

Microbiological quality of colonial cheese sold in Porto Alegre-RS

Qualidade microbiológica de queijos coloniais comercializados em Porto Alegre-RS

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Abstract

Colonial cheese is a traditional dairy product in southern Brazil and is commonly purchased by the citizens of Porto Alegre. However, there is still a lack of technical regulation of colonial cheese, and there is little information about the microbiological quality of this product at the retail level. Thus, the objectives of this study were to (i) evaluate compliance with the legal microbiological standards of colonial cheese sampled from street fairs and the central market of the city of Porto Alegre; (ii) statistically test the hypothesis of an association between noncompliance with the standards and local purchasing (street fairs or central market); (iii) estimate the number of *Listeria* spp. and *Listeria monocytogenes* in the positive samples; and (iv) characterize the *L. monocytogenes* strains by serotyping and macrorestriction (PFGE). For this purpose, 205 cheese samples belonging to 17 different brands were analyzed. The microbiological analyses were conducted according to ISO standardized protocols for the detection of *L. monocytogenes* and *Salmonella* spp. or by enumeration of coagulase-positive *Staphylococcus* and coliforms at 45°C. Among the samples, 47.31% did not comply with at least one of the microbiological standards established by the Brazilian legislation and were thus unsuitable for human consumption. Regarding the coliforms at 45°C and coagulase-positive *Staphylococcus*, 10.73% and 40.48% of the samples presented higher counts than the legal parameter, respectively. There was no association between the frequency of samples with coagulase-positive *Staphylococcus* counts above the legal parameter and local of purchasing; however, the commercial brand influenced the frequency of unsuitable samples. This may indicate failures of hygiene during cheese production. *Salmonella* spp. were not detected. *Listeria monocytogenes* was isolated from 2.9% of the samples. The estimated average populations of *Listeria* spp. and *L. monocytogenes* were low in the positive cheese samples at -3.3 log CFU g⁻¹ and -2.26 log CFU g⁻¹, respectively. The strains of *L. monocytogenes* belonged to serovars 1/2a, 1/2b and 1/2c and could be grouped into five pulsotypes with no evident epidemiological relation among them. The results demonstrate the need to improve the hygiene procedures during colonial cheese production and to strengthen monitoring at the dairy plants and retail levels.

Key words: Dairy products. *Listeria monocytogenes*. *Salmonella*. Positive *Staphylococcus* coagulase. Coliforms at 45°C.

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Resumo

O queijo colonial é um derivado lácteo típico do sul do Brasil, e amplamente adquirido pela população da cidade de Porto Alegre. Porém, esse produto lácteo ainda não conta com regulamento técnico específico e ainda há poucos dados sobre a qualidade microbiológica dos queijos ofertados à população. Sendo assim, os objetivos do estudo foram: (i) avaliar os parâmetros microbiológicos previstos na legislação em queijos coloniais comercializados em Feiras Modelo e Mercado Público de Porto Alegre; (ii) testar hipóteses estatísticas de associação entre violação do padrão microbiológico estabelecido na legislação com o ponto de comercialização; (iii) estimar a distribuição de contagem de *Listeria* sp. e *L. monocytogenes* nos queijos em que esta bactéria foi detectada e (iv) caracterizar as cepas de *L. monocytogenes* por sorotipificação e macro-restrição. Para tanto, foram analisadas 205 amostras de queijo, compreendendo 17 marcas distintas. As análises microbiológicas foram conduzidas conforme protocolos padronizados de análise de alimentos e evidenciaram que 47,31% dos queijos estavam não conformes com pelo menos um dos parâmetros microbiológicos estabelecidos na RDC nº12/2001, portanto impróprios ao consumo humano. Com relação à quantificação de coliformes a 45°C e *Staphylococcus* coagulase positiva, respectivamente, 10,73% e 40,48% das amostras apresentaram contagens superiores ao estabelecido na legislação. A distribuição das amostras não conformes com o parâmetro *Staphylococcus* coagulase positiva não esteve associada ao ponto de comercialização, mas foi influenciada pela marca comercial do queijo, indicando possíveis falhas na elaboração dos mesmos. Não houve detecção de *Salmonella* sp. e em 2,9% das amostras havia presença de *L. monocytogenes*. A população média estimada nos queijos positivos para *Listeria* sp. e *L. monocytogenes* foi baixa, respectivamente, $-3,3 \log \text{ UFC g}^{-1}$ e $-2,26 \log \text{ UFC g}^{-1}$. As cepas de *L. monocytogenes* pertenciam aos sorovares 1/2a, 1/2b e 1/2c e a macro-restrição demonstrou a presença de cinco pulsotipos sem relação epidemiológica evidente. Os resultados demonstram a necessidade de melhorar a qualidade higiênica da elaboração do queijo colonial e aumentar a fiscalização sanitária desse produto.

