MONITORING, PREVENTION AND TREATMENT AS KEY PILLARS IN THE MANAGEMENT OF BOVINE MASTITIS

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Abstract

The quality of milk is a component based on certain individual indicators that allow the correct assessment of the quality of the dairy product starting from the producing animal, the quality of the feed, the exploitation system, the state of health, milking hygiene, the storing and transportation conditions of milk towards the processing unit. The purpose of this study was to monitor udder health and establish a proper treatment based on a specialized protocol that employs the assessment of physicochemical milk parameters, number of somatic cells, as well as identification of the pathogen agent generating the disease. Milk samples from 69 dairy cows were collected at the beginning and middle of lactation, a total number of 19 cases of clinical and subclinical mastitis cases being observed. The physicochemical parameters of milk, somatic cell count, and pathogen prevalence were assessed. A very small significant negative relationship was observed between somatic cell score and lactose percentage, ($r = 0.667$, $p < .001$). The most frequent encountered pathogens were *Streptococcus spp*. A management approach for mastitis, as well as other diseases, should include the following major components: prevention, monitoring, and diagnosis.

Key words: bovine mastitis; somatic cells count; on-farm milk culturing; milk quality

Introduction

A fundamental problem of the twenty-first century will be to ensure a sustainable and secure supply of food to the world's rising population. As a result, the fundamental goal of dairy farming is to reduce milk losses and other losses associated with unhealthy cows. Furthermore, a move from therapeutic to preventative health management for mastitis and a broad range of other infectious and non-infectious bovine illnesses, some of which are known to have significant negative impacts on cow performance and well-being, would be preferred (Farschtschi *et al.*, 2022).

Bovine mastitis is caused by a variety of factors, including infectious pathogens, inadequate nutrition, and ineffective farm management (1, 2). It has an impact on animal welfare and causes huge economic losses owing to lower milk yield, decreased milk quality, higher treatment expenses, and early culling (Ajose *et al.*, 2022).

Milk quality is a component based on certain individual indicators that allow the correct assessment of the quality of the dairy product starting from the producing animal, the quality of the feed, the exploitation system, the state of health, milking hygiene, the storing and transportation conditions of milk towards the processing unit (Leitner *et al.*, 2019).

According to the EU standards, the overall quality of milk refers to the three pillars: compositional, nutritional and biological quality (milk composition, cryogenic point, milk density, milk acidity, presence/absence of antibiotics and inhibitory substances), hygienic quality (<100,000 NTG/ml - the total number of aerobic mesophilic germs/ml milk) and sanitary quality (<400,000 NCS/ml - the number of somatic cells/ml milk). The discharge of somatic cells in milk is the best indicator of the beginning of inflammation in the mammary gland, and it is the most extensively used trustworthy biomarker of udder health (Jadhav *et al*., 2018).

The purpose of this study was to monitor udder health and establish a proper treatment based on a specialized protocol that employs the assessment of Physico-chemical milk parameters, number of somatic cells, as well as identification of the pathogen agent generating the disease.

MATERIAL AND METHOD

The research was carried out in the laboratory and zootechnical biobase within the Research and Development Station for Cattle Breeding Dancu, Iasi, from March to September 2022. A total number of 69 cows were included in the study, from second to forth lactation were included in the study.

Milk sample collection

Milk sample collection was carried out in accordance with the provisions of National Veterinary Sanitary and Food Safety Authority Order no. 35 of 2016, milk samples being collected from cows at the beginning (more than 45 days) or

at mid lactation, being well known that in the last part of lactation, the number of somatic cells is physiologically higher. For the physico-chemical and microbiological analysis, milk samples were collected in a sterile container, refrigerated and transported in short time to the laboratory.

Determination of somatic cells count and physicho-chemical parameters

To determine the number of somatic cells and the physicho-chemical parameters, the milk samples collected from the cows included in the study were warmed up to 37^{O}C. Three determinations were made for each milk sample. The following physico-chemical parameters were assessed: fat, dry matter, protein, lactose, casein and density. The number of somatic cells was assessed according to the fluoro-opto-electronic method (SR EN ISO 13366-2:2007), using the CombiScope 600/300 FTIR System, Delta Instruments.

Identification of affected udder quarters

To identify udder quarters with mastitis, a topical-glandular approach was used based on the assessment of somatic cell count using the "cow side" method, California Mastitest. Briefly, following teat disinfection, the first strips of milk were discharged and a small amount of milk, from each quarter, was collected in the plastic pallet with 4 cups marked A, B, C and D. An equal amount of reagent was added to this milk sample. The two elements were homogenized by rotating movements and the results were read after 10 seconds.

