

Original paper

INFLUENCE OF KEEPING AND MILKING OF COWS ON THE HYGIENIC QUALITY OF MILK

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Abstract

The quality and quantity of milk is significantly influenced by housing conditions, care and feeding of dairy animals. Hygienic correct milk can be obtained if the cows or other dairy cattle are kept in hygienic conditions. The aim of this study was to investigate the effect of housing conditions and milking of cows on the microbiological and chemical quality of milk. We examined six samples of bulk tank milk cows from 6 different farms from the Municipality of Podgorica, where cow milking is done by machines. Chemical parameters of milk - fat, protein, lactose, free fat dry matter and somatic cells were investigated on the device Combi-Foss (Foss Electric, Denmark), and the total number of bacteria on the appliance BactoScan. All obtained data were processed using modern variation statistics. Statistical parameters: mean value, standard deviation, maximum and minimum values, are calculated. In order to assess the hygienic conditions of keeping and milking of cows at these farms a certain building, microclimate and hygienic parameters were studied - by modified method of Hristov and Reljić (2009). Based on the established state, each test parameter is rated from 0 to 5 points. Average value of the fat content in bulk tank milk was 3.39 % - ranged from 2.92 % to 3.94 %, protein content 3.13% - ranged from 2.78 % to 3.65 %, lactose 4.27% - ranged from 4.14 % to 4.47%, the fat free dry substance 8.13 % - ranged from 7.75 % to 8.77 %. The value of the number of somatic cells in bulk tank milk at the first farm was 52000/mL, second 85000/mL, third 63000/mL, fourth 1920000/mL, fifth 1373000/mL and sixth 200000/mL. The total bacteria count in the bulk tank milk at the first farm amounted to 5000/mL, second 41000/mL, third 124000/mL, fourth 1002000/mL, fifth 467000/mL and sixth 31000/mL. Physical, microclimate and hygienic parameters of dairy cows housing are rated with good rating on farms 1, 2, 3, 4 and 6 and on the farm 5 with a sufficient rating. The fourth and fifth farms did not use disinfection of teats after milking and disinfection of milking machines. The fourth and fifth farm had significantly higher number of somatic cells in bulk tank milk. The fifth farm had significantly higher total number of bacteria/mL in bulk tank milk - compared to the number that is permitted under applicable regulations. This can be linked with inadequate building, microclimate and hygiene of cows and failing disinfection of the udder and milking machine after milking.

Key words: *hygiene, milk, milking, somatic cells, total bacteria count*

Introduction

Milk is a food of high nutritional value only if it is obtained from healthy animals. Proper breeding, keeping and exploitation of dairy animals provide a healthy product and production of hygienic proper milk. The main sources of milk contamination by microorganisms are diseased udders, udder skin, air, equipment for milking and storage of milk, and man. When leaving the mammary glands of healthy animals the milk contains a small number of non-pathogenic microorganisms - to 300/mL originating from teat canal. Postsecretory level of contamination of milk depends on the way and milking hygiene and procedure with milk after milking. The number of somatic cells in milk of cows directly indicates the state of health of the mammary gland. The number of somatic cells in milk is influenced by numerous factors, such as factors that cause mastitis (pathogen microorganisms, toxins), physiological and pharmacological factors (stage of lactation, breed), stressful factors (changes in food, transportation, housing conditions, method of milking). Healthy mammary glands of milk contain polymorphonuclear leucocytes, macrophages, lymphocytes, erythrocytes and epithelial cells. In the case of mastitis, the number of polymorphonuclear leukocytes is increased and may be up to 100 % of the total cells in milk. The average number of somatic cells in milk from healthy udders is 50 000/mL and in most cases it is less than 150 000/mL (Katić et al., 2003.). Changes in the composition of milk caused by mastitis affect its suitability for technological processing. Milk of cows with mastitis has a reduced percentage of free fat dry matter, casein, lactose, sodium, calcium, magnesium and an increased amount of chloride, serumalbumines, serumglobulines, and increased pH. In the production of cheese from milk with high somatic cell count a coagulation time is prolonged while the yield and sustainability of cheese is reduced.

