

## Total bacterial count and somatic cell count in refrigerated raw milk stored in communal tanks

*Contagem bacteriana total e contagem de células somáticas em leite cru refrigerado armazenado em tanques comunitários*

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### ■ Summary

The current industry demand for dairy products with extended shelf life has resulted in new challenges for milk quality maintenance. The processing of milk with high bacterial counts compromises the quality and performance of industrial products. The study aimed to evaluate the total bacteria counts (TBC) and somatic cell count (SCC) in 768 samples of refrigerated raw milk, from 32 communal tanks. Samples were collected in the first quarter of 2010, 2011, 2012 and 2013 and analyzed by the Laboratory of Milk Quality – LQL. Results showed that 62.5%, 37.5%, 15.6% and 27.1% of the means for TBC in 2010, 2011, 2012 and 2013, respectively, were above the values established by legislation. However, we observed a significant reduction in the levels of total bacterial count (TBC) in the studied periods. For somatic cell count, 100% of the means indicated values below 600.000 cells/mL, complying with the actual Brazilian legislation. The values found for the somatic cell count suggests the adoption of effective measures for the sanitary control of the herd. However, the results must be considered with caution as it highlights the need for quality improvements of the raw material until it achieves reliable results effectively.

**Key words:** Normative instruction; Legislation; Nonconformity; Microbiological quality.

### ■ Resumo

A atual demanda da indústria por produtos lácteos com *shelf life* prolongado tem resultado em novos desafios para a manutenção da qualidade microbiológica do leite, pois o processamento da matéria-prima com altas contagens microbianas compromete a qualidade e o rendimento industrial de seus derivados. O estudo objetivou avaliar a contagem bacteriana total (CBT) e a contagem de células somáticas (CSS) de 768 amostras de leite cru refrigerado provenientes de 32 tanques comunitários. As amostras foram coletadas no primeiro trimestre dos anos de 2010, 2011, 2012 e 2013 e analisadas pelo Laboratório de Qualidade do Leite. Os resultados apontaram que 62,5%, 37,5%, 15,6% e 28,1% das médias para a CBT em 2010, 2011, 2012 e 2013, respectivamente, estavam acima dos valores preconizados pela legislação. Entretanto observou-se uma redução significativa nos níveis de contagem bacteriana total (CBT) nos períodos analisados. Para a contagem de células somáticas, 100% das médias apontaram valores abaixo de 600 mil células/mL, estando essa contagem de acordo com as legislações vigentes no Brasil. Portanto, os valores encontrados para a contagem de células somáticas sugerem a adoção de medidas efetivas de controle sanitário do rebanho. Porém, os resultados obtidos para a contagem bacteriana total devem ser considerados com cautela, uma vez que eles indicam a necessidade de melhoria da qualidade da matéria-prima, até que se atinjam resultados efetivamente confiáveis.

**Palavras-chave:** Instrução normativa; Legislação; Não conformidade; Qualidade microbiológica.

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### 1 Introduction

Through the technical regulation of identity and quality of refrigerated raw milk, the Brazilian legislation provides quality microbiological standards that indicate the hygienic conditions of processed milk. In establishments under federal inspection, the entire volume of milk began to be analyzed by the Brazilian Network for Milk Quality (BNMQ) laboratories, credentialed by the Ministry of Agriculture (BRASIL, 2011).

An important resource for monitoring the microbiological quality of refrigerated raw milk is the somatic cell count (SCC), whose function is to combat bacteria which cause mastitis and possible economic losses arising there from (DONG et al., 2012). All cells in milk and those originating from both apoptotic desquamation of alveoli, as those removed during the flow of the bloodstream, are considered somatic cells (GAIATO et al., 2012).

Another measure that allows knowing the concentration of microorganisms in milk is the total bacterial count (TBC) that assists the evaluation of milking and storage procedures in the rural property and, at the same time, allows us to infer the likely adverse effects on industrial productivity and safety of the milk (GARGOURI et al., 2013; SILVA, 2011).