Palavras-chave: Derivados lácteos. *Listeria monocytogenes*. *Salmonella*. *Staphylococcus* coagulase positiva. Coliformes a 45°C.

Introduction

Colonial cheese is widely consumed in Rio Grande do Sul, Brazil. For many decades, it had been produced as a traditional subsistence dairy product in small farms of this region. However, a significant growth of its production in dairy plants has been observed in recent years. In the city of Porto Alegre, colonial cheese is often purchased by consumers who shop in street fairs and in the central market. These venues are traditional sale points in the city in which vegetables and dairy products are marketed. These sale points are under municipal health department regulation and must have a valid sale permit.

Regarding the quality of colonial cheese, the absence of specific technical regulation still constitutes an obstacle to its standardization, and thus, a diversity of ingredients and manufacturing protocols are observed. In dairy plants, colonial

cheese is produced with pasteurized milk added to dairy ferments, rennet and sodium chloride. After salting in brine, a maturation period of up to 20 days is carried out in cold chambers (GALVANI; AZEVEDO, 2013). For the monitoring of microbiological standards, colonial cheese is considered a medium-hard semi-fat cheese (between 36% and 46% moisture) (BRASIL, 1996). For medium-hard cheese, the Brazilian legislation established the following standards: absence of *Salmonella* spp. and *Listeria monocytogenes* in a 25 g sample, a maximum limit of 10^3 CFU g^{-1} of coagulase-positive *Staphylococcus* and 10^3 most probable number (MPN) g^{-1} of coliforms at 45°C (BRASIL, 2001). Products that do not comply with these standards are considered a hazard for consumers and cannot be marketed.

Between 2007 and 2017, more than 7,000 outbreaks of foodborne illnesses were reported in

Brazil, with more than 120 thousand people affected. In 2.8% of the outbreaks, milk and dairy products were the food involved (BRASIL, 2018). Cheese in noncompliance with the microbiological standards established by the legislation has often been found in informal food markets, enhancing the risk to the consumer (AMORIM et al., 2014; CASARIL et al., 2017; ZAFFARI et al., 2007; ZEGARRA et al., 2009). However, data on compliance with microbiological standards and the presence of pathogenic bacteria in colonial cheese under official inspection regulations are still scarce.

Thus, the aims of this study were (i) to evaluate compliance with the legislation of colonial cheese sold in street fairs and in the central market of Porto Alegre; (ii) to statistically test the hypothesis of an association between violation of the microbiological standards established by the legislation and the commercialization process; and (iii) to estimate the distribution of *Listeria* spp. and *L. monocytogenes* in positive cheese samples and (iv) to characterize the strains of *L. monocytogenes* by serotyping and macrorestriction.

Materials and Methods

Study outline

The sample size was calculated to have a probability of 99% *L. monocytogenes* isolation in at least one cheese sample (i.e., at least one positive sample), assuming that the number of bacteria has a Poisson distribution (HOELZER; POUILLOT, 2013). The choice of this pathogen as the parameter for the calculation of the sample size was based on the fact that cases of human infection may be severe, outbreaks are frequently associated with the consumption of dairy products (ROCOURT et al., 2003), and monitoring is part of the current Brazilian legislation. The parameters used for the sample size calculation were *L. monocytogenes* expected number of 0.001 CFU g⁻¹ (λ); 25 grams of sample; and 90% sensitivity of the isolation protocol. The

calculation was performed in Microsoft Excel software, resulting in a sample size of 205 colonial cheese units. To achieve a representative sample, the number of colonial cheese samples to be collected was divided among the 15 points in which the product was marketed in Porto Alegre: eight street fairs and seven shops in the central market. Each marketing point was visited from three to five times in a two-week interval from October 2014 to March 2015. At each sampling event, a 300-gram portion of one colonial cheese unit belonging to each of the brands available at the sale point was purchased. The brand and packaging date were recorded, as well as the storage status. The products kept in equipment with cooling capacity were considered refrigerated; however, the temperature could not be measured. The collected samples were kept refrigerated until the bacteriological analysis.

