

**QUALITY PARAMETERS OF REFRIGERATED RAW MILK PRODUCED IN THE
NORTHERN REGION OF ESPÍRITO SANTO, BRAZIL**

**PARÂMETROS DE QUALIDADE DE LEITE CRU REFRIGERADO PRODUZIDO
NA REGIÃO NORTE DO ESPÍRITO SANTO, BRASIL**

**PARÁMETROS DE CALIDAD DE LA LECHE CRUDA REFRIGERADA
PRODUCIDA EN LA REGIÓN NORTE DE ESPÍRITO SANTO, BRASIL**

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ABSTRACT

The objective of this study was to evaluate the quality parameters of raw milk samples stored in individual refrigeration tanks on three family farms in the northern region of Espírito Santo. Microbiological quality was assessed by analyzing the total bacterial count (TBC) and psychrotrophic bacteria count (PBC). Proximate composition was also determined using a Lactoscan® device, and somatic cell count (SCC) was measured using a Somaticell® kit. Some characteristics of the farms, the location of the tanks, and the temperature used during storage were assessed. Total bacterial count values were within the legally established limit, and the psychrotrophic bacteria count was below the 10% suggested TBC. The average somatic cell count obtained in samples collected from tank A was 650,000 SC/mL, from tank B it was 500,000 SC/mL, and from tank C it was 250,000 SC/mL. Regarding the centesimal composition, it was observed that the milk samples obtained from tanks A and B presented percentages of fat, solids-non-fat, and total solids below the legally recommended values, while the milk samples from tank C presented rates above or close to the established minimum values. In contrast, the percentages of protein and lactose approached the recommended minimum limits. The data showed that the temperature values recorded in the three refrigeration tanks were in accordance with the legislation. Regarding psychrotrophic bacteria, the values were below those suggested in the literature, but it is necessary to reinforce care and surveillance regarding somatic cells, which are indicative of mastitis in the

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herd. Continued frequent evaluation of the parameters analyzed in other samples will be of great importance, as it will contribute to monitoring possible failures in production and processing and will enable the supply of adequate milk and dairy products, in addition to the sustainability of the sector in the northern region of the state of Espírito Santo, Brazil.

Keywords: Family Farming. Psychrotrophic Bacteria. Somatic Cells. Refrigerated Raw Milk. Quality.

RESUMO

Objetivou-se neste trabalho avaliar os parâmetros de qualidade em amostras de leite cru armazenado em tanques de refrigeração individuais localizados em três propriedades rurais familiares situadas na região norte do Espírito Santo. Para avaliação da qualidade microbiológica foram realizadas análises de contagem bacteriana total (CBT) e a contagem de bactérias psicrotróficas (CBP). Realizou-se também a composição centesimal por meio do aparelho Lactoscan® e contagem de células somáticas (CCS) utilizou-se o Kit Somaticell®. Verificou-se algumas características das propriedades rurais, o local de instalação dos tanques e a temperatura empregada durante o armazenamento. Valores da contagem bacteriana total apresentaram-se no limite estabelecido pela legislação e a contagem de bactérias psicrotróficas ficou abaixo de 10% sugeridos em relação a CBT. A contagem média de células somáticas obtida em amostras coletadas no tanque A foi de 650.000 CS/mL, no tanque B foi de 500.000 CS/mL e no tanque C foi de 250.000 CS/mL. Com relação à composição centesimal, observou-se que as amostras de leite obtidas nos tanques A e B apresentaram percentuais de gorduras, sólidos não gordurosos e sólidos totais abaixo do que é recomendado pela legislação, enquanto as amostras de leite do tanque C apresentaram taxas acima ou próximas aos valores mínimos estabelecidos. Em contrapartida, o percentual de proteína e lactose aproximaram-se dos limites mínimos preconizados. Os dados mostraram que valores da temperatura registrada nos três tanques de refrigeração estavam de acordo com a legislação. Com relação às bactérias psicrotróficas, os valores ficaram abaixo dos sugeridos pela literatura, porém é preciso reforçar o cuidado e a vigilância em relação às células somáticas, indicativo de mastite no rebanho. A continuidade da avaliação frequente dos parâmetros analisados em outras amostras será de grande importância pois contribuirá para monitoramento de possíveis falhas na produção e processamento e permitirá o fornecimento de leite e produtos lácteos adequados, além da sustentabilidade do setor na região norte do estado do Espírito Santo, Brasil.

