Original Article

Clinical and laboratory differences and role of gene xpert in tuberculous pleural effusion

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Abstract

Background: The diagnosis of pleural effusion and its cause are essential for pleural fluid analysis. We have evaluated clinical and laboratory differences among the tubercular pleural effusion.

Methods: The cross-sectional, observational hospital based study was conducted in Bir hospital, Nepal. All patients were evaluated by clinically and laboratory investigations. Patients enrolled for study have pleural effusion and pleural fluid analysis indicative of an exudative pleural effusion using lights criteria. The criteria of enrollment of the patients were pleural fluid for Adenosine deaminizes value more than 40 IU/L, positive for gene xpert test and pleural effusion of any cases with sputum positive pulmonary tuberculosis. Patients were divided into two groups lymphocytic and neutrophilic predominant pleural effusion.

Results: Among 100 patients with diagnosis of exudative tubercular pleural effusion, the most common symptom was pleuritic chest pain in 85%, followed by fever in 84% and cough in 82%. Among the tubercular pleural effusion, 21% had neutrophils predominant and 79% had Lymphocytes predominant. The patients with neutrophil predominant Tubercular pleural effusion had higher fever rates (90.5vs.82.5%) than those with lymphocyte-predominant Tubercular pleural effusion. The mean value of Neutrophil predominant pleural fluid for lactate dehydrogenase (LDH) level was 1657.5 IU/L and protein was 5.3gm/dl and in lymphocyte predominant pleural fluid for LDH value was 610.2 IU/L and protein was 4.6 gm/dl; the difference wasstatistically significant with P value of <0.001. Only 15% of patients had sputum positive for Acid fast bacilli. Among the sputum positive patients, 47% had positive for pleural fluid for gene xpert test with all patients had rifampicin sensitive. The sensitivity of pleural fluid for gene xpert test was 46.6%, and specificity was 90%.

Conclusion: In pleural effusion, the positivity of gene xpert for pleural fluid was higher among the sputum positive patients. The prevalence of Neutrophil-predominant pleural effusion was common in tubercular pleural effusion.

Introduction

Tuberculosis (TB) is a major global health burden worldwide, but there is significant variation in different geographical areas. Tuberculous pleural effusion is a second common extra pulmonary manifestation in poor country like Nepal.[1,2] Furthermore, it is also complicated by the emergence of MDR. Gene Xpert MTB/RIF is promising innovation for the detection of mycobacterium tuberculosis and its resistance to Rifampicin. The gold standard of pleural TB diagnosis requires detection of Mycobacterium tuberculosis (MTB) in pleural fluid or histological demonstration of a caseous granuloma in the pleura.[3-5] However, due to its paucibacillary in origin, its diagnosis is a challenge. Therefore,

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pleural fluid analysis is essential for diagnosing pleural TB. Previously, lymphocyte-predominant exudates with high adenosine deaminase (ADA) have been classified as pleural TB. Nonetheless, the diagnosis of pleural TB using pleural fluid analysis is still challenging. While previous studies have shown that lymphocytes constitute up to 90% of total cells in pleural fluid with pleural TB [3,6]. Various studies have reported lymphocytes in pleural fluid decreased in patients who were diagnosed with pleural TB[3,7] and encounter neutrophil-predominant pleural fluid3[,8].The objectives of this study was to find clinical and laboratory differences between lymphocyte and neutrophil-predominant Tuberculous pleural effusion and find out the role of gene xpert for the evaluation and its applicability in Nepalese populations.

Materials and methods

Study population

This study included consecutive, adult patients (age ≥17 years) with newly diagnosed Tuberculous Pleural Effusion at Bir Hospital, Kathmandu, Nepal. All included patients underwent thoracentesis at least once during diagnostic workup. Tubercular Pleural effusion was diagnosed as indicated in "Diagnostic criteria", as mentioned below:

1. Age more than or equal to 16 years

2.Patients with clinically and radiologically suspected to have pleural effusion and who had pleural fluid analysis indicative of an exudative pleural effusion using lights criteria with pleural fluid ADA value more than 40 IU/L

3.Any cases of pleural effusion in sputum positive pulmonary tuberculosis.

Patients admitted in medical ward or emergency of Bir Hospital with clinical signs and symptoms suggestive of pleural effusion was evaluated and enrolled after taking the informed consent. Routine investigations including WBC counts and chest x-ray was performed for each patient. Investigations like USG of chest in case of minimal or suspected pleural effusion on chest X-ray will be done.

