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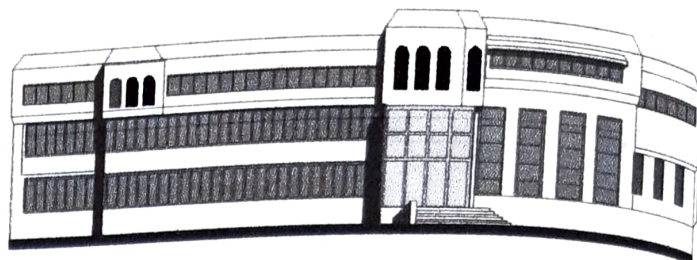


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Somatic cell count as an indicator of the hygiene status of the raw milk

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Abstract: The milk hygiene is one of the most crucial for obtaining good quality dairy products. That's why there is great interest for the examination of the somatic cell count (SCC) in raw milk, as a major indicator of the quality of raw milk. The presented results describe the average cell size as a result of elevated somatic cells in raw milk. The average size of cells in normal milk is 10,88 μm (122×10^3 SCC/ml). But, elevated number of somatic cells more than $200 \times 10^3/\text{ml}$ leads to enlarged size of the somatic cells.

Key words: somatic cells, Lactoscan SCC, raw milk

Introduction

Milk is a product which contains high-value nutritional biological fluid composed of water, proteins, fat, sugars, minerals and other elements (Li N., et al., 2014). Managing and controlling mastitis based on somatic cell counts can help ensure high milk quality and yield (Kim et al. 2017). The optimal SCC cut-off point to distinguish between infected and uninfected at the individual cow level has been established at 200,000 cells/mL (IDF, 2013). Regular SCC testing and associated udder health monitoring programs have a substantial positive influence on single animals as well as the entire herd (Barkema et al. 1998). Somatic cells in milk are mostly white blood cells that infiltrate the mammary gland in response to infection and when present in milk serve as a sensitive indicator of inflammatory conditions. Somatic cells naturally exist in the raw milk including macrophages, polymorphonuclear neutrophil cells (PMNs), and lymphocytes (Boutinaud M., and Jammes H., 2002). Due to the

difficulty of managing and controlling mastitis occurrence in dairy herds, monitoring somatic cells in milk from individual cows is critical for diagnosing clinical and subclinical mastitis as well as securing the quality of milk and milk products (Kim et al. 2017).

What is Milk SCC?

The term SCs mean cells that are normally present at low levels in milk and are derived from the body of the animal. The majority of these cells in normal milk are cells from the udder secretory tissue (epithelial cells), and some are white blood cells (leukocytes). Leukocytes represent the second line of defence, the first being the anatomical and chemical barriers of the teat apex and canal in the mammary gland. The epithelial cells in milk result from the desquamation of the mammary epithelium of the alveoli and the ducts. Majority of exfoliated epithelial cells present in milk are viable and exhibit characteristics of fully differentiated alveolar cells (Boutinaud and Jammes 2002).

Material and Methods

For the purpose of the study, we used milk samples from one farm in the Pelagonia region. The farm is considered a big farm for Macedonian standards, with more than 100 cows. Cows are from Holstein - Friesian breed housed in a tie - stall barn and milked three times per day. Cows are fed mainly with hay, concentrate for milking cows and brewers spent grain. The average herd milk yield on the farm is 9.300,00 kg/year. During the study total of 20 milk samples were analyzed. Samples were collected from the individual cows. Briefly, freshly collected individual cow milk samples were obtained from the hand milking after discarding the first milk. Each sample was preserved with bronopol.

Preparation of the milk samples for counting

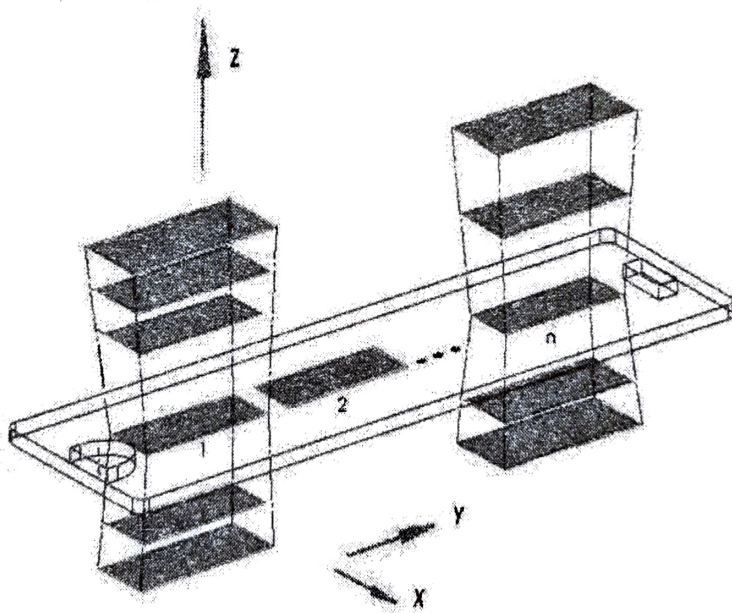
The tempered milk samples on the 200C were first stirred using the Mini Vortex mixer. The stirring process was repeated 3-4 times paying attention to the sample not to reach the cap of the container. Than 100 μL of the milk was pipetted to the micro-tubes with SOFIA GREEN lyophilized fluorescent dye. Micro-tubes with the milk sample and the dye were stirred 8-9 times on a Mini Vortex for 1-2 seconds taking care while stirring the solution not to reach the cap of the micro-tube. In order for the dye to interact with the cell nuclear material, it is necessary to wait at least 1 and maximum 20 minutes before starting to count.