Palavras-chave: Agricultura Familiar. Bactérias Psicrotróficas. Células Somáticas. Leite Cru Refrigerado. Qualidade.

RESUMEN

El objetivo de este estudio fue evaluar los parámetros de calidad de muestras de leche cruda almacenadas en tanques de refrigeración individuales en tres granjas familiares en la región norte de Espírito Santo. La calidad microbiológica se evaluó mediante el análisis del recuento bacteriano total (TBC) y el recuento de bacterias psicrotróficas (PBC). La composición proximal también se determinó utilizando un dispositivo Lactoscan®, y el recuento de células somáticas (RCS) se midió utilizando un kit Somaticell®. Se evaluaron algunas características de las granjas, la ubicación de los tanques y la temperatura utilizada durante el almacenamiento. Los valores del recuento bacteriano total estuvieron dentro del límite legalmente establecido, y el recuento de bacterias psicrotróficas fue inferior al 10% sugerido de TBC. El recuento promedio de células somáticas obtenido en las muestras colectadas del

tanque A fue de 650,000 SC/mL, del tanque B fue de 500,000 SC/mL, y del tanque C fue de 250,000 SC/mL. En cuanto a la composición centesimal, se observó que las muestras de leche obtenidas de los tanques A y B presentaron porcentajes de grasa, sólidos no grasos y sólidos totales inferiores a los valores recomendados legalmente, mientras que las muestras de leche del tanque C presentaron valores superiores o cercanos a los valores mínimos establecidos. Por el contrario, los porcentajes de proteína y lactosa se acercaron a los límites mínimos recomendados. Los datos mostraron que los valores de temperatura registrados en los tres tanques de refrigeración se ajustaron a la legislación. En cuanto a las bacterias psicrótroficas, los valores fueron inferiores a los sugeridos en la literatura, pero es necesario reforzar la vigilancia de las células somáticas, que son indicativas de mastitis en el rebaño. La evaluación frecuente y continua de los parámetros analizados en otras muestras será de gran importancia, ya que contribuirá al monitoreo de posibles fallas en la producción y el procesamiento y permitirá el suministro de leche y productos lácteos adecuados, además de la sostenibilidad del sector en la región norte del estado de Espírito Santo, Brasil.

Palabras clave: Agricultura Familiar. Bacterias Psicrótroficas. Células Somáticas. Leche Cruda Refrigerada. Calidad.

1 INTRODUCTION

Milk is a food of fundamental importance for human health. It has many nutrients available and when the hygiene standards recommended in the production process are neglected, it can provide favorable conditions for microbial multiplication by altering its physicochemical characteristics. It is notorious that its microbiological quality is multifactorial and depends, among others, on the state of health of the mammary gland and the outside of the udder, hygiene of the milking equipment, maintenance and correct functioning of the cooling tank, the quality of the water, as well as the adoption of good practices throughout the production chain, aiming to obtain a product with quality and safety for the final consumer (TEIXEIRA et al., 2018; COSTA, 2006). And if after milking the milk is not in adequate storage and refrigeration conditions, there is the possibility of the development and multiplication of undesirable microorganisms, making it unsuitable for consumption (OLIVEIRA, MARCHIORE, 2017).

According to the Regulation of Industrial and Sanitary Inspection of Products of Animal Origin (RIISPOA), milk is the product of complete, uninterrupted milking, in hygienic conditions, of healthy, well-fed and rested cows (BRASIL, 2020). Even with the predominance of small and medium-sized rural properties, the milk and dairy products sector in Brazil is of great economic and social importance, in addition to promoting job creation around 4 million people, both directly and indirectly (ZOCCAL, 2018). However, it is notorious that to this day, due to various economic, structural and cultural factors, not all rural producers are able to fully comply with the legislation.

Studies show that milk production in some regions of the country faces challenges, requiring the adoption of improvements in the management used to obtain and store milk and its derivatives and, otherwise, it can compromise the quality of raw milk and the competitiveness of the sector, in addition to becoming carrier vehicles for undesirable bacteria such as psychrotrophs, among others (RAMOS et al., 2014; NETA et al., 2015; GUTH et al., 2022; MASSINI et al., 2023; STRÖHER et al., 2023, COTTA et al., 2020). In addition, it is necessary to periodically check animal health, especially in relation to mastitis, which is a disease that contributes to changes in the centesimal composition of milk and implies a reduction in the volume produced, generating economic losses for the producer.

