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DETERMINATION OF RAW MILK QUALITY BY TOTAL BACTERIAL COUNT AND SOMATIC CELL COUNT IN DIFFERENT REGIONS IN REPUBLIC OF KOSOVO

SUMMARY

The aim of this research was to determine the quality of raw milk by total bacterial count (TB) and somatic cell count (SCC) in the Republic of Kosovo. This research was carried out in a dairy plan in the territory of the Republic of Kosovo. The raw milk processed in the dairy was used for analysis during the research period. Raw milk samples were taken by every producer who delivered milk at the dairy for a period of four months (January to April). Raw milk samples came from five different areas of the territory of the Republic of Kosovo. In total 90 raw milk samples were analyzed. During the research it is determined that the quality of raw milk deviates in terms of the regulations requirements for milk quality. The average number of somatic cell counts was 657,055 cells/ml and the total bacterial count was 2,626,616 CFU/ml of the total number of the analyzed samples. There were not statistically significant differences of the raw milk of the five examined regions.

Keyword: raw milk, Kosovo, somatic cells, total bacterial count

INTRODUCTION

Milk must be specific to human consumption i.e. come from wellnourished healthy lactating animals. This means that the milk of infected animals (resulting from inflammation of the udder), undergoing veterinary treatment is excluded. Milk must be maintained at a temperature of 4 °C or below during all operations (Kohler, 2013), and because it is a perishable product transporters must ensure proper handling and hygiene (Guetouache *et al.*, 2014). Milk is sterile during secretion in the alveoli of the udder. Its contamination with

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microorganisms starts during and after milking (Tamime, 2009). The first contamination of milk with bacteria occurs in the teat canal. Just passing through the channel, the milk is contaminated with 100 - 1,000 CFU (Colony Forming Units)/ml. Microorganisms in raw milk originate from different types of contamination, such as: the udder of the cows, the surface of the cows, from the air as well as from the surfaces of the equipment with which the milk is in contact during milking, storage and transport (Bytyqi *et al.*, 2011). Contamination can be with thermophilic or mesophilic microorganisms. Milk is an excellent environment for the development of such microorganisms, especially mesophilic ones, whose reproduction depends on temperature. So their number can double every 15 minutes at a temperature of $30-37^{\circ}$ C (Alcázar, 2011). Milk is a good medium for the growth of many microorganisms due to its high water activity, near-neutral pH, and available nutrients (Frank, 2007).

The microbiological quality of raw milk can be affected by several factors, such as milking, type of farm (conventional, organic) and season of the year (Bogdanovicova K, *et al.*, 2016). Other factors that have an influence on the microbiological quality are the type of nutrition of the animals, the stage of lactation as well as the way of keeping the animal, i.e. the environment in which they live (Dunge, 2016). The type of microorganisms present in raw milk depends on the temperature and time of storage, as well as the procedure with the milk during and after milking (Tamime, 2009).

The total number of somatic cells count (SCC) in milk is a significant indicator of the health of the mammary udder. In addition, somatic cells are a good diagnostic tool that allows early detection of the subclinical or acute form of mastitis (Green *et al.*, 2004). That is why they represent a significant component in the monitoring program at the farm level (Schukken *et al.*, 2003). The number of somatic cells in the milk is an indicator of the health status of the mammary gland in cows, an indicator of the stability of the milk and its quality for consumption, as well as an indicator of the losses that producers have during mastitis (Brahma *et al.*, 2017). A high number of SCC reduces the durability of milk, and negatively affects the sensory characteristics of the final products (Bytyqi *et al.*, 2011). Also, SCC are correlated with subclinical mastitis and changes in physicochemical properties in raw milk (decreased milk quantity, changes to milk consistency (density), decreased the percentage of protein). Additionally, a higher number of SCC can lead to worsening the milk hygiene and even contain pathogenic microorganisms (Toepel., 2004).

