Identification of Mycobacterium Tuberculosis Complex, Using Molecular Methods

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Abstract

Background and Objective: A high level of homogeneity observed within all bacteria in the *Mycobacterium tuberculosis* complex makes a property that seriously challenges traditional biochemical-based identification methods of these pathogens in the laboratory. The work presented here was conducted to characterize *Mycobacterium tuberculosis* complex isolates in Golestan, Northern Iran.

Material and Methods: Between 2008 and 2010, 42 mycobacterial isolates were collected from clinical tuberculosis-suspected patients in Golestan province. The isolates were sub-cultured on fresh Mycobacterium-specific culture media including glycerinated and pyruvated Lowenstein-Jensen slopes. The isolates were subsequently subjected to a PCR-based identification scheme coined Huard-Warren method. This strategy consisted of three individual algorithms namely, 16SrRNA; RV typing (Rv0577, Rv3877.8, Rv1970, Rv3120, Rv1510 and IS*1561*) and RD typing (RD1, RD 4, RD9 and RD12).

Results: All isolates were proved to be *M. tuberculosis*. Furthermore, none of the patients were being infected with any other member of the *M. tuberculosis* complex or simultaneously co-infected with two mycobacteria. This fundamental observation was independently obtained by specific culture media, RV typing and also RD typing.

Conclusion: Considering the fact that cattle and sheep farming play an important role in the economy of the region, absence of *Mycobacterium bovis* in the studied isolates can be unexpected to some extent. Huard-Warren which is a simple and cost-effective identification method can be used in both reference and regional laboratory for differential diagnosis of tuberculosis.

Keywords: *Mycobacterium Tuberculosis* Complex, Huard-WarrenMethod, 16SrRNA, Golestan Province, RD Typing, RV Typing