TITLE: QUALITATIVE EVALUATION OF *ENTEROBACTERIACEAE* ISOLATES IN MINAS FRESCAL CHEESE ACQUIRED IN MARKETS IN RIO DE JANEIRO

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ABSTRACT: Minas Frescal cheese is extensively consumed in Brazil, due to its peculiar characteristics such as lightness and freshness. It is poorly or moderately-fat cheese with high water content obtained by milk enzymatic coagulation. The Collegiate Board Resolution n. 12/2001 of ANVISA determines low counts of coagulase-positive *Staphylococcus* (n=5, c=2, $m=10^3$, $M-=5x10^3$) and thermotolerant coliforms (n=5, c=2, $m=10^2$, $M=10^3$), and absence of *Listeria monocytogenes* and *Salmonella* spp. as acceptance criteria for microbiological quality of Minas Frescal cheese. However, little is known about the specific *Enterobacteriaceae* genera other than *Salmonella* contaminating this kind of cheese. The objective of the study was to determine the variety of *Enterobacteriaceae* genera present in a sample of unfractionated Minas Frescal cheeses obtained in Rio de Janeiro city. The study was performed with 23 pieces of cheese from 12 different brands (here named A to L), all within the validity period and presenting state or federal inspection seal, marketed in stores located in the city. Portions of each cheese (25g) were homogenized in 225mL of 1% peptone water, and serial dilutions were inoculated onto MacConkey agar plates. After 18h incubation at 36ºC, three colonies of each morphology per cheese, isolated in any dilution, were selected for identification by MALDI-TOF mass spectrometry. In total, 375 *Enterobacteriaceae* isolates were obtained: *Enterobacter* (30.7%); *Raoultella* (11.7%); *Hafnia* (10.7%); *Kluyvera* (10.1%); *Escherichia* (8.3%); *Citrobacter* (6.4%); *Serratia* (5.3%); *Buttiauxella* (5.1%); *Pantoea* (2.7%); *Ewingella* (2.4%); *Klebsiella* (2.1%); *Lelliottia* (1.9%); *Rahnella* (0.8%); *Leclercia* (0.5%); *Proteus* (0.5%); *Cronobacter* (0.3%); *Pluralibacte*r (0.3%) and *Yersinia* (0.3%). Brands I and J were the source of 112 (29.8%) isolates (56 each) of the total collection. In these brands, eight different genera were identified. Brand A presented the highest diversity of *Enterobacteriaceae* (11). *Enterobacter*, *Raoultella* and *Hafnia* were recovered in the largest number of brands, being present in 11, 9 and 8 cheese brands, respectively. *Enterobacteriaceae* was absent in only one brand (L). This study identified the diversity of *Enterobacteriaceae* present in inspected and ready to sell Minas Frescal cheese and establishes a starting point for studies on the impact of the occurrence of these bacterial genera on this kind of food that is normally consumed raw.

Keywords: *Enterobacteriaceae;* food safety; Minas Frescal cheese

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