

Quantification of mesophilic aerobic bacteria, coliform bacteria and *Staphylococcus aureus* and in the production of fresh cheese in a rural community in Brazil

Abstract

Cheese is a milk derivative widely consumed in Brazil, having a great nutritional importance. The microbiological quality of food is of vital importance, as existing microorganisms, especially in products of animal origin, can cause serious diseases, so the objective of this study was to evaluate the quality of Minas Frescal cheese produced in a rural community in Brazil. Twenty samples of Minas Frescal cheese were analyzed, these samples were submitted to analysis for counting mesophilic aerobic bacteria, total coliform bacteria, coliform bacteria at 45°C and *Staphylococcus aureus*. Brazilian legislation was used as a parameter for the evaluation of the analyzed cheese samples. All samples analyzed showed high counts of mesophilic aerobic, bacteria total coliform bacteria, coliform bacteria at 45°C and *Staphylococcus aureus*. The high count of total coliform bacteria, coliform bacteria at 45°C and *Staphylococcus aureus* observed in the analyzed cheese samples suggests that this product may pose a health risk to consumers.

Keywords: dairy products, food contamination, microbiological parameters

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Introduction

Foodborne diseases are one of the most important causes of mortality worldwide, despite technological advances in industry and medicine. The prevention of these diseases depends on care in the stages of production and handling of raw materials, processing, distribution and marketing of the finished product. Obtaining milk hygienically is the crucial point in the process of making cheese and dairy products, as the animal, equipment and the environment can be contaminated.¹ Cheese is a highly nutritious product, however, if outside the standards of hygiene and food safety, it can promote the growth of microorganisms during processing and sale.² Milk is a biological fluid of high nutritional value for mammalian species. When extracted from the mammary gland of healthy animals, under aseptic conditions, the milk shows average bacterial counts from 5.0×10^2 to 10^3 CFU.mL⁻¹, mainly comprised by the saprophytic microbiota.³ In general, the microbial load of milk depends on the number of microorganisms that come into contact with the milk before milking or through subsequent contamination.⁴ The hygienic obtaining of milk is the first critical point in the process of making cheese and other dairy products, since the animal, the equipment and the milking environment can represent an important source of contamination by microorganisms.

Staphylococcus aureus is one of the main agents of food poisoning worldwide. In general, foods that require a lot of handling during preparation are at greater risk of causing staphylococcal poisoning. Among the foods most involved with staphylococcal poisoning are shellfish, cooked meats, salami and cheeses. Although pasteurization causes a decrease in the population of microorganisms present in milk, some toxins, such as staphylococcal enterotoxin, are not inactivated and can cause food poisoning in the consumer. The presence of *Staphylococcus aureus* in food is directly related to poor hygiene and inadequate handling.⁵ Milk products, especially cheese, are considered a frequent vehicle for pathogens, especially those produced in an artisanal way, without proper hygiene care and good

handling practices.⁶ Therefore, it is necessary to adopt good handling practices, storage, transport and commercialization of artisanal fresh cheese, since contamination can occur at any stage of production, and inspection in the in order to offer a better quality product to consumers.⁷

Artisanal cheeses produced in rural communities in the municipality of Januária-MG are usually not evaluated for their microbiological quality, and inspection by sanitary surveillance agencies does not occur. Thus, this study aims to evaluate the quality of Minas Frescal cheese produced in a rural community in Brazil.

Material and methods

A total of 20 samples of fresh cheese from artisanal production were acquired from producers in a rural community in the municipality of Januária-MG, Minas Gerais State, Brazil. The collected samples (Figure 1) were transported in an isothermal box to the Microbiology Laboratory of the Federal Institute of the North of Minas Gerais (IFNMG) Januária campus. All samples were analyzed for total coliform and coliform bacteria counts at 45°C, *Staphylococcus aureus* counts and mesophilic aerobic bacteria. Samples of 50 g of the cheeses were added to sterile plastic bags containing 225 mL of 0.1% peptone water solution, and then homogenized in a stomacher (Figure 1). The homogenized samples were used to count total coliforms, coliforms at 45°C and *Escherichia coli* bacteria using the Most Probable Number (MPN) method. For *Staphylococcus aureus* analysis, 50 g samples of the cheeses were added to sterile plastic bags containing 225 mL of 0.1% peptone water solution, then homogenized in a stomacher (Figure 1). A total of three decimal dilutions were obtained, of which 0.1 mL of each was spread on the surface of three Baird-Parker potassium tellurite agar plates. The plates were further incubated at 35°C/45-48h. Subsequently, the number of Colony Forming Units (CFU) was counted. After the incubation period, typical colonies were inoculated into flasks containing Brain Heart Infusion (BHI) broth and incubated at 35°C/18-24h.



