TITLE: Detection of *Mycobacterium* spp. in Coalho cheese commercialized in the city of Caicó-RN, Brazil.

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ABSTRACT

Artisanal Coalho cheese is a popular dairy product, widely consumed in the Northeast region of Brazil, and is produced with raw or pasteurized milk. Microbial contamination of cheeses has very important consequences, both for the dairy industry, given the potential economic losses, and for public health, due to the risk of outbreaks of foodborne diseases. The present study aimed to detect Mycobaterium spp. in Coalho cheese commercialized in the city of Caicó-RN Brazil, using bacterial culture and molecular diagnosis. 50 samples of Coalho cheese obtained from local street market were analyzed, 35 of which were made from raw milk (artesanal cheese) and 15 with pasteurized milk (industrialized cheese). Material was extracted from these samples, which were submitted to processes: to obtain DNA; cultivation (Stonebrink); conventional PCR for M. bovis; and finally, nested-PCR for Mycobacterium spp. In laboratory research, it was not possible to isolate tuberculous mycobacteria in the cheeses studied. However, two samples (4%) showed growth of non-tuberculous mycobacteria (NTM) in culture medium, which through molecular diagnosis showed hsp65 identity of: Mycobacterium lehmanii (sequence ID: KY933786.1, e value: 2e-133, identities: 312/363 [86%]); and Mycobacterium rutilum (sequence ID: LT629971.1, value e: 3e-108, identities: 331/371 [89%]), being indicated as indicative of environmental contamination. There are published results on mycobacterioses in humans and animals caused by agents with high genetic similarity to those found in this study. NTMs are emerging microorganisms of a ubiquitous nature and the consumption of contaminated Coalho cheese represents a risk to public health, since it could be a diffusing agent for micro-organisms. Cheeses should be produced under greater control of sanitary inspection. Due to these characteristics, they deserve greater attention in the cheese production chain, both in good agricultural practices (GAPs) and in good food manufacturing practices (GMPs).

Keywords: Coalho cheese; *Mycobacterium* ssp.; *Mycobacterium lehmanii*; *Mycobacterium rutilum*; tuberculosis.