



ORIGINAL RESEARCH PAPER

Pulmonary Medicine

COMPARISON OF CONVENTIONAL METHODS WITH GENE Xpert MTB/RIF ASSAY FOR DETECTION OF MYCOBACTERIUM TUBERCULOSIS AND RIFAMPICIN RESISTANCE IN PLEURAL BIOPSY SAMPLES AMONG SUSPECTED CASES OF TUBERCULAR PLEURAL EFFUSION

KEY WORDS: Gene Xpert, Pleural Biopsy,

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ABSTRACT

BACKGROUND- The Gene Xpert MTB/RIF assay is an automated, closed system that performs real-time PCR and can be used by operators with minimal technical expertise, enabling diagnosis of tuberculosis and simultaneous assessment of rifampicin resistance within 2 hours. Aim is to compare the conventional methods with Gene Xpert MTB/RIF assay for rapid detection of Mycobacterium tuberculosis and rifampicin resistance in pleural biopsy samples among suspected cases of tubercular pleural effusion. **MATERIALS & METHODS-** Clinically suspected tubercular pleural effusion 100 cases were enrolled for the study. Following investigations were carried out: - Routine blood investigations (CBC, RBS, RFT, LFT, Electrolytes, HIV Test), Chest x-ray, Mantoux test, sputum for AFB. Pleural biopsy investigations- Histopathological examination, pleural biopsy culture by MGIT and pleural biopsy Gene Xpert MTB/RIF assay. Pleural fluid investigations- Cytological, biochemical, mycobacterial analysis and Gene Xpert MTB/RIF assay. **RESULTS:** Gene Xpert of pleural biopsy was performed on 100 patients .it yielded 28 positive cases which were rifampicin sensitive and 4 positive cases with rifampicin resistance & the rest were showing a negative report. **CONCLUSION-** The study used the gene Xpert MTB/RIF assay for direct Mycobacterium Tuberculosis detection in pleural fluid and pleural biopsy specimens in a high Tubercular endemic country. The study demonstrates that Gene Xpert MTB/RIF assay performs well in biopsy specimen for rapid and accurate diagnosis of pleural TB.

BACKGROUND-

India is the country with the highest burden of tuberculosis. Out of the estimated global annual incidence of 10 million tuberculosis cases in 2019, 2.8 million were estimated to have occurred in India with the estimated tuberculosis.

Among the extra pulmonary tuberculosis cases, tuberculosis lymphadenitis is the predominant manifestation followed by tubercular pleural effusion. In a study by Arora et al (2006) among EPTB cases, tuberculosis lymphadenitis was the predominant manifestation in 53.7%, followed by tubercular pleural effusion in 28.7%.

Pleural effusion patients happen to be common presenter in clinical setup of both respiratory & non-respiratory.

In practice, pleural fluid analysis including the microbiological, biochemical and cytological tests diagnose majority of the situation.

The Gene Xpert MTB /RIF assay is an automated, closed system that performs real time PCR and can be used by operators with minimal technical expertise, enabling diagnosis of tuberculosis and spontaneous assessment of rifampicin resistance within 2 hours

Several studies have evaluated the performance of gene Xpert MTB /RIF assay using pleural fluid. As there are limited studies about the use and efficacy of the gene Xpert MTB/RIF assay in pleural biopsy, there is a need for more research on this novel approach in pleural biopsies.

Materials & Methods- This study was a cross-sectional study. Suspected tubercular pleural effusion patients aged 14 years or more were en-rolled for this study. Total duration of study is 6 months.

The total sample size is 100. The pleural biopsy sample was tested for tuberculosis by smear microscopy, histopathology, MGIT culture and gene Xpert.

A sample which is found to be positive by any of three tests (smear microscopy, histopathology, MGIT culture) were considered as final positive as well as was considered as

composite reference standard and was compared with Gene Xpert.

Clinically suspected tubercular pleural effusion cases coming in outpatient department was enrolled for the study. The diagnosis of tubercular pleural effusion was established when any of the following criteria were met: 1) identification of bacilli in pleural fluid, or pleural biopsy specimen by direct smear or MGIT culture ; 2) presence of granuloma in pleural biopsy tissue ; 3) lymphocytic exudate with adenosine delaminates (ADA) levels > 35 U/l in the absence of any other obvious causes of pleural effusion .

Data was collected and analysed statistically. Numerical variables were reported as mean ± standard deviation and categorical variable as number and percentage.

INCLUSION CRITERIA:-

1) Patients aged > 14 years, with clinically suspected tubercular pleural effusion.

