of the retrospective analysis of the electrical conductivity and quarter yield data, thresholds for the individual parameters and a combination of both were established separately. Analysis methods associated with 'moment' and 'time' were also tested. Those associated with the momentary survey considered only the actual value, whereas those associated with consecutive survey compared the actual value with the preceding values. Table 3 presents the methods of analysis employed in the present study, along with their thresholds; table 4 shows their corresponding values for sensitivity, specificity and PM.

The results of the analyses show that between 12 and 70 % of all newly diseased quarters can be readily identified, using these parameters. The combination of different thresholds increases the sensitivity. In particular it is interesting that the decrease of relative quarter yield from one sampling to the subsequent one by 10 %, alone and in combination with other parameters is the method with the best identification rates. The specificity

*Table 2. Udder health status (%) at the onset of sampling.*

| Farm | NS | LI | UM | M |
|---|---|---|---|---|
| 1 (n = 54 cows) | 27,8 / 68,2* | 3,7 / 1,9 | 55,6 / 25,7 | 13,0 / 4,2 |
| 2 (n = 42 cows) | 35,2 / 70,2 | 7,1 / 3,0 | 31,0 / 18,5 | 26,2 / 8,3 |

*= The first value refers to the cows, the second to the quarters.

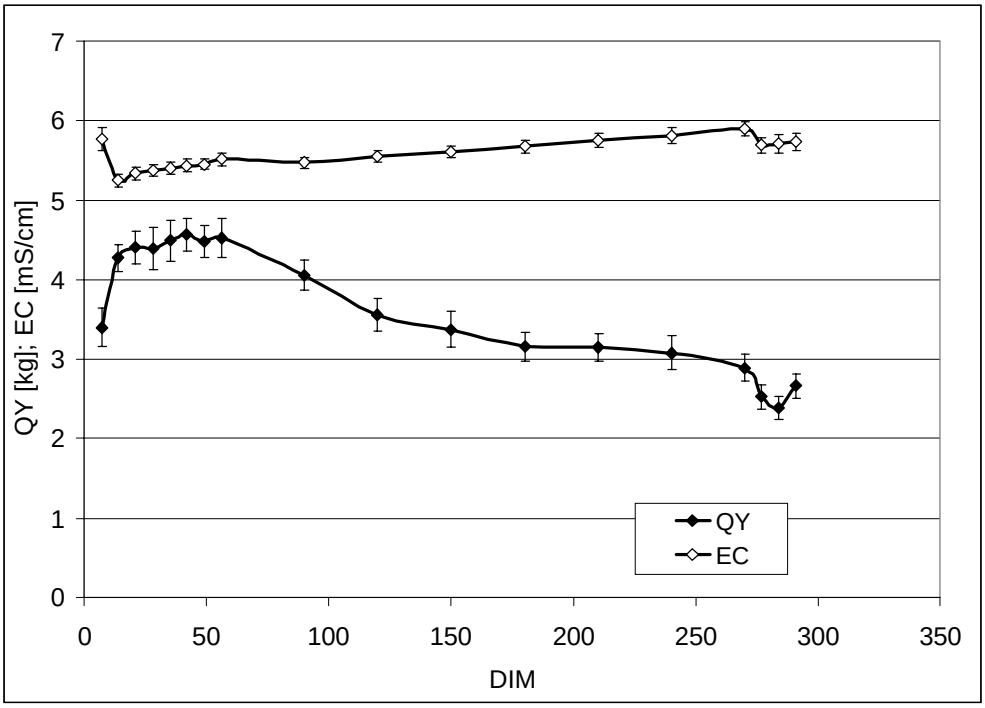


*Figure 1. Physiological course of quarter milk yield (QY) [kg] and electrical conductivity (EC) [mS/cm], mean and standard error of the mean; DIM = days in milk (n = 6 cows).*

is usually above 70 %. The PM's demonstrate, that considering this material, 30 to 40 % of all quarters are always misclassified. The number of PM's for clinical mastitis, that are particularly low, do not indicate a high recognition rate of clinical cases, but are the result of the high number of non-clinical cases.

## Discussion and conclusion

Using a retrospective analysis of electrical conductivity and lactation corrected yield data, from quarter machine milk samples and corresponding cyto-bacteriological findings, the diagnostic value of these parameters in single usage or in combination, was calculated, in order to identify mastitis cases, omitting clearly the influence factor of the milking interval. By means of these parameters on quarter machine milk level and using appropriate analysis procedures, the identification of ca. 70 % of all sub-clinical new infections and of 40 % of all clinical mastitis cases is feasible. Zero to 20 % of all non-diseased quarters were misclassified with this method. Although an extension and consecutively an optimization of the analysis systems should promise better results, automatic analysis systems – integrated in milking devices – can not be expected to provide a secure early diagnosis of mastitis under the tested conditions. Actually,

*Table 3. Summary of analysis methods employed*

| Method No. | Analysis | Threshold healthy / not healthy |
|---|---|---|
| 1 | EC, sub- clinical | 6,5 mS/cm |
| 2 | EC, sub- clinical | 15 % of difference between quarters |
| 3 | EC, sub- clinical | Combination No. 1 & No. 2 |
| 4 | EC, sub- clinical | Value before vs. value after + > 10% |
| 5 | EC, clinical | 8 mS/cm |
| 6 | EC, clinical | 9 mS/cm |
| 7 | QY, sub- clinical | 15 % of decrease of QY in relation to the previous sample after lactation correction |
| 8 | QY, sub- clinical | 10 % of relative QY reduction in one quarter in relation to the previous sample |
| 9 | EC, QY sub- clinical | Combination No. 1 and/or No. 7 |
| 10 | EC, QY sub- clinical | Combination No. 4 and/or No. 7 |
| 11 | EC, QY sub- clinical | Combination No. 1 and/or No.4 and/or No. 7 |
| 12 | EC, QY sub- clinical | Combination No. 1 and/or No.4 and/or No. 7 and/or No.8 |

*Table 4. Misclassification of different analytical methods.*

| *Method No.* | *Sensitivity (%)* | *Specificity (%)* | *PM * (%)* |
|---|---|---|---|
| 1 | 26.7 | 99.4 | 37.0 |
| 2 | 21.2 | 95.6 | 41.6 |
| 3 | 11.9 | 99.1 | 44.4 |
| 4 | 54.9 | 77.5 | 33.8 |
| 5 | 37.2 | 98.8 | 3.0 |
| 6 | 17.9 | 99.5 | 2.5 |
| 7 | 44.4 | 76.9 | 39.7 |
| 8 | 68.6 | 72.2 | 29.6 |
| 9 | 52.8 | 63.5 | 41.9 |
| 10 | 54.9 | 77.5 | 41.1 |
| 11 | 58.3 | 77.5 | 32.1 |
| 12 | 67.1 | 76.0 | 29.4 |

* Probability of misclassification [percentage of false (+) & false (-) diagnoses].

## References

these techniques should be regarded as alarm systems that require verification by a cyto-bacteriological examination before any therapeutic measure is taken.

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