For this, there are analyzes that seek to identify the microbial groups present in milk and dairy products, such as mesophilic bacteria, psychrotrophic bacteria, and pathogenic microorganisms, among others. The total mesophile count or total bacterial count (TBC) is

an important indicator of milk quality, as high counts indicate the presence of spoilage microorganisms (CASSOLI et al., 2016). TBC is influenced by several factors, such as milking hygiene, storage time and temperature, seasonal factors, such as time of year and climatic conditions, among others (TAFFAREL et al., 2013). However, when good production practices are neglected, refrigeration contributes to the development of psychrotrophic bacteria, which have the capacity to produce thermostable lipolytic and proteolytic enzymes, which maintain their enzymatic activity after pasteurization, or even after the treatment used to obtain UH (ultra-high temperature) milk (BELOTI, 2015; FELIPUS, 2017).

There are several species of psychrotrophic bacteria that can develop in milk at refrigeration temperature, such as *Pseudomonas fluorescens*, *Pseudomonas putida*, *Aeromonas hydrophila*, *Aeromonas sóbera*, *Aeromonas caviae*, *Burkholderia cepacia*, *Klebsiella oxytoca*, *Ewingella americana*, *Hafnia alvei*, *Chryseomonas luteola*, *Alcaligenes faecalis*, *Methylobacterium mesophilicum* and *Sphingomonas paucimobilis*. And among these bacteria, the genus *Pseudomonas* spp., stands out for its proteolytic activity associated with the deterioration of dairy products (LAMPUGNANI et al., 2019; ARCURI et al., 2008).

Regarding the proximate composition, milk is formed by lipids, carbohydrates, proteins, mineral salts and vitamins and represent approximately 12 to 13% of milk, and water approximately 87%. Solid elements, their distributions and interactions are determinant for the structure, functional properties and suitability of milk for processing and manufacturing dairy products (DALPIAZ et al., 2018).

Another important analysis in verifying milk quality, both for producers who store milk in individual and collective tanks, is the somatic cell count (SCC), as it is possible to make an assessment of the health of the mammary gland, since this test is an indication of the degree of inflammation. Bovine mastitis is characterized by an infection of the mammary parenchyma that affects a large part of dairy herds, and the highest incidence of cases is of bacterial etiology, but can also occur due to fungal infection (de GOUVEIA et al., 2022). And this disease can present itself in clinical or subclinical forms. The first presents evident signs, such as edema and increased udder temperature, hardening, pain, lumps and pus at the site, and changes in the characteristics of the milk (COSER et al., 2012). The subclinical form, on the other hand, despite not showing visible signs of inflammation in the udder, is characterized by an increase in the number of somatic cells, an increase in chlorine and sodium contents, in addition to a decrease in the levels of casein, lactose and fat, affecting

the quality and volume of the milk produced. (COSTA et al. 2017; DEMEU et al. 2016; STRÖHER et al. 2023).

The studies reinforce that mastitis significantly alters the physicochemical parameters of milk, compromising both nutritional and industrial quality. In a study carried out by Liu et al. (2023), the authors highlighted that molecular interventions represent a promising strategy to mitigate the inflammatory impacts associated with subclinical mastitis in cows, contributing to the reduction of antimicrobial use and would involve the use of technologies that would modulate gene expression and cellular processes related to the inflammatory response.

This analysis should be seen as an extremely valuable tool that, among other purposes, allows the monitoring of the prevalence of subclinical mastitis in the herd, especially those caused by contagious microorganisms. In addition to providing important information about the quality of raw milk to the industry, it indicates the hygienic conditions under which the milk was produced on the properties, the possibility of estimating milk production losses, guides the producer to make decisions in order to prevent the transmission of mastitis during lactation, identification of cows for treatment, drying and disposal (LANGONI, 2000; SILVA, 2015; VEIGA, 2016).

The Ministry of Agriculture, Livestock and Supply (MAPA) issued Normative Instruction 76 (IN 76), which establishes technical requirements for the identity and quality of refrigerated raw milk, pasteurized milk and type A pasteurized milk. This standard is part of a set of regulations, including Normative Instruction 77 (IN77), which aim to ensure the quality of milk produced in Brazil and which are currently in force (BRASIL, 2018a and 2018b).