EXCLUSION CRITERIA:-

- 1) Sputum AFB positive cases.
- 2) Contraindications to pleural biopsy procedure like:-
 - Uncorrectable coagulation defects like Haemophilia ,low platelets or platelet function abnormalities, abnormal PT & PTT.
 - A Dry Tap
 - Empyema
 - Uncooperative patient
 - Uremia

Results-

The mean age of the enrolled patients was 32.41(±12.47) years with range of 16 years to 66 years. The majority of patients (70 %) were in the age group 20 to 45 years. This is comparable to results from previous studies. Jinghui et al (2015) from China reported the mean age of their patients as 38.6 ± 13.2 years, with male-to-female ratio of 1.25.18 Vadvai et al (2011) also reported median age of their patients as 37 years (range, 8 months to 94 years), with male-to-female ratio of 0.85.31

Pleural biopsy culture of the Patients

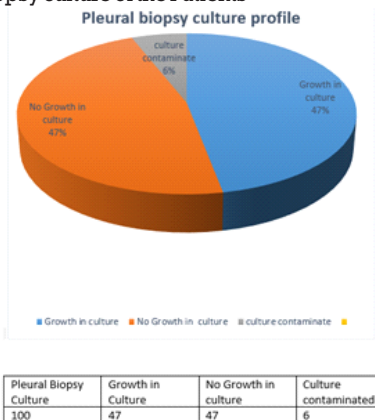


Figure - 1

The majority of the patient (80%) were found non-smokers. Rest of the twenty patients were smokers with a mean peak year history of 6.17.

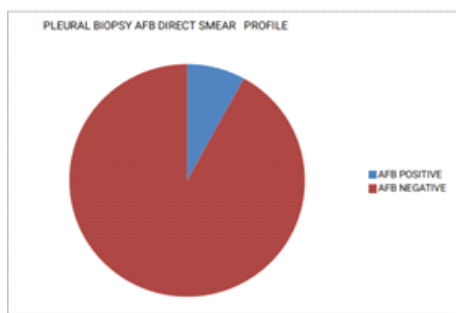


Figure - 2

Ninety percent of our patient had lymphocytic predominance in pleural fluid. Neutrophil predominance and mixed picture were found in five percent of cases each.

Sixty six percent of enrolled patients had granulomatous lesions in pleural biopsy samples. In majority of patients AFB in pleural biopsy direct smear microscopy only eight percent was positive for AFB.

Histopathology examination of pleural biopsy sample is better diagnostic modality as compared to the Gene Xpert assay for diagnosis of tubercular pleural effusion.

Gene Xpert assay of pleural biopsy sample is better diagnostic modality compared to the Gene Xpert assay of pleural fluid for diagnosis of tubercular pleural effusion.

DISCUSSION-

The main presenting respiratory symptoms of our enrolled patients were cough (52%), pleuritic chest pain (55%), and shortness of breath (8%). Constitutional symptoms included fever (42%), loss of appetite (35%), and loss of weight (39%). The finding is similar to that reported in previous studies of tubercular pleural effusion. In a similar study from France, Le Palud et al in 2014 found cough (51.9%) as the main symptom followed by general symptoms (45.1%).

Majority of our patients (73%) were found to have normal weight with a BMI of 18.5 to 22.9. Only two patients were overweight with BMI of more than 23. Twenty-five patients were underweight with BMI less than 18.5. Majority of our patients (80%) were non-smokers. Rest of the twenty patients were smokers with a mean pack year history of 6.17(±3.2). Half of our patients were educated only upto high school. Approximately two third were unskilled/semi-skilled workers or were students. Only six patients had a history of taking antitubercular treatment in the past. Four of these had taken treatment for pleural effusion while two had taken for

pulmonary tuberculosis. Lusiba et al (2014) from Uganda reported that participants with pleural tuberculosis were younger when compared to those without pleural tuberculosis [mean (±SD) age 32±11 years versus 41±15 years respectively, p,0.001] and were more likely to have a high temperature. There were more participants without pleural tuberculosis who smoked cigarettes when compared to participants with pleural tuberculosis [31% (9/29) versus 11.5%, (10/87), p = 0.014 respectively].

CONCLUSION-

This study demonstrates that Gene Xpert MTB/RIF assay performs well in biopsy specimens for rapid and accurate diagnosis of pleural tuberculosis. Pleural biopsy Gene Xpert assay was found positive in 28% cases; among these, four cases were rifampicin resistant. The sensitivity and specificity of the Gene Xpert MTB/RIF assay in pleural biopsy against culture were 70% and 100% respectively; against the histopathological examination, were 57% and 100% respectively, and against our Composite Reference Standard were 28% and 100% respectively.

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