Microbiological analyses

The analytical units (25 g of cheese) were collected from the sample unit in a vertical laminar flow hood using knives, tweezers and/or sterile spoons. After the outer layer was removed from one side of the cheese, portions of different points of the sample were collected and transferred to a sterile plastic package until the 25 g required for the analytical procedures were completed. The enumeration of coagulase-positive *Staphylococcus* and coliforms at 45°C were performed according to the ISO (1999) protocol and the most probable number (MPN) method by the multiple-tube technique standardized by the American Public Health Association and described in the Compendium of Methods for the Microbiological Examination of Food (KORNACKI; JOHNSON, 2001). The detection of *Salmonella* spp. and *L. monocytogenes* was performed according to ISO (2001) and ISO (2004), respectively. Typical colonies of *Listeria* spp. were transferred to TSA-YE agar and incubated at 37°C for 18-24 h. The phenotypic confirmation of the suspected colonies

was determined by the catalase test, gram staining, motility, the presence of β -hemolysis, xylose and rhamnose fermentation and the CAMP test (SILVA et al., 2010). In *Listeria* spp.-positive samples, quantification by the MPN method was conducted (KORNACKI; JOHNSON, 2001).

Characterization of Listeria monocytogenes by serotyping and macrorestriction (PFGE)

Strains confirmed as *L. monocytogenes* were sent to the Laboratory of Bacterial Zoonosis of the Oswaldo Cruz Foundation for serotyping and subjected to macrorestriction of total DNA followed by pulsed-field gel electrophoresis (PFGE). The total DNA was extracted and digested with the enzyme *ApaI* (20 units) (New England BioLabs, United Kingdom) as previously described (PULSENET, 2009). The DNA fragments were electrophoresed in a 1% agarose gel (Pulsed Field Agarose, BioRad, California, USA) in 0.5X TBE buffer (0.9 M Tris base, 0.9 M boric acid, 0.02 M EDTA, pH 8.0 in a CHEF-DRII (BioRad, California, USA). The following running parameters were used: 6 V.cm⁻¹ and 14°C, with an initial switch of 5 s and a final switch of 40 s, for 23 h. After the electrophoresis run was completed, the gel was stained in a 1 μ g.mL⁻¹ ethidium bromide solution for 20 min and destained in distilled water for 45 min. The images were captured and scanned by a Kodak 2200 system (Eastman Kodak Company, New York, USA) and saved as a TIFF file. The pulsotypes were analyzed by the Dice similarity coefficient with a 1.0% tolerance. The profiles were grouped by UPGMA to construct the dendrogram in the software GelCompar II (AppliedMaths, Belgium).

Statistical analysis

The frequencies of nonconformity and enumeration of coagulase-positive *Staphylococcus* and coliforms at 45°C were calculated by descriptive statistical methods. Statistical assumptions

were tested to verify the association between the frequency of noncompliance (according to ANVISA's RDC 12/2001) and the sale point (central market or street fairs). In addition, the effects of the sale point on the frequency of noncompliance of coagulase-positive *Staphylococcus* and coliforms at 45°C, with the inclusion of the cheese brand variable as a control, were tested. The model was a Poisson regression with robust variance using the "sandwich" packages (ZEILEIS, 2004) and "epiDisplay" (CHONGSUUVIVATWONG, 2018) of R software (R CORE TEAM, 2017). The model estimates were interpreted as the prevalence ratio (PR) of noncompliance using the street fairs as a reference. The parameters of *Listeria* spp. and *L. monocytogenes* [$\log((\mu; \sigma)$ UFC g⁻¹] in the sampled cheeses were estimated using the maximum likelihood method (BUSSCHAERT et al., 2010) with the fitdistrplus package (DELIGNETTE-MULLER; DUTANG, 2015) of R software (R CORE TEAM, 2017). The method is suitable for estimating distributions with censored data, as is the case for MPN results and microbiological testing. In this model, the results of the screening test performed on 25 g of sample are considered together with the results of the MPN. In this approach, a negative result in the screening is considered left censored (i.e., less than one bacterium in 25 g or less than 0.04 CFU g⁻¹). Positive samples in the screening that were negative in the MPN are considered interval censorship, while positive samples in the screening and in a finite number of MPN tubes are considered uncensored data. Finally, samples positive in the screening and presenting all positive MPN tubes are considered right censored data. The goodness of fit of the model, according to the lognormal distribution, was tested by the chi-square test as well as by the comparison of the number of positive cheese samples, estimated by the model, with those observed in the collected samples, considering a detection limit of 1 CFU in 25 g of sample, i.e., -1.4 log of bacteria.