The State of Espírito Santo is located in the southeastern region of Brazil and has an area of 1.46 million hectares of pastures intended mostly for beef and dairy cattle. There are about 1.94 million head of cattle in these pastures (IDAF, 2019), with emphasis on the municipalities of Ecoporanga, Linhares and Nova Venécia, which have the largest herds in the state (24% of the total herd) (INCAPER, 2020; IBGE, 2018a). And dairy farming is an activity of significant relevance in the state and stands out for its expressive volume of production and socioeconomic importance. In 2018, 417 million liters of milk were produced, generating a value of around 495.6 million reais (OTAVIANO et al., 2020; IBGE, 2018b).

Regarding production, seventy-five (75%) in the state is supplied by small properties with daily production of up to 100 liters. This activity is of significant importance because it is present in practically all municipalities and contributes to the economic development of Espírito Santo. In the northern region of the state located above the Doce River, which has

higher values both in relation to animal herd and milk production (SANT'ANNA, SESSA, 2021).

Considering these facts and the production characteristics of the northern region of Espírito Santo, it is understood the importance of encouraging milk production that guarantees better yields to small producers and the dairy industry, as well as consumer security in acquiring adequate products and in addition, in this region there is a lack of published data on the quality of raw milk.

In this chapter, the quality parameters of raw milk stored in individual refrigeration tanks located in three family farms located in the northern region of Espírito Santo will be addressed. Analyses of microbiological quality, centesimal composition and somatic cell count were carried out, in addition to checking the conditions of the refrigeration tank site and the temperature used during collection. Obtaining this data will allow producers to direct the adoption of more actions aimed at improving the production and obtaining of adequate milk and dairy products, in addition to promoting the sustainability of the sector in the region.

2 METHODOLOGY

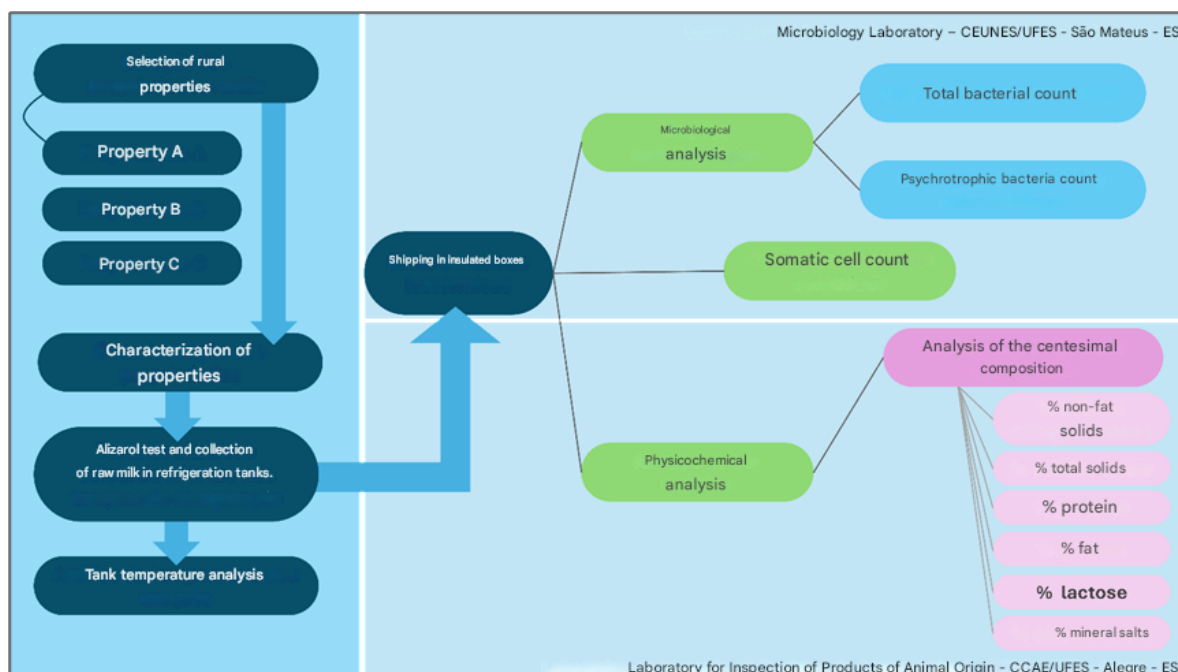
2.1 GENERAL CHARACTERISTICS OF PROPERTIES, INSTALLATION LOCATION AND TEMPERATURE OF TANKS

This research was carried out in a municipality in the northern region of Espírito Santo and three family-based rural properties were inserted and whose producers accepted to participate in the project. The selection of properties was carried out by convenience sampling and in addition, properties that had accessibility difficulties were not included, especially with regard to the infrastructure of side roads and bridges.