On-farm milk culturing

For the individual milk samples with somatic cells values above 200.000 cells/ml, with or without visible signs of clinical mastitis, a microbiological evaluation was carried out to identify the potential pathogenic agent, using the ClearMilk kit. The milk sample was placed on the surface of the Petri dish using the inoculation wand and incubated for 22-26 hours at 37.5°C. The Atlas of Agents was used to identify the pathogen. The presence of one colony for major pathogen agents such as *E. coli*, and *Staphyloccocus aureus* was considered positive. For other major pathogens such as *Streptococcus spp.* five or more colonies were considered as positive. The samples in which more than two pathogens were identified were discharged and considered as contaminated.

Statistical analysis

The statistical analysis was carried out using Graphpad software. The mean and standard deviation for all physico-chemical parameters were determined to evaluate possible differences between healthy cows and those with mastitis. To create a normal distribution, milk somatic cell count (SCC) was converted to somatic cell score (SCS) (SCS = (log2 (SCC /100000) + 3) (Antanaitis*, et al*., 2021). The mean and standard error of the mean were used to represent descriptive statistical variables.

RESULTS AND DISCUSSIONS

Bovine mastitis initially affects only one mammary quarter, after which the infection may also spread to the others. For this reason, the diagnostic tests were performed individually (individual approach) and for each mammary quarter separately (topic-glandular approach). Figure 1 depicts the several steps employed in the diagnostic of bovine mastitis, as well as the various therapeutic choices based on the diagnostic findings.

The presence of inflammatory changes in the mammary gland may be characterized by an increase in the number of somatic cells. Previous studies have shown that permeability changes related to inflammatory reactions in secretory cells can cause changes in milk components, such as the percentage of protein, fat and lactose, as well as a decrease in milk production (Table 1).

Early detection of bovine mastitis may help improve treatment strategies, increase milk production and reduce antibiotic use on farms. Thus, current scientific research focus is being orientated in identifying new and less expensive biomarkers for early diagnosis. One such proposed biomarker is the percentage of lactose in milk.

In our study, we investigated the possible correlations between the somatic cell score (SCS) and other physico-chemical parameters of milk, in healthy cows and in cows with mastitis, using the Pearson correlation coefficient and linear regression. A very small significant negative relationship was observed between somatic cell score and lactose percentage, ($r = 0.72$, $p < .001$) (Figure 2).

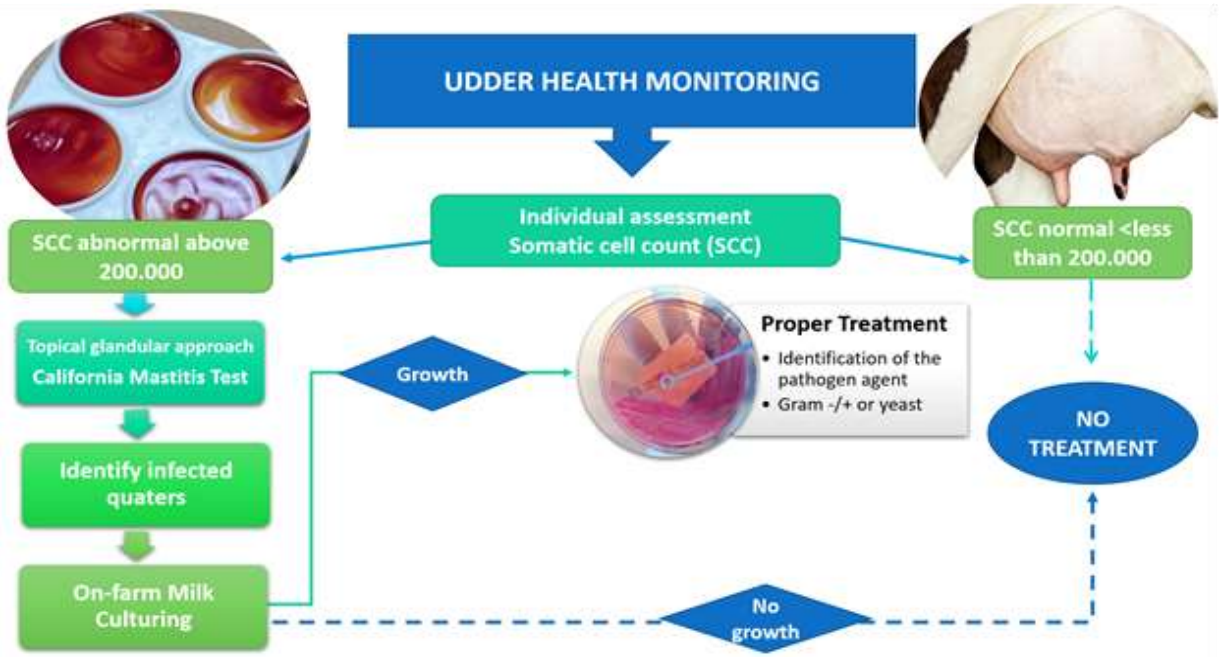

Figure 1 **Diagram of decision making for diagnosing and treatment of mastitis on dairy farms**

