Detection of Mycobacterium bovis by Multiplex PCR in artisanal cheeses

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The consumption of artisanal cheeses is very common in Brazil, especially the Minas cheese, also known as "fresco" or white cheese. This product, however, can be the vehicle for various pathogens, since this cheese in general is made of raw milk. The Mycobacterium bovis is an example of a micro-organism that can found in artisanal cheeses, being part of the M. tuberculosis complex alongside with M. tuberculosis, M. africanum and M. ulcerans. The informal commerce of this kind of cheese on the countryside facilitates the spread of the disease. It can be spread by contact with infected animals or consuming products made with milk contaminated by this pathogen. In order to extinguish tuberculosis of the Brazilian herd was created the National Control and Eradication of Brucellosis and Tuberculosis Plan (PNCEBT) in 2001 by MAPA (Ministry of Agriculture Livestock and Food Supply) where the indirect immune tuberculin test is the main form diagnostic on animals. Due to the fact that milk and dairy products are widely consumed by the population it was standardized the PCR technique to search for M. bovis gens in order to check if the consumables were produced from infected cows. Therefore, this study aimed to evaluate the occurrence of M. bovis in artisanal cheeses sold in local markets of Minas Gerais, São Paulo and Paraná. Twenty-eight samples cheeses were subjected to chemical extraction of DNA using phenol-chloroform. Total DNA was subjected to multiplex PCR using primers specific for the species. The amplified material was analyzed by agarose gel electrophoresis and, subsequently, it was observed that all samples were negative, namely, M. bovis gene was not present in the samples. Thus, it can be concluded that the milk used for the production of these cheeses was not infected with M. bovis.

Keywords: Artisanal cheeses, tuberculosis, *Mycobacterium*, PCR.

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