Refrigerated raw milk samples were collected and the temperature record values of the individual tanks were recorded. Afterwards, the samples were placed in sterile flasks, using sterile instruments, and then stored in isothermal boxes under refrigeration (between 4 and 8° C). Subsequently, they were transported for microbiological and physicochemical analysis, at the Microbiology Laboratory of UFES – CEUNES in São Mateus - ES and to the Laboratory of Inspection of Products of Animal Origin - LIPOA of the Department of Veterinary Medicine of UFES in Alegre - ES respectively. The order and organization of the project were maintained using the flowchart presented below (Figure 1).

Figure 1

Microbiological, physicochemical analysis and somatic cell count in refrigerated raw milk samples



Source: The authors.

2.2 MICROBIOLOGICAL ANALYSES

Total bacterial count (TBC)

Dilutions of milk samples were performed in sterile saline solution (0.85%) (w/v) and sowing on Standard Count Agar (PCA), using the depth seeding technique. After solidification, the plates were incubated at 35° C/24 h. Plate counting was performed and the results obtained were expressed in CFU/mL according to APHA (2001). The Ministry of Agriculture, Livestock and Supply (MAPA), through Normative Instruction No. 76 of November 2018, establishes a maximum limit of 300,000 CFU/mL of raw milk (BRASIL, 2018a).

Psychrotrophic Bacteria (PBC) Count

To perform the psychrotrophic bacteria count, the same methodology described in the previous item was used, with changes only in temperature and incubation time, which were 7° C/10 days. After the incubation period, the counts were performed and the results were expressed in CFU/mL according to the APHA (2001).

2.3 PHYSICOCHEMICAL ANALYSIS:

Determination of the centesimal composition of milk samples

The analyses of protein, fat, lactose, total solids and mineral salts were carried out at the Laboratory of Inspection of Products of Animal Origin (LIPOA) of the Center for Agrarian Sciences and Engineering of UFES in Alegre - ES. The analysis was performed in an automated manner using the Lactoscan® device. The tests were performed in triplicate and the results found were compared with the legislation (Normative Instruction No. 76, of MAPA) (BRASIL, 2018a).

2.4 DETERMINATION OF SOMATIC CELL COUNT (SCC)

The somatic cell count (SCC) was determined using the Somaticell® Kit. The analyses were carried out following the manufacturer's guidelines. In a specific tube provided by the manufacturer, in the vertical position, 2 mL of reagent was added and then, with the aid of a Pasteur pipette, 2 mL of milk was added. The mixture was homogenized with the aid of a stick, making 30 consecutive vertical movements for 20 to 24 seconds. Afterwards, the tube was sealed and inverted for 20 seconds and returned to its initial position, after 5 seconds the tube was read on a graduated scale. Refrigerated raw milk from individual tanks or for community use must have quarterly geometric means of SCC of a maximum of 500,000 CS/mL (BRASIL, 2018a).

3 RESULTS AND DISCUSSION

3.1 GENERAL CHARACTERISTICS OF THE PROPERTIES, INSTALLATION LOCATION AND TEMPERATURE OF THE TANKS

In this study, periodic collections were carried out in three different family farms with different profiles and were named A, B and C and manual milking was performed and all samples were stable to the alizarol 72° GL test. Property A is located in the rural area and, in terms of herd size and infrastructure, the property is small and easily accessible, in terms of the structural quality of the roads and geographical location. Property B was located in the rural area, is medium-sized and difficult to access, as the structural quality of the roads was relatively compromised. Property C was located in the urban area, medium in size and easily accessible.