Results and Discussion

Among the 205 samples of colonial cheese analyzed, 84 (40.9%) were collected from the central market and 121 (59.1%) from street fairs (Table 1). A minimum of nine and a maximum of 18 cheese samples per shop were purchased at the central market. From six to 35 cheese samples were purchased at each street fair. The majority of the purchased cheese samples (89.7%) were stored in cooling equipment. At the street fairs, a higher frequency (14.9%) of cheese samples stored at room temperature was observed compared to that of

the central market (3.6%). The 205 cheese samples belonged to 17 brands, which were acquired between one and 39 times, depending on the availability at the sale points. Among the cheeses sampled, eight commercial brands (A, E, G, J, K, M, N, O) were sold only at the street fairs, three (I, L, P) were sold only at the central market and six (B, C, D, F, H, Q) were available in both. Half (51.2%) of the collected samples belonged to three commercial brands (C, F, P), which are thus the most available to the consumer. On the other hand, less than five cheese samples were acquired from four commercial brands (A, G, I, N).

Table 1. Distribution of colonial cheese samples that did not comply with the microbiological limits established by the Brazilian legislation from sale points at the central market and street fairs of Porto Alegre.

Sale points	Colonial cheese brands available (n)	Samples (n)	Noncompliant samples (n)			Total (%)
			Coagulase-positive <i>Staphylococcus</i>	Coliforms at 45°C	<i>Listeria monocytogenes</i>	
F1	4	12	4	0	0	4 (33.3%)
F2	7	18	6	2	0	7 (38.9%)
F3	4	6	5	0	0	5 (83.3%)
F4	4	12	7	0	0	7 (58.3%)
F5	4	17	12	1	0	12 (70.6%)
F6	8	35	13	5	1	16 (45.7%)
F7	4	6	5	2	0	6 (100%)
F8	4	15	7	2	0	8 (53.3%)
Subtotal	-	121	59 (48.8%)	12 (9.9%)	1 (0.8%)	65 (53.7%)
M1	3	9	4	3	2	6 (66.7%)
M2	4	13	5	1	0	5 (38.5%)
M3	3	12	4	2	0	5 (41.7%)
M4	4	9	2	0	0	2 (22.2%)
M5	7	18	7	4	2	11 (61.1%)
M6	2	9	2	0	0	2 (22.2%)
M7	3	14	0	0	1	1 (7.1%)
Subtotal	-	84	24 (28.6%)	10 (11.9%)	5 (5.9%)	32 (38.1%)
Total	-	205	83 (40.5%)	22(10.7%)	6 (2.9%)	97 (47.3%)

F= street fairs; M= central market.

Of the total samples of colonial cheese, 47.3% were not in compliance with at least one of the microbiological parameters established in the current legislation and were thus unfit for human consumption (Table 1). Although the cheese samples analyzed were produced in dairy plants under official inspection regulations and marketed in outlets licensed by the health authorities, the results showed that there is a need to improve the hygienic-sanitary quality of this product.

Regarding the enumeration of coliforms at 45°C, 10.7% of the samples presented counts above the maximum limit established by the legislation. There was no significant association between the marketing point (street fairs or central market) and noncompliance with this parameter. Additionally, no significant difference between the brands was detected. Much higher frequencies (above 50%) of coliforms at 45°C were reported in colonial cheese by Zaffari et al. (2007) in Rio Grande do Sul and Lucas et al. (2012) in Paraná. However, in both studies, the cheeses sampled may have been manufactured with raw milk, since they were not produced in dairy plants under official regulations. In the present study, on the contrary, pasteurization was a mandatory step in the production of colonial cheese in dairy plants. Therefore, the observed noncompliance with the coliform at 45°C is possibly associated with failure in pasteurization or postprocess contamination due to errors in good manufacturing practices during processing (SILVA et al., 2010).