For the examinations we used LactoChips4x, so we took 8 μL of stained samples and transferred that amount to each of the four chamber of the

Lactochip. Then lactochips were placed in the automatic counter where the somatic cells were counted, and totally 16 images per sample were taken, and the average number and average size of the cells were calculated by the Lactoscan SCC software.

Working principle of LACTOSCAN SCC

Unique, 3D, multi-image, sequential scanning process, based on a precise fluorescent optics and low magnification, images analysis software, LACTOSCAN SCC is a fast, precise and reliable counter of somatic cells. Via automatic displacement of the mechanism on axes X – Y and liquid lens Z, the device is capturing a maximum of 60 images. After capturing, the images are being processed by the embedded software and the average result, calculated by using the formula from IDF/ISO13366, of all the filmed images is displayed. The whole process, after placing the LACTOCHIP in the cartridge, is automatic.



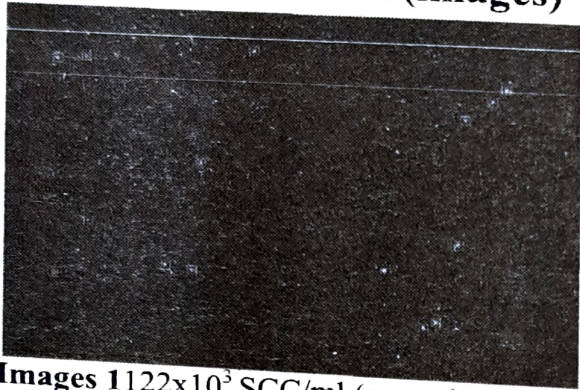
Picture 1: Working principle of the Lactoscan SCC

Results and discussion

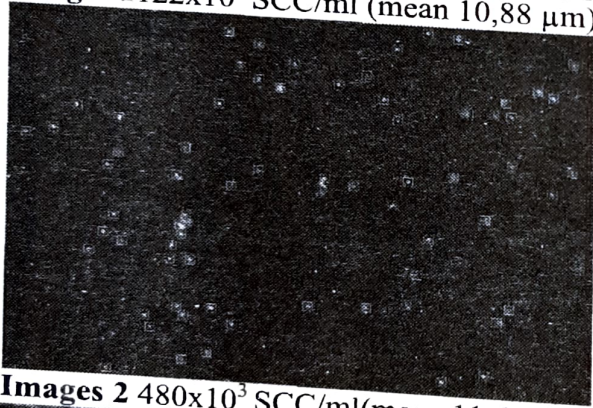
Somatic cells are influenced by many factors like cow productivity, health, parity, lactation stage, and breed of an animal, also changes in environmental and stressful conditions may lead to increase the number of somatic cell in raw milk (Alhussien, M. N., & Dang, A. K. 2018). The number of somatic cells may be elevated in response to infectious bacteria like *Staphylococcus aureus* and *Streptococcus agalactiae* which lead to losses in milk production and changes in

milk composition (Malek dos Reis et al. 2011; Sharma, Singh, and Bhadwal 2011). Somatic cellscomposing a four main cell types: macrophages, polymorphonuclear neutrophils cells (PMNs), lymphocytes and epithelial cells (Li, N., Richoux et al., 2014). Milk is produced by mammary epithelial cells (Li, N., Richoux et al., 2014).These cells are shed from the mammary epithelium during lactation and they are the first defense line of the mammary glands (Boutinaud and Jammes, 2002).