Raw milk should be analyzed in order to identify potential deviations of physicochemical properties and microbiological contamination. Microbiological contamination during milking and after milking is a huge problem in the dairy industry. Some microorganisms can reduce the quality of the milk and can be responsible for milk spoilage, and affect the shelf life of raw milk (Kadriu, 2021).

The first step for controlling the quality of raw milk should be done on the farm level. Farmers are the ones who need to improve the conditions of keeping, feeding and milking the cows in order to obtain quality and safe raw material for

processing (Tamime, 2009). Additional control should be followed by the dairy industry as well as by government institutions in order to improve the current situation in the Republic of Kosovo (Kadriu, 2021).

MATERIAL AND METHODS

This research was carried out in a dairy plan in the territory of the Republic of Kosovo. The raw milk processed in the dairy was used for analysis during the research period. Raw milk samples were taken by every producer who delivered milk at the dairy for a period of four months (January to April). Raw milk samples came from different areas of the territory of the Republic of Kosovo (region 1 - Mitrovica, region 2 - Pristina, region 3 - Gnjilane, region 4 - Pec, and region 5 - Prizren). In total 90 raw milk samples were analyzed 27 samples were analyzed from region 1, i.e. 30% of the total number of samples, 13 samples (14.44%) were analyzed from region 2, 21 samples (23.33%) were analyzed from region 3, and 21 samples (23.33%) were analyzed from region 4.19 samples (21.11%) and 10 samples were analyzed from region 5, i.e. 11.11% of the total number of analyzed samples.

The total number of microorganisms was examined using the BactoScan TM FC - which counted the bacterial colonies in the milk (CFU). The instrument works on the principle of fluorescence flow cytometry according to the ISO 21187 standard. The number of somatic cells was examined using the Fossomatic TM Minor. The instrument works on the principle of the fluoro-opto-electronic method according to the standard ISO 13366-2.

The obtained results were compared with official quality regulations for raw milk in Kosovo (Table 1) (MAFRD., 2006).

Standard fo	or Extra Class	I Class	II Class	III Class
fresh milk				
CFU/M1	<80,000	<100,000	<200,000	<500,000
SCC/M1	<300,000	<400,000	<500,000	<600,000

Table 1. Milk quality standard in Kosovo (CFU/mL and SCC/mL in milk).

Additionally, the number of somatic cell counts was categorized into four categories in order to see how many samples meet the regulations criteria.

-Category I ($\leq 200,000$ cells/ml)

- Category II (from 200,001 400,000 cells/ml)
- Category III (from 400,001 600,000 cells/ml)
- Category IV (≥ 600,001 cells/ml)

RESULTS AND DISCUSSION

From the data presented in Table 2, we can determine that the average number of the somatic cell count, in the examined samples, varies in the range from 414,383 SCC/ml (region 2) to 783,052 SCC/ml (region 4), with an average

value of 657,055 SCC/ml. In all five regions the average number of SCC, do not meet the requirements according to the Rulebook for determining the specific requirements of hygiene of food of animal origin (Official Gazette R. Kosovo 12/2011). Similar results were presented by the Food and Veterinary Agency of Kosovo where for 2007 year, 57.31% of samples had lower than or equal to 400,000 SCC/ml and on the other hand 29.44% of the samples have more than 600,000 SCC /ml (Musliu *et al.*, 2009).

Parameter	Region	N	<u>x</u>	SD	Min	Max
	1	27	713,777	370,521	78,000	1,499,000
	2	13	414,384	436,251	77,000	1,616,000
800/ml	3	21	581,238	1,587,920	8,000	7,443,000
SCC/ml	4	19	783,052	913,432	192,000	2,802,000
	5	10	739,200	726,330	78,000	2,691,000
	Total	90	657,055	932,507	8,000	7,443,000

Table 2: Changes of SCC/ml in raw milk according to the studied regions

Even more, when we look at the variations of the minimum and maximum values of the number of somatic cells, which range from 8,000 SCC/ml to 7,443,000 SCC/ml, respectively, they point to the fact that there are huge differences between regions, but also in the regions themselves. Even if the average number of SCC in the raw milk is 370,521 SCC/ml (region 1) the minimum and maximum number of somatic cells ranges from 78,000 SCC/ml to 1,499,000 SCC/ml.