The most frequently detected noncompliant standard found in the colonial cheese samples evaluated in the present study was related to the number of coagulase-positive *Staphylococcus*, since 40.5% of the samples presented counts higher than the maximum limit established by the legislation. Of the 83 cheese samples presenting noncompliance with this parameter, 12 (14.45%) also had coliforms at 45°C, which is above the limit of the legislation. The frequency of noncompliance found in the sampled cheeses was higher than that

reported in previous studies conducted in southern Brazil. In studies adopting a more restricted sampling of cheeses available to the consumer, the frequency of noncompliance with the coagulase-positive *Staphylococcus* limit ranged from 12.5% in Paraná (LUCAS et al., 2012) to 25% in Rio Grande do Sul (ADAMI et al., 2015). In studies that sampled Minas fresh cheese, similar or higher noncompliance frequencies were reported (KOMATSU et al., 2010; PINTO et al., 2011). However, the high moisture level and the absence of maturation characteristics of this cheese type are factors that favor the survival of *Staphylococcus* spp., increasing the risk of the occurrence of high coagulase-positive *Staphylococcus* counts.

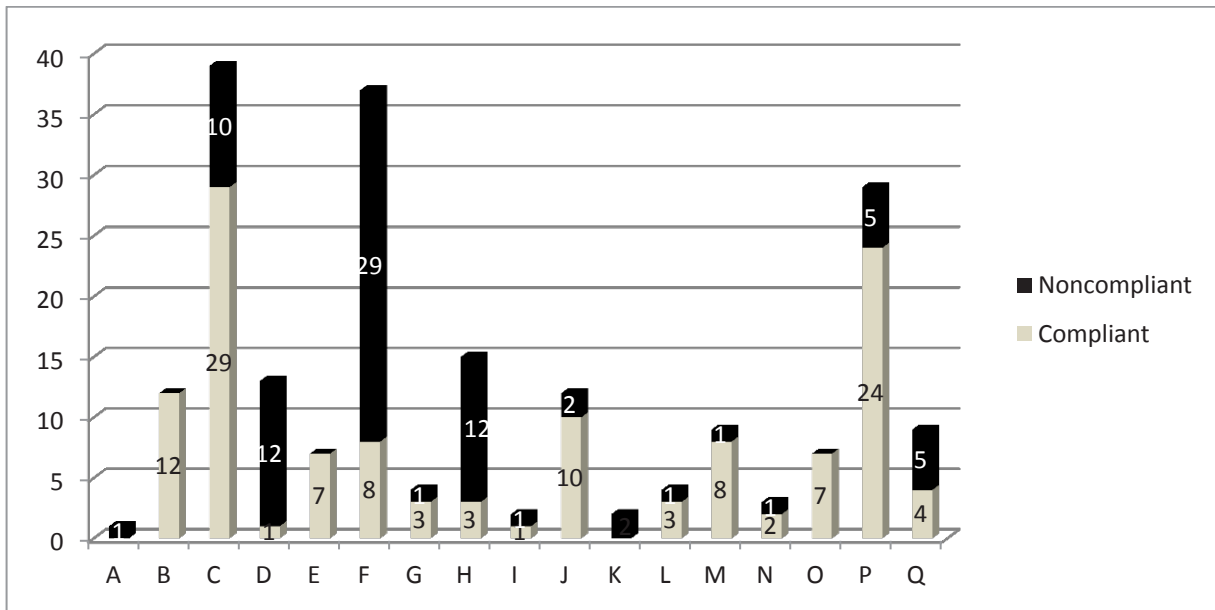
The relative prevalence (RP) of noncompliance for coagulase-positive *Staphylococcus* was 57% higher at street fairs, where the storage of cheeses at room temperature was more frequent, than in the central market ($p < 0.05$, RP = 1.57, 95% CI: 1.09-2.27). However, when the effect of the point of sale estimate was adjusted by the inclusion of the cheese brands, the point of sale was no longer significant ($p > 0.05$, RP = 1.02, 95% CI: 0.7-1.48), suggesting that the noncompliance may be related to manufacturing processes in some dairy plants. The enumeration of coagulase-positive *Staphylococcus* has been used to verify the risk of enterotoxin presence in the cheese and as an indicator of postprocess contamination related to the handling of the product or contact with surfaces with poor sanitation (SILVA et al., 2010). Considering that cheese is a product with several stages involving handling during manufacturing, it is possible that the absence of good hygiene practices by food handlers contributed to the high frequency of colonial cheese samples unfit for consumption.