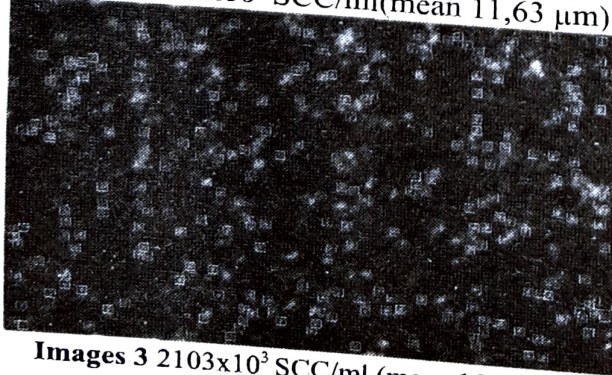
LACTOSCAN SCC(images)



Images 1122x10³ SCC/ml (mean 10,88 μm)

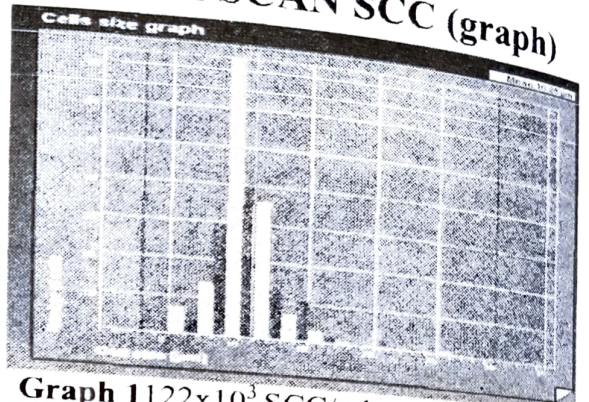


Images 2 480x10³ SCC/ml(mean 11,63 μm)

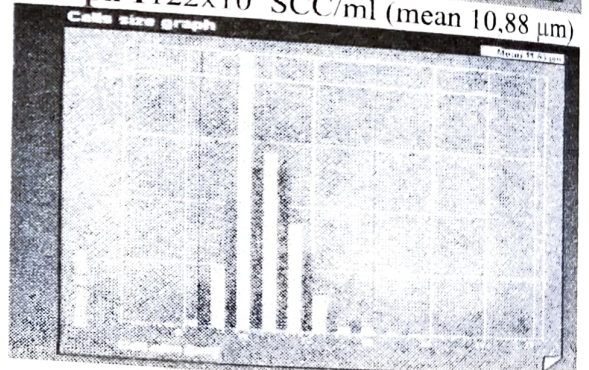


Images 3 2103x10³ SCC/ml (mean 15,91 μm)

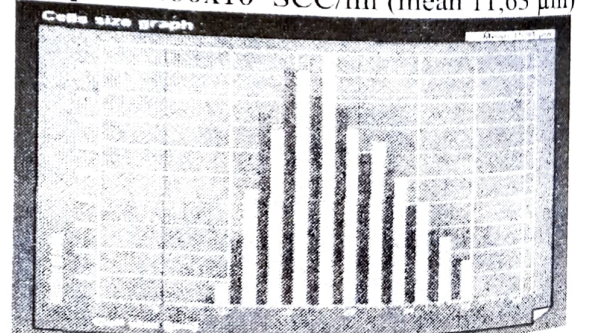
LACTOSCAN SCC (graph)



Graph 1122x10³ SCC/ml (mean 10,88 μm)



Graph 2 480x10³ SCC/ml (mean 11,63 μm)



Graph 3 2103x10³ SCC/ml(mean 15,91 μm)

Presented results indicate that the average size of the cells is rising as a result of increased somatic cell in raw milk. In the image 1 and graph 1 we can see that the average size of cells is 10,88μm (122x103SCC/ml). According to Li, N.,

Richouxet al., (2014) the predominant cell type in healthy cow milk generally are macrophages, which fight against bacterial invasion quickly by engulfing action. Also, macrophages participate in the specific immunity as do lymphocytes (Burvenich et al. 2003). Elevated number of somatic cells mostly is related with infection of the mammary gland. When SCC are more than $200 \times 10^3/\text{ml}$, the udder is considered to be infected (Li, N., Richouxet al., 2014). In the image 2 and 3 are presented results with $480 \times 10^3 \text{ SCC/ml}$ and $210^3 \times 10^3 \text{ SCC/ml}$, respectively. The graph 2 and 3 presented the average number of cell size $11,63 \mu\text{m}$ and $15,91 \mu\text{m}$, respectively. PMNs can be recruited and increase milk SCC when the infection continues (Li, N., Richouxet al., 2014)

Conclusion

Somatic cells are secreted in milk during the normal lactation and are used as an index for estimating mammary health and milk quality of dairy animals worldwide. This research may help to understand the importance of implementing good hygienic standards on the farm, to reduce milk somatic cell counts (SCCs) and to establish differential SCC standards. Farmers should use California mastitis test to prevent infection, because high milk SCC affect the quality of the milk and shelf life of the dairy products. Further, managing the SCC on farm will help to improve quality and quantity of milk.

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