According to the obtained data on the number of somatic cells, there are also data on the number of CFU/ml, where it is clearly seen that the average number of CFU/ml for all regions in the examined period was 2,616,616 CFU/ml (table 3). Variations of the same parameter ranged from 1,365,071 CFU/ml (region 4) to 3,803,000 CFU/ml (region 2). Such results are not in accordance with the Rulebook establishing the specific requirements of hygiene of food of animal origin (Official Gazette R.Kosovo 12/2011) for the hygienic quality of the processed milk. These data indicate poor hygiene during milking, also improper milk storage, as well as a higher prevalence of subclinical mastitis.

Parameter	Region	Ν	<u>x</u>	SD	Min	Max
CFU/ml	1	25	3,363,520	9,022,032	70,000	45,742,000
	2	12	3,803,000	4,164,898	124,000	14,791,000
	3	14	2,144,642	1,946,187	126,000	5,674,000
	4	14	1,365,071	2,117,198	85,000	5,299,000
	5	8	1,519,125	1,727,863	90,000	4,574,000
	Total	73	2,616,616	5,698,770	70,000	45,742,000

Table 3: Changes of CFU/ml in raw milk according to the studied regions

In general, if we consider the quality of milk obtained in the territory of the Republic of Kosovo during a period of four months, it can be noted that in terms of hygienic quality, the obtained results exceed the requirements according to the Regulation (Standards for quality and categorization of raw milk, 20/2006). According to Bytyqi et al., (2014) about 29.6% of the analyzed milk samples belong to extra class milk (SCC/mL<300.000), followed by milk quality class IIIrd (24.3%), I-st (8.5%) and II-nd (8,2%), where the SCC/mL are ranged from >300.000 till <600.000 for the respective year. On contrary of the total number of analyzed bulk milk samples, 29.5% samples do not fulfill the milk quality standards, poor quality (SCC/mL >600.000). Additionally, Uka E et al., (2018) analyzed 11,482 milk samples, in 2017 where 35% from the samples belong to extra class milk (CFU<80,000) and 46% to III-rd class (CFU<500,000). Similar results where presented in 2018, where 33% from the analyzed 8,795 milk samples were classified as extra milk class, and 44% to III-rd class. Different results were presented by Bytyqi et al., (2010) where in their research for 2008 and 2009, the requirements for extra milk (in terms of SCC/ml) meet 78.45%, and 70.88%, from farms, respectively. They indicate that there are significant differences between the examined 13 farms in the territory of the Republic of Kosovo in terms of the hygienic quality of the milk. According to the results presented by Visciano & Schirone (2022) who analyzed milk samples in Italy (Pescara and Teramo) only 23 (9.6%) out of 240 samples exceeded the regulatory limit for SCC, and the maximum found value was 2,351,000 cells/mL.

On the other hand, Bytyqi *et al.*, (2011) in their research analyzed 70% of raw milk processed in 14 largest dairies in Kosovo. According to their results for 2008 only 20% of the milk producers in Kosovo meet the requirements and around 10% meet the standards given in the Regulations for the following calendar year 2009 (CFU<100,000 CFU/ml). Regarding the total somatic cell count, 80% of the raw milk samples met the 2008 Regulations in Kosovo (SCC<600,000 cells/ml) and 66% met the 2009 Regulations (SCC<400,000 cells/ml).