Given the importance of production proper hygienic milk in human nutrition, the aim of our study was to investigate the effect of keeping and milking hygiene of cows on the microbiological, chemical quality and the number of somatic cells in milk.

Materials and methods

We have examined six bulk tanks milk samples of cows from six farms of the Municipality of Podgorica, where milking cows is done by machines. Milk samples were collected in sterile plastic containers with an added preservative. Immediately after taking, the samples were placed in the hand refrigerator at a temperature of 4-5⁰C and transported to the laboratory in 12 hours maximally. Milk samples were tested for the following parameters: fat content, protein content, lactose content, free fat dry matter content, number of somatic cells and the total number of bacteria. Determination of chemical parameters and the number of somatic cells in bulk tank milk of cows was done on the machine Combi - Foss (Foss Electric, Denmark) and the total number of bacteria on the appliance BactoScan. All data were analyzed using modern variation statistics. The following statistical parameters were calculated: mean value, standard deviation, maximum value and minimum values.

In order to assess the hygienic conditions of keeping and milking the cows were determined visually by some space, microclimate and hygienic parameters, using modified method of Hristov and Reljić (2009).

Rating of spatial conditions was performed by measuring the dimensions of stables, bearings, windows, doors and outlets. We have taken into account the position of the stables, the type

and condition of ventilation, as well as the characteristics of the material for the construction of walls, ceilings and floors, their current status of functionality and possible damage, and the type and amount of litter. Evaluation of sanitation was carried out on the basis of visual assessment of hygiene litter, bearings, floors, walls, ceilings, windows, outlets, feeders, drinkers, channels and manure corridors and ventilation ducts. Assessment of microclimate conditions was performed using the following parameters: dust, noise, ventilation, air circulation, harmful gases and illumination. Based on the established state of each test parameter was evaluated with a minimum of 0 to a maximum 5 points (5 - excellent, 4 - very good, 3 - good, 2 - satisfactory, 1 – insufficient - there are resources to improve, 0 – insufficient - no resources to improve). Dividing the total number of points with the number of estimated parameters we obtained final rating of a minimum 0 to a maximum 5 (evaluation is done as follows: 0 - 1.99 insufficient, 2.00 - 2.49 sufficient, 2.5 - 3.49 good, 3.5 - 4.49 very good and 4.5-5.00 excellent).

Results and discussion

Results of chemical composition of bovine bulk tank milk are shown in Table 1.

Table 1. *The results of examination of chemical and microbiological parameters and the number of somatic cells in bovine bulk tank milk samples*

Farm	Fat content (%)	Protein content (%)	Lactose content (%)	Free fat dry matter (%)	Freezing point	Somatic cells number	Total bacteria number
1	3.32	3.18	4.47	8.38	0.534	52000	5000
2	2.92	2.91	4.14	7.79	0.517	85000	41000
3	3.32	2.96	4.17	7.86	0.525	63000	124000
4	3.65	3.65	4.39	8.77	0.531	1920000	1002000
5	3.19	3.28	4.24	8.25	0.524	1373000	467000
6	3.94	2.78	4.24	7.75	0.514	200000	31000
Average	3.39	3.13	4.27	8.13	0.524	615500	18000
Max	3.94	3.65	4.47	8.77	0.534	1920000	1002000
Min	2.92	2.78	4.14	7.75	0.514	52000	5000
Sd	0.357	0.314	0.128	0.404	0.007	818826.7	18.385

The results in Table 1 show that in the bulk tank milk samples fat content varied from 2.92 % to 3.94 %, protein content from 2.78 % to 3.65 %, and the content of lactose from 4.14 % to 4.47%. The content of free fat dry matter varied from 7.75 % to 8.77 %. Freezing point varied from 0.514 to 0.534 and the number of somatic cells from 52000 to 1920000/mL. The number of bacteria varied from 5000 to 1002000/mL of milk.

The results of evaluation of the stable spatial parameters of cows keeping are shown in Table 2.