Figure 1 Processing of cheese samples for microbiological analysis.

Results and discussion

The count of total coliform bacteria, coliform bacteria at 45°C, *E. coli*, *S. aureus* and aerobic mesophiles in Minas Frescal cheese samples

Table 1 Count of mesophilic aerobic bacteria, total coliform bacteria, coliform bacteria at 45°C, *Escherichia coli* and *Staphylococcus aureus* in Minas Frescal cheese samples collected in a rural community in Brazil

Sample	Total Coliforms (MPN.g-1)	Coliforms at 45°C (MPN.g-1)	<i>E. coli</i> (MPN.g-1)	<i>S. aureus</i> (CFU.g-1)	Mesophilic Aerobes (CFU.g-1)
1	> 1.100	> 1.100	> 1.100	> 10 ³	> 10 ³
2	> 1.100	> 1.100	> 1.100	> 10 ³	> 10 ³
3	> 1.100	> 1.100	> 1.100	> 10 ³	> 10 ³
4	> 1.100	> 1.100	> 1.100	> 10 ³	> 10 ³
5	> 1.100	> 1.100	> 1.100	> 10 ³	> 10 ³
6	> 1.100	> 1.100	> 1.100	1,0 × 10 ⁷	4,2 × 10 ⁹
7	> 1.100	> 1.100	> 1.100	8 × 10 ⁶	4,5 × 10 ⁹
8	> 1.100	> 1.100	> 1.100	5 × 10 ⁶	3,8 × 10 ⁹
9	> 1.100	> 1.100	> 1.100	5,4 × 10 ⁶	4,1 × 10 ⁹
10	> 1.100	> 1.100	> 1.100	1,0 10 ⁷	4,7 × 10 ⁹
11	> 1.100	> 1.100	> 1.100	2,4 × 10 ⁶	4,1 × 10 ⁹
12	> 1.100	> 1.100	> 1.100	1,9 × 10 ⁶	5,5 × 10 ⁸
13	> 1.100	> 1.100	> 1.100	1,0 × 10 ⁶	2,5 × 10 ⁸
14	> 1.100	> 1.100	> 1.100	1,9 × 10 ⁶	5,3 × 10 ⁸
15	> 1.100	> 1.100	> 1.100	2,1 × 10 ⁶	4,5 × 10 ⁸
16	> 1.100	> 1.100	> 1.100	1,9 × 10 ⁶	4,8 × 10 ⁸
17	> 1.100	> 1.100	> 1.100	2,5 × 10 ⁶	9,5 × 10 ⁸
18	> 1.100	> 1.100	> 1.100	1,9 × 10 ⁶	3,9 × 10 ⁸
19	> 1.100	> 1.100	> 1.100	2,7 × 10 ⁶	3,3 × 10 ⁸
20	> 1.100	> 1.100	> 1.100	2,2 × 10 ⁶	2,9 × 10 ⁸

Conclusion

The high count of mesophilic aerobic bacteria, total coliform bacteria, coliform bacteria at 45°C and *Staphylococcus aureus* observed in the analyzed cheese samples suggests that this product may pose a health risk to consumers.

Acknowledgments

None.

Conflicts of interest

The author declares there is no conflict of interest.

(n = 20) is presented in Table 1. In all analyzed samples, the count of coliform bacteria at 45°C was above the tolerance limit determined in RDC 12 of 2001 of the National Health Surveillance Agency of the Brazil, characterizing the product as unfit for consumption. Analyzing five samples of artisanal cheese sold in establishments in the city of Castro state of Paraná, found that 80% of the samples analyzed were characterized as unfit for consumption, due to high contamination by coliform bacteria at 45°C. The high count of total coliform bacteria is an indication of a possible hygienic-sanitary deficiency in the manufacture, handling and storage of the analyzed product. Were analyzed 20 samples of Minas Frescal cheese obtained from street markets in the city of Uberlândia, Minas Gerais State, noting that 70% of the samples had a count above 10³ MPN.g⁻¹ for total coliform bacteria. High total coliform counts, often described in Minas Frescal cheese, can reduce shelf life due to product deterioration and are indicative of failures in hygiene procedures.⁸

All cheese samples analyzed in this study showed high counts of *S. aureus* and the higher the count of *S. aureus*, the greater the possibility of having coagulase-positive species. Bacteria of the genus *Staphylococcus* have a habitat in humans and animals, being found on the skin, glands and mucous membranes,⁹ and their presence in cheeses indicates that the raw material used may be from infected animals or that the product may have been handled under poor hygienic conditions, which may produce toxins when found in large quantities, and attention to hygienic issues in the handling of these products is important.¹⁰

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