From the total number of analyzed samples for (N=90), 20 samples, i.e. 22.22% refer to samples where the number of somatic cells is up to 200,000 cells/ml, while 21 samples refer to the number of somatic cells from 200,001 to 400,000 cells/ml, i.e. 23.33% (table 4) or about 45% of the tested samples meet the standards for hygienic quality of the milk. Similar results were presented by Bytyqi *et al.*, (2011) and Bytyqi *et al.*, (2010). Statistically significant results were obtained between the fourth category with the other three categories (p<0.01). However, these data, as well as those from previous studies, point to the fact that almost half of the dairy animals do not show signs of inflammation of the udder, but as an additional problem are the poor hygienic conditions during milking, as well as the inadequate storage and transportation of the raw milk to the dairy is not standardized. In addition, not only the means of transport are under different ownership, the transport also differs, that is, a total

of six different types of milk transport are defined in the territory of the Republic of Kosovo. For each transport system, it is specifically defined where and when the milk sample should be taken (Bytyqi *et al.*, 2011).

Table	4:	Changes	in	the	hygienic	quality	of	raw	milk	shown	by	categories
accord	ing	to the nu	mb	er of	somatic c	ell coun	t					

Parameter	Category of SCC	N	<u>x</u>	SD	Min	Max
	Ι	20	91,800 ^a	58,250	8,000	192,000
SCC/ml	II	21	289,428 ^a	67,284	201,000	377,000
	III	17	470,647 ^a	34,660	415,000	519,000
	IV	32	1,350,625 ^b	1,295,216	623,000	7,443,000
	Total	90	657,055	932,507	8,000	7,443,000

*Differences of values with different superscripts in the same group are statistically significant at the level: a:b p<0.05

Table 5 shows the total number of microorganisms, by category, according to the number of somatic cells. The obtained results point to interesting data where it can be seen that in the first category (\leq 200,000 SCC/ml) we have an extremely high total number of microorganisms 1,978,333 CFU/ml, which indicates the poor hygienic conditions in which raw milk is obtained and stored. The total number of microorganisms also increases in the other two categories (the second and the third). In addition, the raw milk that had the highest somatic cell count (\geq 600,000 SCC/ml) also had the highest total bacterial count of 3,151,290 CFU/ml. In addition to the fact that there are variations in this parameter, no significant differences were observed between the examined groups (p>0.05).

Table 5: Changes in CFU/ml in raw milk shown by category according to somatic cell count

Parameter	Category according to SCC	N	<u>x</u>	SD	Min	Max
CFU/ml	Ι	12	1,978,333	2,603,418	124,000	8,671,000
CI U/III	II	18	2,407,944	3,695,364	70,000	14,791,000
	III	12	2,186,666	1,064,303	545,000	3,489,000
	IV	31	3,151,290	8,169,771	78,000	45,742,000
	Total	73	2,616,616	5,698,770	70,000	45,742,000

CONCLUSIONS

The dairy industry needs quality raw milk in order to produce quality dairy products. A large number of factors can influence the quality of milk, among which, the most important component, naturally present, is the number of somatic cells. Due to these reasons the number of somatic cells is used as an indicator of the health condition of the udder and the quality of milk. Furthermore, the increase of the total number of microorganisms can be one additional factor for worsening the hygienic quality of milk.

The quality of the raw milk in Kosovo still is very poor and must be improved. Quality milk production is the ultimate goal and goal of any milk producer. The low quality of milk has a negative impact on all segments of the dairy industry, especially in the fresh milk processing segment. Milk quality is the primary factor and ultimate goal of any milk producers in order to obtain quality milk products. Increased SCC is associated with milk reduction and additionally shorter shelf life of the produced milk products. Only when the primary raw material has good quality, quality milk products will be obtained by milk producers.

During the research it is determined that the quality of raw milk deviates in terms of the regulations requirements for milk quality. The average number of somatic cell count was 657,055 cells/ml and the total number of microorganisms was 2,626,616 CFU/ml of the total number of the analyzed samples (N=90). There were not statistically significant differences of the raw milk of the five examined regions.

Current milk quality standards applied in Kosovo do not meet the EU criteria, therefore, Kosovo farmers should be aware that in the very near future they have to meet the EU milk quality standards in order to remain and compete on the market.

The results obtained in this research will make significant contributions to the scientific community, while they also can have applicative character. In this way, the producers of dairy products and the dairy industry can see the possibilities for mutual cooperation in order to improve the quality of the primary raw material.

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