Comparison of Multiplex PCR and Acid Fast and Auramine-Rhodamine Staining for Detection of Mycobacterium tuberculosis and Non tuberculosis Mycobacteria in Paraffin-Embedded Pleural and Bronchial Tissues with Granulomatous Inflammation and Caseous Necrosis

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Abstract

Objective
The aim of this study was to compare the sensitivity and specificity of Acid fast and Auramine-Rhodamine staining and Multiplex PCR for the detection of Mycobacterium tuberculosis complex and non tuberculosis Mycobacteria on formalin fixed paraffin embedded tissues (FFPE)

Materials and Methods
Forty cases of FFPE pleural and bronchial tissue with chronic granulomatous inflammation and caseous necrosis and 10 cases with bronchogenic carcinoma as controls were investigated. We designed a Multiplex PCR DNA amplification method with two targets: 123bp DNA fragment from IS6110, which is present only in mycobacterium tuberculosis complex and 162bp DNA encoding Ag 85complex which is present in all of mycobacteria. The FFPE also stained by Acid fast and Rhodamine-Auramine staining method.

Results
In 26 samples (65%) 123 bp and 162 bp DNA fragments were detected together (12 in bronchial samples and 14 in pleural samples). The 162 bp fragment wasn't detected alone. The sensitivity of PCR was 65% and the specificity was 100%. Eleven cases were positive for Acid fast staining. There was 27.5% sensitivity and 100% specificity. Thirteen cases were positive for Auramine-Rhodamine staining (A-R-S); there was 32.5% sensitivity and 100% specificity. All of the 10 controls were negative for 123 bp, 162 bp DNA fragments, for Acid fast and Auramine-Rhodamine staining.

Conclusion
Multiplex PCR is a sensitive, specific and rapid method for detection of M. tuberculosis in FFPE tissues.

Keywords: Acid fast stain, Auramine-Rhodamine stain, Bronchial tissue, M. tuberculosis, Multiplex PCR, Pleural tissue

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