The objective of this study was to verify the characteristics related to the microbiological quality through somatic cell count and total bacterial count of refrigerated raw milk stored in communal tanks for industrial processing.

### 2 Material and methods

Refrigerated raw milk was collected in 32 communal tanks, from rural properties at Colorado do Oeste, Rondônia, Brazil. The collection procedure consisted of milk homogenization, through activation of the tank agitator for 5 minutes, followed by the collection and transference of the samples to appropriate containers, which were sent to the Milk Quality Laboratory, at the Federal University of Goiás (CPA, 2010).

Data collection refers to samples analyzed during the months of January, February and March, in 2010, 2011, 2012 and 2013, period classified as rainy (October to April) (RONDÔNIA, 2007). Two samples of milk were collected every month, once for CBT and one for CCS. Therefore, in the quarter, six samples per tank were analyzed, allowing the calculation of the geometric mean, according to the methodology suggested by the Normative Instruction n° 51 (NI-51) and the Normative Instruction n° 62 (NI-62) (BRASIL, 2002; 2011).

Table 1 shows the amount of communal tanks of the milk suppliers and the geometric mean size used to infer the results.

Results of the microbiological quality presented in this study were extracted from reports issued by LQL for a dairy industry inserted in the Federal Inspection System (FIS), at Colorado do Oeste, Rondônia, Brazil.

According to information provided by the laboratory LQL (CPA, 2010), TBC analyses were performed using the equipment BactoScan FC®, with a capacity of 150 samples/hour. For SCC analysis, the Fossomatic 5000 Basic®, equipment was used, with capacity of 300 samples/hour. Both devices are based on the analytical principle of flow cytometry.

We start from the following premise in assessing the microbiological quality of refrigerated raw milk:

- The collected sample is suitable for industrial processing if the results of the geometric mean for SCC and TBC are up to 1.000.000 cells/mL for samples of 2010; 750.000 cells/mL for samples of 2011 and 2012, and 600.000 cells/mL for samples of 2013.
- If the samples are above the specified value, it will be considered unsuitable for processing and/or for marketing purposes (BRASIL, 2002; 2011).

The research project was approved by the Ethics Committee in Research of the Federal Institute of Education, Science and Technology of Rondônia, Brazil.

### 3 Results and discussion

The results indicated that, in the studied period, milk TBC showed geometric mean in disagreement with the legislation (Table 2).

The microbiological content for TBC of refrigerated raw milk was above the maximum permissible limit

**Table 1.** Sample data and geometric mean of milk suppliers in Colorado do Oeste in the first quarter of 2010 - 2013.

Amount of tanks	Geometric mean of the quarter	Total of analyzed samples
32	128	768

**Table 2.** Percentage results of geometric means (n=128) of refrigerated raw milk for TBC in the first quarter of 2010 - 2013 (Colorado do Oeste, RO).

First quarter (Jan. Feb. Mar.)	TBC (cells/mL)			
	≤ 600 k	> 750 k	> 1000 k	> 10.000 k
2010	25% (8)	12.5% (4)	50% (16)	12.5% (4)
2011	62.5% (20)	6.25% (2)	31.2% (10)	-
2012	84.3% (27)	3.1% (1)	12.5% (4)	-
2013	69.6% (23)	18.1% (6)	9.0% (3)	-