In addition, the properties of the present study had individual refrigeration tanks that specifically met the demands of the respective properties. The milk cooling and storage tank, for individual or community use, must be installed on the rural property in an appropriate place, provided with walls, roofing, paving, lighting, ventilation and running water point

(BRASIL, 2018a). And in relation to the hygiene and cleaning of the installation site, no non-conformities were observed and not even at the time of collections, such as insects or the presence of domestic animals. It was found that the average temperature value recorded in tank A was 3.87° C, that of tank B was 4.3° C and that of tank C, 2.4° C and that they were in accordance with the legislation being a determining factor in the quality of the milk, since the inadequate temperature directly influences bacterial growth. The legislation establishes that milk at the refrigeration station must be kept at a maximum temperature of 4° C (BRASIL, 2018a).

In studies carried out on milk production conditions in some family farms located in the municipality of Alegre – ES and in São Mateus – ES, the authors observed several non-conformities related to good production practices not only during milking, but also before and during its storage in refrigeration tanks, requiring adjustments in milk production management (JUNQUEIRA et al., 2015; RAMOS et al., 2014; LACERDA, 2014; NETA et al., 2016; NETA et al., 2018; PICCOLO et al., 2018; ULISSES et al., 2022; MASSINI et al., 2023).

3.2 TOTAL BACTERIAL COUNT (CBT) AND PSYCHROTROPHIC BACTERIA COUNT (CBP)

The mean values of total bacterial count (TBC) and psychrotrophic bacteria count (PBC) are shown in Table 1.

Table 1

Mean values of total bacterial count (TBC) and psychrotrophic bacteria count (PBC) in refrigerated raw milk samples.

Cooling tank (individual)	CBT (CFU/mL)	CBP (CFU/mL)	% BP/BT
A	7.24x10 ⁴	4.98 x10 ²	0,68%
B	2.57 x 10 ⁴	NR	-
C	6.60 x 10 ⁴	NR	-

NR: Non-representative (count less than 25 CFU/mL); CBT: Total Bacterial Count; CBP: Psychotrophic Bacteria Count. % BP/BT: Percentage of psychrotrophic bacteria/total bacteria.

In this study, regarding the total bacterial count (TBC), no sample presented values above what is recommended by IN 76/2018, which establishes that refrigerated raw milk from individual tanks or for community use must have quarterly geometric averages of a maximum

of 300,000 CFU/mL. And the psychrotrophic bacteria (PBC) count was less than 10% of the TBC. One hypothesis for this low count may be related to the shorter time the milk was stored in the refrigeration tanks and because they are individual. In Brazil, there are no values for psychrotrophic bacteria in the legislation, however, these results are in accordance with what was suggested by Cousin (1982). According to the author, values higher than 10% of mesophilic values should not be recommended considering that many species belonging to this group of microorganisms have high spoilage potential, and thus can compromise the harmlessness of the product mainly due to the proteolytic capacity of these microorganisms.

In addition, it was also observed the conformity in relation to the effect of maintaining the adequate temperature and storage time of milk in cooling tanks on dairy farms with microbial growth being a parameter of importance for the maintenance of the microbiological and physicochemical quality of raw milk. In other words, maintaining the appropriate temperature contributed both to the quality of the milk and to the reduction of economic losses, generating sustainability for the sector. It is important to emphasize that cooling milk after obtaining it is the widely accepted method to minimize the speed of multiplication of mesophilic microorganisms, which are usually responsible for the deterioration of this product.

Massini et al., (2023), obtained high values for the count of psychrotrophic bacteria in raw milk collected in a municipality in the southern region of Espírito Santo. In one of the samples that was obtained in a collective tank, it presented values of 107 CFU/mL. These values could be attributed to the mixture of milk from several properties in a single tank, which favors contamination and subsequent bacterial multiplication.

Neta et al., (2016), when analyzing the quality of raw milk stored in four refrigeration tanks in the southern region of ES, concluded that high values of mesophilic and psychrotrophic bacteria found were indicative of failures in the procedures used in hygiene and due to inappropriate production and storage conditions. In addition, all refrigeration tanks were of the collective type and in this case, there were greater possibilities of contamination considering the mixture of milk from several rural properties.

Some genera of psychrotrophic bacteria produce thermostable extracellular enzymes, mainly proteases and lipases, which degrade milk components and, consequently, alter the characteristics of dairy products. Goulart et al. (2021), evaluated the physicochemical characteristics of natural yogurts produced from milk contaminated by proteolytic psychrotrophic bacteria. The presence of proteases can alter the composition of yogurt,

degrading the proteins into smaller water-soluble peptides that are lost in whey separation. The authors observed that the longer the time spent in refrigeration, without appropriate heat treatment, favored the multiplication of these bacteria and found that good production practices should not be neglected, which are related to lower CBT values and, consequently, lower values of psychrotrophic bacteria.