Table 2. *Results of evaluation of the stable spatial parameters of cows keeping*

Farm	1	2	3	4	5	6
Stable	4	4	5	3	3	5
Bearing	3	3	4	2	2	4
Hallway nutrition	2	3	3	1	2	4
Feeding equipment	3	4	4	3	3	4
Equipment for drinking	4	4	4	3	4	4
Hallway manure	3	4	3	2	1	3
Channel manure	3	2	3	3	2	3
Outlet	4	3	3	3	4	2
Ventilation	2	3	2	1	0	4
Windows	3	4	3	1	1	3
Doors	3	4	2	1	1	3
Walls	4	3	4	2	2	4
Roof	1	2	2	0	1	2
The total number of points	39	43	42	25	26	45
Evaluation	3.00	3.30	3.23	1.92	2.00	3.46
Descriptive rating	good	good	good	insufficient	sufficient	good

Results in Table 2 show that the evaluation of the spatial parameters were insufficient on farm 4, sufficient on farm 5 and good on other farms: 1, 2, 3 and 6.

The assessment results of microclimate conditions in the stables for keeping the cows are shown in Table 3.

Table 3. *Results of evaluation of microclimate conditions in the stables for cows*

Farm	1	2	3	4	5	6
Dust particles	3	2	3	3	3	3
Noise	4	3	2	4	3	4
The air flow	2	4	3	2	1	3
Hazardous gases	2	2	4	2	2	2
Illumination	2	3	2	3	2	4
The total number of points	13	14	14	14	11	16
Evaluation	2.6	2.8	2.8	2.8	2.2	3.2
Descriptive rating	good	good	good	good	sufficient	good

Rating of microclimate conditions was sufficient on the farm number 5 and good on the other farms.

The assessment results of hygienic parameters in the stables for keeping cows are shown in Table 4.

Table 4. *Results of evaluation of hygiene in stables for cows*

Farm	1	2	3	4	5	6
Hygiene bearings	5	4	4	2	2	4
Hygiene litter	4	3	4	2	1	5
Hygiene drinkers	5	4	5	3	3	4
Hygiene hall for food	3	3	4	3	2	3
Hygiene discharge	3	4	4	4	3	4
Hygiene window	4	3	3	3	2	3
Hygiene walls	3	4	3	3	2	3
Hygiene roof	4	5	3	3	2	4
Total number of points	31	30	30	23	17	30
Evaluation	3.87	3.75	3.75	2.87	2.12	3.75
Descriptive rating	very good	very good	very good	good	sufficient	sufficient

The results presented in Table 4 show that the evaluation of the hygienic conditions in the stables was sufficient on the fifth farm, good on the fourth farm and very good on the other farms. The lowest rating (rating 2) on the fourth farm had parameters for hygiene of bearings and litter. On the fifth farm a litter hygiene was evaluated the lowest rating (rating 1).

The results of total evaluation of accommodation conditions on the farms are shown in Table 5:

Table 5. *Total rating of accommodation conditions on tested farms*

Evaluated parameters	Farm					
	1	2	3	4	5	6
Spatial	3.00	3.30	3.23	1.92	2.00	3.46
Microclimatic	2.60	2.80	2.80	2.80	2.20	3.20
Hygienic	3.87	3.75	3.75	2.87	2.12	3.75
Rating	3.15	3.28	3.26	2.53	2.10	3.47
Descriptive rating	good	good	good	good	sufficient	good

The results presented in Table 5 show that the rating of the accommodation conditions for cows on farms 1, 2, 3, 4 and 6 was good and on the farm 5 insufficient.

The results of implementation of washing and disinfection of udder and milking machines are shown in Table 6:

Table 6. *Test results of implementation of washing and disinfection of udder and milking machines*

Farm	Washing the udder by lukewarm water before milking	Teat disinfection after milking	Washing of milking machines by detergent and lukewarm water	Disinfection of milking machines
1	Yes	Yes	Yes	Yes
2	Yes	Yes	Yes	Yes
3	Yes	Yes	Yes	Yes
4	Yes	No	Yes	No
5	Yes	No	Yes	No
6	Yes	Yes	Yes	Yes

The results in Table 2 show that washing the udder with lukewarm water before milking is enforced on all farms. Washing the milking machines on all six farms was conducted using detergent and lukewarm water. Teat disinfection after milking and disinfection of milking machines is not implemented on the fourth and fifth farm.