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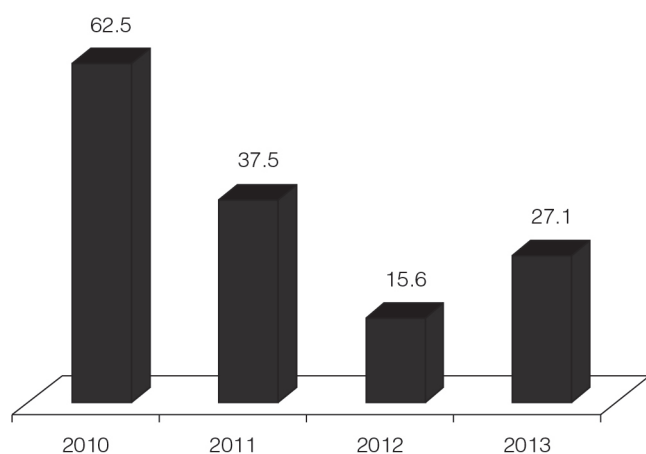
(Table 2), that is  $1.0 \times 10^6$  cells/mL for 2010;  $7.5 \times 10^5$  cells/mL for 2011 and 2012 and  $6.0 \times 10^6$  cells/mL for 2013 (BRASIL, 2002; 2011).

Considering the values of geometric means for total bacterial counts, 20 (62.5%) samples had counts exceeding 1.000.000 cells/mL in 2010; 12 (37.5%) and 5 (15.6%) samples were higher than 750.000 cells/mL in 2011 and 2012, respectively; and 9 (27.1%) samples were above 600.000 cells/mL in 2013 (Table 2), considering the maximum limits established by legislation (BRASIL, 2002; 2011).

In several studies on the incidence of TBC in milk, it was observed that high microbial counts are sourced from disability in washing and sanitizing of the equipments, milking utensils and inadequate cooling system (TRONCO, 2008). In Lacerda et al. (2010), high bacterial counts were due to failure in handling, especially with regard to hygiene and health care in milking. Another explanation for high values of TBC is the season, which influences the feeding of the herd and milk production, interfering with the bacterial content (HOOPERHEIDE and MATTIODA, 2012; PAIVA et al., 2012). This aspect can be seen in a study about the quality of refrigerated raw milk in Ouro Preto do Oeste, Rondônia, Brazil, showing that the mean TBC in March, a period considered rainy (October-March), of 1.891.000 cells/mL, was higher than the average of July, a period considered dry, of 760.000 cells/mL (ALMEIDA, 2010).

According to the results, there were significant reductions in the levels of TBC of refrigerated raw milk; however, values in disagreement with the legislation were found (Figure 1).

The values in disagreement to TBC found in 2010 (62.5%, Figure 1) is a serious problem for the industry regarding to quality, especially in the results above



**Figure 1.** Percentage of quarterly geometric means (from 2010 to 2013; n=128) in disagreement with the legislation, for total bacterial count of refrigerated raw milk at Colorado do Oeste.

10.000 cells/mL. Buying milk with high bacterial counts, the company may be putting at risk the safety of dairy products produced with this type of raw material. TBC values above the reference parameters of the legislation are among the major deficiencies in microbiological quality of milk as stated by Ponsano et al. (2011), based on studies on training farmers to improve the quality of raw milk produced in Araçatuba, SP. Then, it becomes evident the need to adopt control measures to improve the microbiological quality of milk before submitting it to industrial processing.

It is observed that over the study period (Figure 1) there were reductions of TBC in compliance with the legislation in 2011 and 2012, with a slight increase in 2013. This result is alarming, since in the second half of 2014 the limit for the TBC must present 300.000 cells/mL. Increased TBC in the first quarter of 2013 can be explained by the gradual reduction of the maximum allowable values for the microbiological requirements of milk (BRASIL, 2011).

Although the percentage of TBC samples in disagreement with the legislation is high, it is noted (Figure 1) that, over the analyzed period, there were reductions of TBC, in compliance with the Brazilian legislation (BRASIL, 2002; 2011) with a slight increase in 2013. Studies by Paiva et al. (2012) showed an annual quality evolution of refrigerated raw milk with a reduction rate of TBC due to technical guidance to farmers. However, Ponsano et al. (2011) warns that it is necessary a greater time working together with farmers so that they may assimilate information and put them into practice, until improvements in milk quality are met.

Therefore, in this research, one of the factors that may have contributed to improvements in the levels of TBC are the intensification of technical orientation performed by the milk collection companies in line with the requirements of Brazilian legislation, which gradually is being implemented throughout the national territory (BRASIL, 2002; 2011).