According to Ströher et al. (2023), when analyzing refrigerated raw milk quality parameters from 33 (thirty-three) small dairy farms in Vale do Taquari, RS, the authors concluded that three milk producers showed non-conformities in the analysis of Standard Plate Count or TBC of refrigerated raw milk (8.57%), with values of 11,000 CFU/mL as the overall average. This is an analysis that classifies the microbiological quality of milk, as it measures the aerobic mesophilic bacteria present in it.

Alves (2024), evaluated the microbiological quality of raw milk samples obtained from 155 rural properties located in the interior region of the state of Goiás, through the analysis of total bacterial count, psychrotrophic bacteria and somatic cells. The author reported that for the values obtained for total bacterial count, they were mostly low, ranging from 2,000 to 125,000 CFU/ml, suggesting good hygiene and milking practices and positive results for psychrotrophic bacteria with CFU/mL counts ranging from 10^1 to 10^3 .

Santana et al. (2021), evaluated the quality of refrigerated raw milk collected in 12 different tanks, which were collective and individual located on rural properties in the municipality of Ouro Preto do Oeste -RO and found lower CBT values ranging from 8.9×10^5 and 7.1×10^5 CFU/mL in samples collected in tanks with lower temperature values around 4.9°C and 6.1°C , respectively, while in another tank that presented a temperature of 14.7°C at the time of collection, the values were CBT 2.2×10^6 CFU/mL.

Psychrotrophic bacteria have high deteriorating potential, and thus can compromise the harmlessness of the product, mainly due to the proteolytic capacity of these microorganisms. Another risk factor inherent to the presence of these microorganisms in milk is the formation of biofilm by bacteria of the genus *Pseudomonas* spp. (MANN; WOZNIAK, 2012), which are bacterial communities surrounded by a polysaccharide matrix, conferring high resistance and persistence to contamination control methods (SOUZA et al., 2021).

When evaluating samples of cheese from raw milk in the northwest region of São Paulo, researchers identified the presence of pathogenic agents, including *Listeria* spp., detected in 68 (64.14%) of the 106 samples obtained from bovine feces, swabs from the hands of milkers and cheesemakers, buckets, raw milk, whey, water, surfaces and utensils

of cheese production (RIBEIRO et al., 2022). And in this same study, the authors evaluated 391 isolates of *Staphylococcus* spp., which were obtained from samples of raw milk, milkers' hands, whey, utensils, and cheeses. Of this amount, 60 (15.31%) were identified as *Staphylococcus aureus* by PCR (*Polymerase Chain Reaction*), and of these, 15.31% had virulence genes (*eta*, *hlg*, *seg*, *seh*, *sei*).

Ribeiro Júnior et al., (2018) in order to identify, quantify and evaluate the deteriorating activity of *Pseudomonas* spp. in bovine milk produced in the state of Paraná, obtained plate counts of *Pseudomonas* spp. that ranged from <10 to 1.3×10^3 CFU/mL, with an average of $0.89 (\pm 3) \times 10^2$ CFU/mL. The detection of this psychrotrophic bacterium in milk is a risk for producers and consumers, due to its ability to interfere with the shelf life and sensory characteristics of this product, in addition to being an important pathogenic agent.

3.3 COMPOSITIONAL EVALUATION OF MILK SAMPLES

The results of the centesimal composition analysis are shown in Table 2.

Table 2

Centesimal composition of raw milk samples stored in individual refrigeration tanks

Parameters	A	B	C	Legislation IN 76/2018 ¹ Recommended minimum
Fat (%)	2,47 %*	2,86 %*	3,67 %	3,0 %
Non-greasy solids (%)	7,97 %*	8,05 %*	8,00 %*	8,4 %
Total Solids (%)	10,44 %*	10,92 %*	11,68 %	11,4 %
Protein (%)	2,87 %*	2,95 %	2,92 %	2,9 %
Lactose (%)	4,38 %	4,43 %	4,40 %	4,3 %
Mineral salts (%)	0,64 %	0,65 %	0,65 %	—

¹Normative instruction 76 (BRASIL, 2018a). * values lower than the standard established by IN 76/2018.