The modes of keeping and feeding the dairy animals significantly affect the quality and quantity of obtained milk. Hygienically correct milk can be obtained only if the cows are kept in hygienic conditions. The proper treatment of animals is closely related to their welfare, and to technological, hygienic and other requirements that provide the necessary comfort and hygiene to dairy farm. Raw milk is usually contaminated as a result of mistakes in the implementation of hygiene during milking and storage of milk. Cows suffering from mastitis secrete a large number of bacteria through milk, which is significant source of contamination. Streptococci and staphylococci, as the most common causes of mastitis, are transmitted from animal to animal through milking. Reduction of new infections can be achieved by milking teat disinfection after milking, proper handling of the milking machine, disinfection of teat cups, washing the udder with lukewarm running water and other hygienic – sanitary measures. Teat cup, if the milking technology is poor, may be an important factor in the transmission of mastitis pathogens. Mastitis increases the number of somatic cells, in particular those caused by *Streptococcus agalactiae* and *Staphylococcus aureus* (Katić et al., 2003).

Stable climate, as zoohygienic significant factor, is determined by the characteristics of the roof, walls and floors of the stable. Regular ventilation and outdoor air significantly affect the indoor climate. Construction materials used to build the stable, should be adapted to the geographical area (Stojanović and Katić, 2004). Appropriate microclimate in the stable and cleanliness of floors provides a constant dry and clean udder, which is an important prerequisite for mammary gland health and hygienic quality of milk.

For the preparation of dairy animals for milking, washing teats and udder, which requires lukewarm water and clean cloths for cleaning, must be regularly performed. Improved hygiene

of milking and milking machines can reduce the number of somatic cells in milk. Control of milking machines has great importance in the prevention of new infections in the herd (Hristov et al., 2005). Stable air contains particles of dust, which during hand milking contaminate milk by microorganisms (Katić and Stojanović, 2004). According to Article 87 of the Rule Book on the specific hygiene requirements for food of animal origin, Gazette of Montenegro, no. 14/2009, the allowed number of bacteria in bovine raw milk is 100000/mL and of somatic cells 400000/mL. If the raw milk is used for the manufacture of products from milk, the maximum number of bacteria is 300000/mL milk. With these criteria in mind, bulk tank raw milk originating from farms 3, 4 and 5 does not correspond to the criteria for the total number of bacteria. On the fourth and fifth farm significantly higher number of somatic cells in bovine bulk tank milk was found compared to the number permitted under the Rule Book of the specific hygiene requirements for food of animal origin, Gazette of Montenegro 14/2009 (Table 1).

Katić et al. (1994) determined the mean number of somatic cells in cow milk to be 93361/mL, in the milk of cows with subclinical mastitis challenged with *S. aureus* 182163/mL, in the milk of cows with subclinical mastitis caused by *S. uberis* 2214000/mL and the in milk of cows with nonspecific mastitis 980750/mL. In the bulk tank milk of healthy cows they found the mean fat content of 3.39%, protein 3.37%, lactose 4.81% and free fat dry matter 8.71%. According to the Regulations on the quality and other requirements for milk and milk products and starter cultures (Gazette SRJ 26/2002, 56/2003, 4/2004 and 5/2004) fat content should not be less than 3.3%, protein content 2.9% and free fat dry matter 8.5%. The value of the free fat dry matter content in milk obtained in our tests (8.13 ± 0.404) is below the minimum value of the dry matter prescribed by the regulations. Lactose content obtained in our study on bovine bulk tank milk is $4.27\% \pm 0.128$ which is lower value compared to the lactose content in milk of healthy cows (4.7%) (Table 1).

Rating of spatial parameters was insufficient on farm 4, sufficient on farm 5 and good on other farms: 1, 2, 3 and 6. On the fourth farm, with minimum rating, the following parameters were evaluated: the hall for food, ventilation, windows, doors, roof, air flow, harmful gases, hygiene bearings and hygiene litter (Table 2). On the fifth farm the lowest rating had the parameters: hallway manure, ventilation, windows, doors, roof, air flow and hygiene litter (Table 2). Rating of microclimate conditions was sufficient on farm 5 and good on other farms (Table 3). Rating of hygiene in stables was sufficient on the fifth, good on the fourth and very good on the other farms. The lowest rating (rating 2) on the fourth farm obtained parameters: hygiene of bearings and litter. On the fifth farm the lowest rating was for hygiene of litter (rating 1) (Table 4). Total rating of conditions for accommodation of cows on farms 1, 2, 3, 4 and 6 was good, while the rating on the farm 5 was sufficient (Table 5).