Among the cheese brands most available for marketing (C, F, P), brand "F" contributed to the increase in the frequency of noncompliance of the coagulase-positive *Staphylococcus* parameter, since 78.3% of the samples collected were above the allowed limit (Figure 1). This brand was available in 13 of the 15 marketing points, which

increases the risk to consumers. Some less available colonial cheese brands sampled at street fairs (D, H, Q) also showed a high frequency (over 50%) of noncompliance, contributing to the higher occurrence of cheeses unfit for consumption at

this point of sale. Products unfit for consumption with widespread distribution in the market increase the risk to the population; therefore, continuous monitoring conducted by the health authorities at the dairy plants and sale points is highly advisable.

Figure 1. Distribution of colonial cheese brands (A-Q) according to their compliance with the limits for coagulase-positive *Staphylococcus* established by the Brazilian legislation.



All cheese samples analyzed were in compliance with the *Salmonella* spp. parameter. The absence of *Salmonella* spp. in colonial cheese, which were manufactured in dairy plants under official inspection, may be related to the fact that it is made with pasteurized milk. Other studies that investigated medium-hard cheese in Brazil also reported the absence of *Salmonella* spp. (AMORIM et al., 2014; LUCAS et al., 2012; MELO et al., 2013). However, there are reports of *Salmonella* isolation from Minas fresh and soft cheese, especially when produced with raw milk (ANTONELLO et al., 2012).

In contrast, *Listeria* spp. was detected in 12.2% of the colonial cheeses sampled. In a single sample, both *L. monocytogenes* and *L. innocua* were isolated. Among the species identified, *L.*

innocua predominated (n = 14), followed by *L. monocytogenes* (n = 6), *L. welshimeri/L. seeligeri* (n = 4) and *L. grayi* (n = 2). Therefore, six cheese samples (2.9%) were noncompliant with the absence of *L. monocytogenes*. Of these, only two samples showed concomitant coagulase-positive *Staphylococcus* or coliforms at 45°C counts higher than the legislation limit, demonstrating that contamination by this pathogen can occur even under adequate hygienic-sanitary conditions. In Rio Grande do Sul, Schwab et al. (1996) made the first report of *L. monocytogenes* isolation from colonial cheese; subsequently, Zaffari et al. (2007) also found this pathogen in artisanal produced ricotta cheese. However, studies that target the detection of *L. monocytogenes* in cheese are still scarce in Brazil.

Both the presence of *L. monocytogenes* in raw milk and cross-contamination during processing may influence the prevalence and concentration of this pathogen in cheese (FERREIRA et al., 2011; TENENHAUS-AZIZA et al., 2014). In medium-hard cheese produced with pasteurized milk, as is the case of colonial cheese, postpasteurization contact of milk or cheese with populations of *Listeria* spp. present in biofilms formed on the contact surfaces can lead to cross-contamination (CARPENTIER; CERF, 2011; TENENHAUS-AZIZA et al., 2014). The introduction of *L. monocytogenes* into the food can also occur at the retail level, especially in products that are fractionated and thus may be at risk of cross-contamination (PRADHAN et al., 2011). In 2017, the occurrence of *L. monocytogenes* in ham sliced by a supermarket chain in Porto Alegre was reported (PORTO ALEGRE, 2017), indicating that this may be a risk scenario for exposure to this pathogen. In the present study, there was a higher percentage of positive samples for *L. monocytogenes* in the central market (5.9% of the total samples) compared to that of the street fairs (0.8% of the total samples). However, as observed for coagulase-positive *Staphylococcus*, the brand of the colonial cheese influenced the result. Among the six cheese samples positive for *L. monocytogenes*, three belonged to the brand “P”, marketed only at the central market, indicating that the origin of the contamination was possibly the dairy plant.

