Isolation and molecular identification of *Mycobacterium* from commercially available pasteurized milk and raw milk samples collected from two infected cattle farms in Alborz Province, Iran

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**Abstract**

Objective/Background: Mycobacterium avium subsp. paratuberculosis (MAP) is an etiological agent of Johne’s disease in ruminant including cattle, sheep and goats. This disease is considered an economically important disease in cattle. Animals with paratuberculosis shed viable MAP, particularly in their milk and feces. MAP may be involved in the development of Crohn’s disease in humans through the consumption of contaminated milk and dairy products. Common methods of pasteurization are not enough to kill all MAP present in the milk and the bacterium has been isolated from raw milk, pasteurized milk and cheese samples. The purpose of this study was to evaluate two different methods for detecting MAP in milk and milk products. We analyzed the commonly used methods such as culture and molecular biology for identification of MAP.

Methods: For this study, 50 milk samples from cows with suspected Johne’s disease located in two dairy farms and 10 commercially available pasteurized milk and cheese samples from the market in Karaj city, Iran were selected. Following Ziehl–Neelsen staining of milk samples, direct microscopic detection of MAP was performed. All milk samples were centrifuged, and the concentrated samples were decontaminated using hexadecyl pyridinium chloride. The decontaminated milk suspensions were washed three times by centrifuging, and the collected filtrates were cultivated on Herrold’s egg yolk medium enriched by Mycobactin J. Finally, identification and confirmation of isolates to MAP was performed using IS900-nested polymerase chain reaction (PCR).

Results: According to the obtained results by culture and PCR methods, none of the pasteurized milk and cheese samples showed the presence of MAP. However, 10% of the tested raw milk samples collected from suspected cattle showed the presence of MAP by both culture and PCR methods.

Conclusion: Culture and PCR methods are reliable for identification of MAP from milk samples.
Conflicts of interest

The authors have no conflicts of interest to declare.