Table 1

Descriptive statistical variables of the physico-chemical parameters of milk in cows with mastitis and healthy cows

Type	Mastitis (n=19)	Healthy (n=50)	*p-value*
SOMATIC CELL SCORE	15.10 ± 2.25[a]	12.36 ± 1.19	< 0.0001
FAT (%)	3.48 ± 0.82[b]	3.96±0.89	0.0488
PROTEINS (%)	3.39±0.51[c]	3.10±0.27	0.0033
LACTOSE (%)	4.65±0.68[d]	5.04±0.22	< 0.0001
DRY MATTER	12.52±1.78[e]	11.65±1.02	0.0063
CASEIN (%)	2.54±0.23[f]	2.79±0.42	0.0028
a,b,c,d,e,f - Superscript letters indicate that the difference is considered statistically significant.			

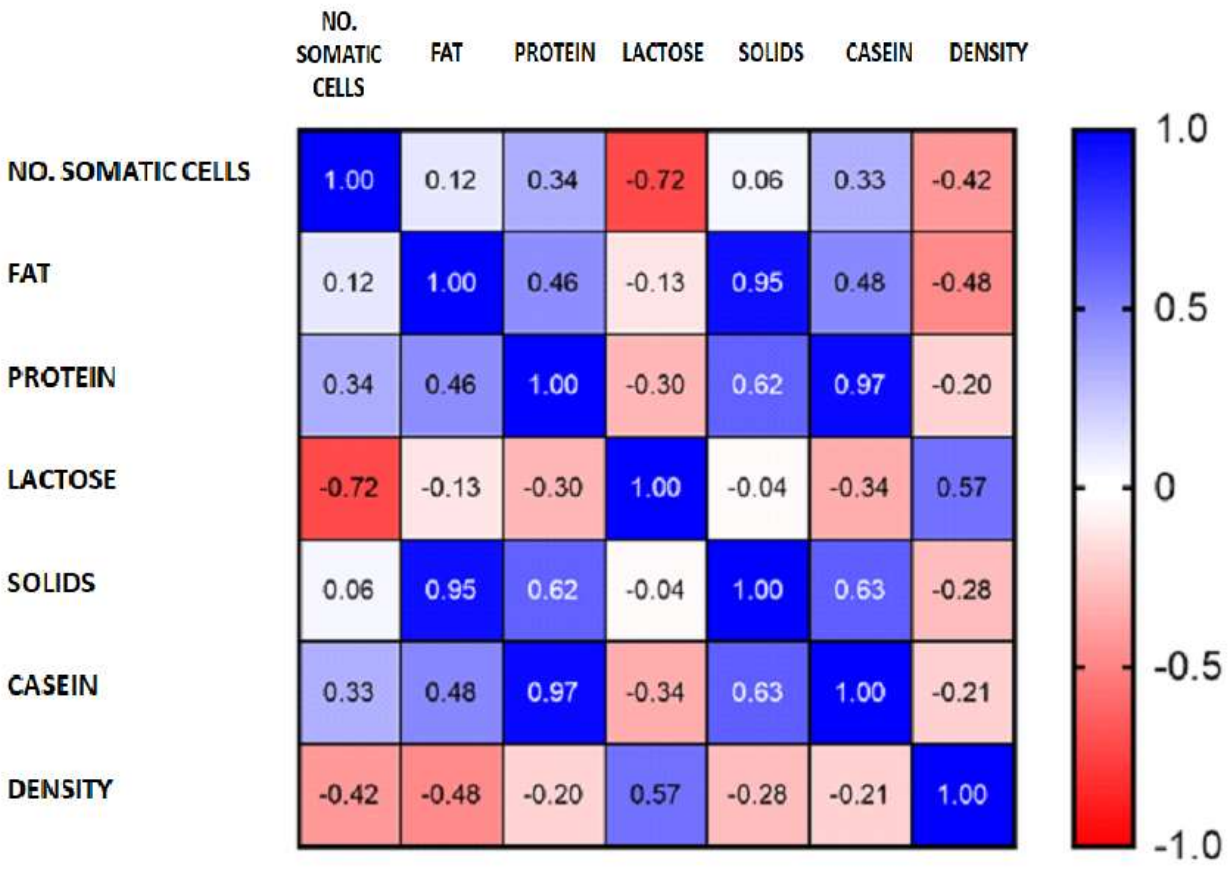

Figure 2 **Correlations between the physico-chemical parameters of milk and the somatic cell score (SCC)**