The six strains of *L. monocytogenes* were classified into three serotypes: 1/2a, 1/2b and 1/2c. These serotypes are the most frequently reported in food and food processing plants (SWAMINATHAN; GERNER-SMIDT, 2007). In Brazil, serotype 1/2a has also been the most frequently isolated from dairy products (BRITO et al., 2008; DE NES et al., 2010; HOFER et al., 2006). Serotypes 1/2a and 1/2b, in turn, have been frequently related to clinical disease and, together with serotype 4b, are involved in the majority of cases reported in humans (HOFER et al., 2006; ORSI et al., 2011). The six strains of *L. monocytogenes* were grouped into five PFGE pulsotypes with similarity between 53.6% and 100% (Figure 2). Two strains presenting patterns with 100% similarity were isolated from samples belonging to different cheese brands and marketed in different shops at the central market. Among the three strains originating from cheese samples belonging to a common brand (P), two presented patterns with high similarity (95.6%) but belonged to different serotypes. The third strain presented a low similarity (62.6%) with the others. Thus, there was no evident epidemiological relationship between the strains, indicating that there were multiple origins of contamination. The typing results, together with the association of positive samples to a particular brand and the occurrence of *L. monocytogenes*-positive samples at distinct marketing points, support the hypothesis that contamination may occur at dairy plants.

Figure 2. Dendrogram based on ApaI-macrorestriction (PFGE) patterns of *Listeria monocytogenes* strains isolated from colonial cheese samples purchased at street fairs and the central market in Porto Alegre. Similarity analysis was performed using the Dice coefficient and UPGMA method (tolerance, 1.0%).



To conduct the risk assessment of a given food for the consumer, it is necessary to obtain information about the number of bacteria present in the food, consumption level and the minimal infective dose (ROCOURT et al., 2003). For this reason, the positive samples for *Listeria* spp. were analyzed by a protocol that allowed an estimation of the number of bacteria. Two positive cheese samples presented 3.6 NMP g⁻¹ and 11 NMP g⁻¹ of *Listeria* spp., respectively. In the other 23 positive samples, the populations found were <3.0 MPN g⁻¹. The *Listeria* spp. mean count was thus estimated at -3.3 log CFU g⁻¹ (SD = 1.63) and *L. monocytogenes* at -2.26 log CFU g⁻¹ (SD = 0.46). The model was well adjusted to the lognormal distribution and, according to the estimated parameters, the frequency predicted by the detection model of *Listeria* spp. was 12.1%, similar to that observed in the samples (25/205, or 12.2%). Similarly, the predicted frequency of *L. monocytogenes* was 3.05% and that observed was 2.9% (6/205). These results show that the number of *Listeria* spp. in cheese sold at the central market and at street fairs is low. In this sense, it is estimated that 15.9% of cheeses have a concentration >1 CFU of *Listeria* spp. per 10 g of cheese, and only 0.6% of them have a concentration >100 CFUs of *Listeria* spp. Likewise, when considering the concentration derived from the distribution, the results suggest a very low concentration of *L. monocytogenes*. In theory, only 0.31% of the cheeses present >1 CFU of *L. monocytogenes* per 10 g of product. However, the presence of risk groups in the population - pregnant, elderly and immunosuppressed - for which the infective dose may be very low must be taken into account. Therefore, the detection of *L. monocytogenes* in a ready-to-eat product, such as colonial cheese, is a hazard *per se* and thus should receive special attention from the health authorities.

Conclusions

The microbiological analysis of colonial cheese sold at the central market and street fairs of Porto

Alegre showed a high frequency of noncompliance with the current legislation, mainly regarding the number of coagulase-positive *Staphylococcus*, indicating the need to improve good manufacturing practices and strengthen the official inspection regulations at the dairy plants.

Listeria monocytogenes was detected in colonial cheese, and the contamination is possibly related to processing at the dairy plants. The estimated number of *L. monocytogenes* in positive cheese was low; however, the existence of very susceptible risk groups points to the need to strengthen the monitoring of this product.

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Conflict of Interest

The authors declare that they have no conflict of interest.

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