In this work, in relation to the centesimal composition, it was observed that the milk samples obtained in the tanks of properties A and B presented percentages of fats, non-fat

solids and total solids below what is recommended by IN 76/2018, while the milk samples of tank C presented rates above or close to the values established by the current legislation.

On the other hand, the protein and lactose values were closer to the recommended minimum limits. These values can be influenced by several factors, such as season, health, breed and, especially, the animal's diet (FRIGERI et al., 2020; PACHECO, 2011). Neiva Júnior et al., 2021, when analyzing the centesimal composition of bovine milk samples, observed the following results: fat 3.5%; protein: 3.1%; lactose: 4.5%; total solids: 12.04% and non-fat solids 8.5%, without observing changes in composition between the dry and rainy seasons, all within the desirable standard.

The substantial reduction of the lactose concentration or total solids in the milk suggests fraudulent addition of water after milking, which affects the quality and authenticity of the product. On the other hand, variations in the concentration of proteins, fats, and lactose can directly impact the physical and functional properties of milk, affecting its processing, stability, and the quality of dairy products (BRITO et al., 2021).

3.4 SOMATIC CELL COUNT (SCC)

In this study, the mean somatic cell counts were 650,000 CS/mL in milk samples stored in tank A, 240,000 CS/mL in tank B, and 500,000 CS/mL for tank C. Thus, only the milk samples obtained in tank A presented higher counts in disagreement with the current legislation, and it was the same sample that presented the most alterations in the centesimal composition, for several parameters analyzed. High somatic cell counts are directly related to the centesimal composition of milk, with a decrease in noble groups of nutrients such as fat and protein (MONTANHINI et al., 2013). Cell count is a parameter related to herd health and directly interferes with centesimal composition.

The main etiologic agents of mastitis are *Staphylococcus aureus*, *Streptococcus agalactiae*, *Corinebacterium* sp. *Mycoplasma bovis*, although mycoplasmas, yeasts and algae of the genus *Prototheca* are also reported (BARKEMA et al., 2009; ZADOKS et al., 2011). Mastitis represents a serious concern within the milk production chain due to the great losses it causes and not only changes in quality and impact on the reduction of the volume of milk produced (MASSINI et al., 2023; SANTOS et al., 2022; PICCOLO et al., 2018). In addition, it is a disease and should not be neglected and can be clinical, when the externalization of symptoms occurs, and subclinical, when the externalization of symptoms does not occur. The value obtained from somatic cells may be indicative of subclinical

mastitis, a very frequent situation in herds and which requires monitoring (MASSOTE et al., 2019). On the other hand, the average value of somatic cells obtained in samples from tank C indicates a count within the limit established by legislation. This means that greater care must be taken with the hygiene of the teats, as proper hygiene of the mammary gland may be the most important single measure in the prevention of new intramammary infections (LIMA et al., 2022). The main form of prophylaxis for this clinical condition is to follow good manufacturing practices, such as the adoption of "milking line" practices, adequate pre- and *post-dipping*, personal hygiene care of milkers, keeping animals standing after milking, among others (MASSOTE et al., 2019).

In a study carried out by Alves (2024), the author evaluated the microbiological quality of raw milk samples obtained from 155 rural properties located in the interior region of Goiás, through somatic cell count (SCC) analyses. It was found that 81.81% of the samples analyzed had high counts and outside the standard established by the legislation, most of them exceeding 500,000 CS/ml, which indicated health problems in the herd.

Thus, it is imminent to continue the work with the participation of other rural producers to monitor the quality of refrigerated raw milk in the northern region of ES.

4 CONCLUSION

With these facts presented, it was concluded that the milk samples were in accordance with the current legislation, with regard to the total bacterial count. And the adequate storage temperature verified in the refrigeration tanks played a crucial role in preserving the quality of the milk, however, somatic cell values above those established by the legislation indicated the presence of mastitis, in addition to directly impacting the centesimal composition of the milk and influencing the process of action of the lactic cultures used in the preparation of dairy products.

It is of fundamental importance to adopt good agricultural practices, including verification of animal health and diet, as such procedures are indispensable to ensure the quality and supply of milk and dairy products to the consumer. By implementing and adopting rigor in relation to hygiene and sanitization at all stages of the production chain, producers can mitigate risks related to public health and raise the quality of milk for the consumer. The continuation of research is imminent and all this will contribute to the sustainability of the milk and dairy products sector in the northern region of Espírito Santo and to the health of the population.

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