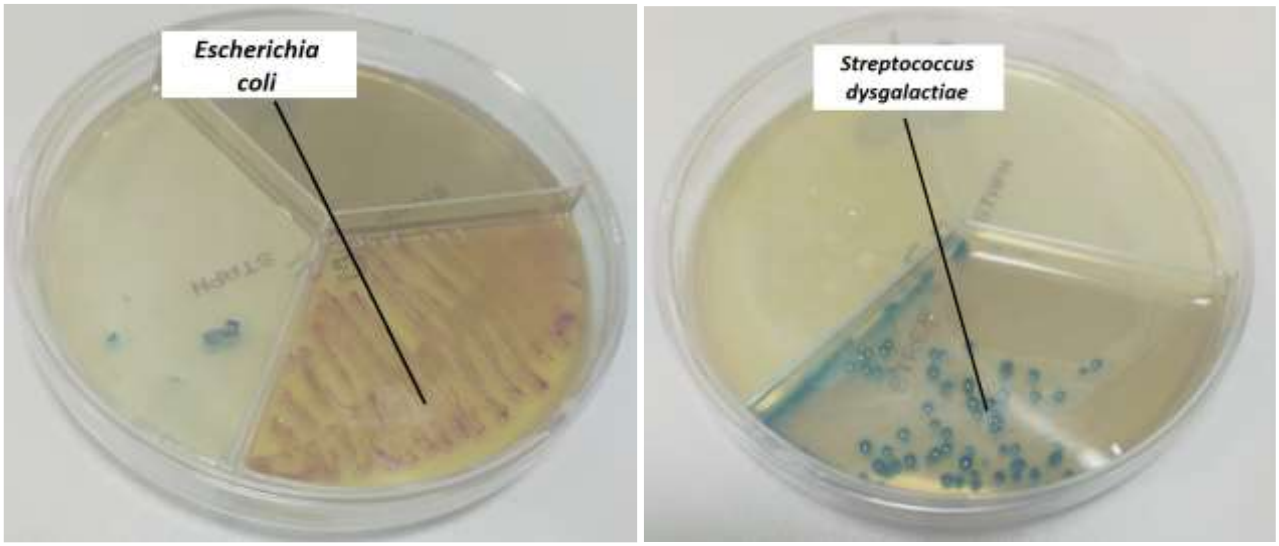

Figure 3. **Pathogen identification in milk samples from cows with clinical mastitis using the Clear Milk test**

Identifying the pathogen agent responsible for inducing bovine mastitis, samples collected from cows with visible signs of clinical mastitis, as well as milk samples with a somatic cell count above the threshold of 200.000 cells/ml were analyzed using the Clearmilk kit (Figure 3).

The on-farm culturing system allows practitioners to distinguish between pathogens. Antibiotic treatment may be chosen for mastitis cases that are likely to benefit from this type of therapy, such is the case of mastitis generated by Gram-positive pathogens (*Strep. dysgalactiae*, *Strep. uberis*, or coagulase-negative staph.). In the case of Gram-negative infections (E. coli or Klebsiella) antibiotics should not be used unless it becomes toxic (Smulski et al., 2020).

In our study, the most frequent encountered pathogen agents were *Streptococcus spp*. A narrow group of bacteria (*Staphylococcus aureus, Streptococcus uberis, Streptococcus dysgalactiae*, and *Escherichia coli*) are accountable for 80% of mastitis cases, with *S. uberis* consistently identified as the predominant pathogen in dairy farms worldwide (Maciel-Guerra *et al.*, 2021).

Reducing udder pathogen exposure is one of the most effective mastitis prevention methods and may be achieved by implementing proper milking parlor procedures, such as adequate pre-dipping, ensuring that the whole teat skin is coated to the base while providing adequate time for the pre-dipping solution to kill the pathogens, and subsequently dry wiping of teats (Ruegg, 2017).

CONCLUSIONS

A management approach for mastitis, as well as other diseases, should include the following major components: prevention, monitoring, and treatment. Due to the fact that mastitis is a pluri-factorial disease, the preventative component should target the primary risk factors for mastitis. Independently of the kind of mastitis and the pathogen agent, there are three basic farm management aims that should be considered: stimulation of the udder's natural defensive systems, ensuring complete and proper milking and lowering udder pathogen susceptibility.

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