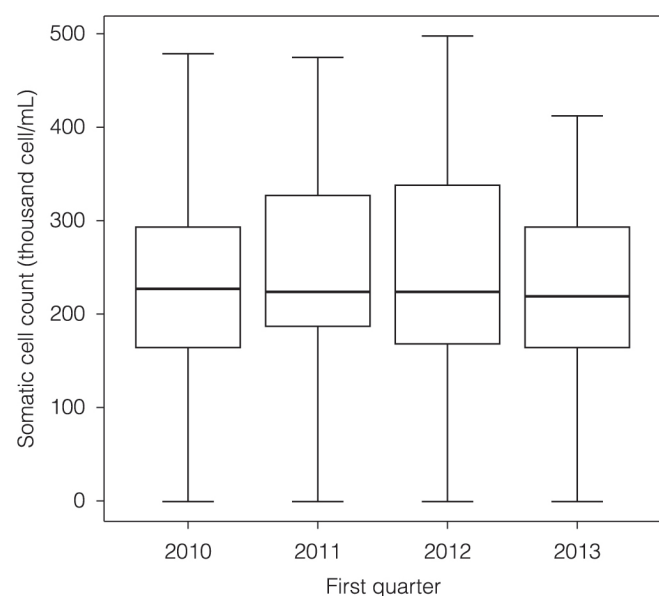
For somatic cell count, no statistical differences between the studied periods (Figure 2) occurred. In Figure 2, the SCC had amplitude ranging from 53.000 to 483.000 cells/mL, results compliant to the legislation (BRASIL, 2002; 2011).

Regarding the description of SCC variable, on average, samples showed 346.000 cells/mL, with a standard deviation of 93.33. The SCC variable had a coefficient of variation of 27%, showing an average variability (FONSECA and MARTINS, 1996).

Similarly to the results of this survey, Silva (2011) found mean values below 600.000 cells/mL in the first six months of 2007, 2008 and 2009 in refrigerated raw milk stored in communal tanks of northeastern Brazil, concluding that the elevation of SCC in milk (up to 200.000 cells/mL), which indicates the occurrence of

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**Figure 2.** Box plot of the geometric mean for somatic cell count of refrigerated raw milk in the first quarter of 2010 – 2013 (Colorado do Oeste).

mastitis, affects the composition of the milk and the shelf-life of products. Hortet et al. (1999) found losses of 0.30 kg of milk, which occurred for 100.000 cells/mL, and 0.61 kg, for 200.000 cells/mL. In addition, with the increase in SCC, milk composition, enzymatic activity, clotting time, productivity and quality of dairy products, are negatively influenced (KITCHEN, 1981).

Considering the results in accordance to the SCC, it was expected a better result for TBC, since the practices that prevent mastitis are largely the same that prevent bacterial contamination of the milk. However, it must be considered that crossbreed and low production animals are less susceptible to mastitis, which most often affects purebreds and high production animals (BELOTI et al., 2011).

Further, according to the results shown in Figure 2, it is observed that the values for somatic cell showed a similar behavior. These results also indicate that milk somatic cells varied slightly depending on the period studied. Corroborating this behavior Mesquita et al. (2008), researching the quality of milk in the central west and northern Brazil in January 2007 to July 2008 found that the geometric mean for SCC (300.000 CFU/mL) had no significant change over the period. With the results obtained in this research (Figure 2), milk suppliers would not have difficulties to meet the parameters set by legislation (BRASIL, 2002; 2011).

## 4 Conclusions

The high percentages for total bacterial count in milk indicate failures in milking hygiene procedures and/or storage that affect the quality of the raw material. However,

there were reductions of TBC over the studied period probably due to the training of farmers in accordance with the requirements of the legislation.

Although all parameters of somatic cell counts were within the microbiological standards established by legislation, this result must be considered with caution as it highlights the need for improvements in the quality of the raw material until it achieves reliable results effectively.

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