Washing and sanitizing the milking equipment is a critical point in the process of milking, because it affects the level of bacterial contamination of bovine bulk tank milk. Proper cleaning of milking machine provides better milk quality. The residual milk or water after washing the milking machine promote the growth of various microorganisms (Bava et al., 2009). Poor hygiene during milking, with failures in disinfection, particularly of the teats and hand milker, facilitate milk contamination from the environment (Hristov et al., 2005.). If hygiene is not maintained, milking machines are always the source of milk contamination. The main sources of contamination from the milking machine are: teat cups, hoses, main

sewer and dairy milk container. Milk is usually contaminated by psychrotrophic gram negative organisms from equipment for milking,

From the equipment the milk can also become contaminated by the sporogenic aerobic bacteria of genus *Bacillus* and the sporogenic anaerobic bacteria of genus *Clostridium*. A common source of bacterial contamination of equipment is unfit water. Proper sanitation of milking machines and their proper application produces milk with a small number of microorganisms (Stojanović and Katić, 2004). Irregular, rough machining or hand-milking has a significant role in the incidence of mastitis.

Improper preparation of the udder for milking leads to transferring the causes of mastitis from cow to cow. In our tests, washing the udder with warm water before milking was enforced on all farms. Washing the milking machine on all six farms was conducted using detergent and warm water. However, teat disinfection after milking and disinfection of milking machine was not conducted on the fourth and fifth farm (Table 6). In view of these results, the high number of somatic cells and microorganisms in bulk tank milk of cows on farms 4 and 5 can be related to inadequate space, microclimate and sanitation of accommodation conditions for cows, non-implementation of disinfection of the udder after milking and non-implementation of disinfection of milking machine.

Conclusion

In bulk tank cow milk samples fat content ranged from 2.92% to 3.94%, protein content from 2.78% to 3.65 % and content of lactose from 4.14% to 4.47%. The content of free fat dry matter varied from 7.75% to 8.77%. Somatic cells count ranged from 52000 to 1920000/mL and number of bacteria from 5000 to 1002000/mL milk. Total rating of conditions for accommodation of cows on farms 1, 2, 3, 4 and 6 was good, and on the farm 5 was sufficient (Table 5). The high number of somatic cells and microorganisms in bulk tank milk of cows on farms 4 and 5 may be associated with inadequate space, microclimate and hygiene conditions of accommodation, as well as by failed teat disinfection after milking and failed disinfection of milking machine.

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References

1. Bava L, Zucali M, Brasca M, Zanini L and Sandrucci A 2009. Efficiency of cleaning procedure of milking equipment and bacterial quality of milk. *Ital. J. Anim. Sci.* 8 (Suppl. 2), 387-389.

2. Hristov S, Stanković B and Relić R 2005. Klinički i subklinički mastitis u krava. *Biotechnology in Animal Husbandry* 21, 1-2, 29-39.
3. Hristov S and Relić R 2009. Ocena uslova smeštaja sa osvrtom na dobrobit krava. *Zbornik naučnih radova* 15, 3-4, 79-88.
4. Katić V, Babić Lj, Popović J and Tayeb H. 1994. Uticaj mastitisa na kvalitet mleka, *Veterinarski glasnik* 48, 3-4, 271-276.
5. Katić V, Stojanović L and Bojanić M 2003. Uticaj nespecifičnih uzročnika mastitisa na broj somatskih ćelija u mlijeku. Peto savjetovanje iz kliničke patologije i terapije, Budva, 9-13. jun. *Clinica veterinaria*, 177-182 pp.
6. Regulations on the quality and other requirements for milk and milk products and starter cultures (Gazette SRJ 26/2002, 56/2003, 4/2004 and 5/2004).
7. Rule book of the specific hygiene requirements for food of animal origin, Gazette of Montenegro, 14/2009.
8. Stojanović L and Katić V 2004. Higijena mleka, Arandelovac.