Those patients with pleural effusion was undergo thoracocentesis. 50 ml of pleural fluid was obtained by a disposable plastic syringe maintaining all aseptic precautions. The plain sample of pleural fluid thus obtained will be sent immediately to the laboratory for analysis. If analysis of these samples was not be feasible immediately, it was kept in refrigerated till analysis was done within 24 hours of sample collection. The pleural fluid was analyzed for total protein, LDH, glucose, TC, DC, malignant cells, ADA, Gram's stain, AFB stain, bacterial culture and gene xpert along with simultaneous serum sample for total protein and LDH levels was measured by the standard method practiced in the hospital laboratory. Simultaneously pleural fluid was send for gene xpert if clinically suspicious of tubercular pleural effusion or refrigenated sample or by re aspiration of samples are send for gene xpert if ADA value is more than 40 IU/L

If at least one of the following three lights criteria is fulfilled, the fluid is defined as an exudate The cutoff value for pleural fluid for ADA >40 IU/L was taken to classify pleural fluid as a case of tubercular pleural effusion in this study. The sensitivity of the gene xpert for the diagnosis of tubercular pleural effusion (with ADA >40) was seen in this study.

The reports of the total protein concentration, LDH, ADA and GeneXpert of the pleural fluid and serum concentration of total protein and LDH level were tabulated for each patient. Each patient was divided into two groups: 1st group lymphocytic predominant pleural fluid where presence of more than 50% of lymphocytes, 2nd group neutrophil predominant pleural fluid where presence of more than 50% of neutrophils. All relevant data including final diagnosis on discharge were then compared.

Results

In this study 100 patient with diagnosis of exudative Tubercular pleural effusion were included. Among them, the mean age was 35.35±14.62 years(17-73yrs) with 44% were male and 56% were female. Thus male to female ratio was 1:1.27. Most of the patients were between the age group of 15-35 years which constituted the 59%.

Table-1: Clinical symptoms of patients

Clinical features	Male (%, N)	Female (%, N)	Total (N=100 or %	P value
Fever	84.09(37)	83.9(47)	84	0.98
Cough	88.6(39)	76.8(43)	82	0.12
Pleuritic Chest pain	84.1(37)	85.7(48)	85	0.82
Hemoptysis	18.2(8)	21.4(12)	20	0.68
Shortness of Breath	59.1(26)	57.1(32)	58	0.84
Weight loss	34.1(15)	33.9(19)	34	0.98
Loss of Appetite	40.9(18)	35.71(20)	38	0.59

As shown in table 1, the most common symptom was pleuritic chest pain in 85%, fever in 84% and cough in 82%, followed by shortness of breath 58%. Other symptoms were loss of appetite in 38%, hemoptysis 20%, loss of weight in 34% patients.

Among 100 tuberculous pleural effusions, 39% was the current smoker and 16% was alcohol consumer. The gender disaggregated analysis indicated that more male (52.3% and 27.27) than female (28.57 and 7.1 %) were smoker and alcohol consumer and the association was statistically significant at $p<0.01$.

Table 2. Comparison of clinical characteristics between lymphocyte-predominant and neutrophil-predominant tuberculous effusion

Clinical features	Lymphocyte (n=79) number (%)	Neutrophil (n=21)	P value
Fever	65(82.3	19(90.5)	>0.05
Cough	64(81)	18(85.7)	>0.05
Pleuritic Chest pain	64(81)	21(100)	0.03
Hemoptysis	15(19)	5(23.8)	>0.05
Shortness of Breath	46(58.2)	12(57.1)	>0.05
Loss of Appetite	29(36.7)	9(42.9)	>0.05
Hb<12 gm/dl	45(57)	14(66.7)	>0.05
ESR>20mm in 1st hr	77(97.5)	21(100)	>0.05
PF LDH/ADA<16.2	64(81.01)	9(42.8)	0.006

Table 2 demonstrates comparison between the symptoms and laboratory finding between Lymphocytes and neutrophil predominant tuberculous effusion. The patients with neutrophil predominant TPE had higher high fever rates (90.5vs.82.5%) than those with lymphocyte-predominant TPE. In addition, the patients with PMNL-predominant TPE had PF LDH/ADA<16.2 (42.8vs 64). However, it was statistically significant between PF LDH/ADA ratio (p<0.05).

Table 3. Comparison of Pleural fluid analysis of Lymphocyte- and Neutrophil-Predominant Pleural TB (N=100)

Parameters	Lymphocyte (79%)	Neutrophil (21%)	P-value
Pleural fluid ADA (IU/L)	65.5	58.7	0.000
Pleural fluid LDH (IU/L)	610.2	1657.5	0.000
Pleural fluid Protein (mg/dL)	4.6	5.3	0.002
Pleural fluid Glucose (mg/dL)	85.0	59.9	0.000

Table 3 shows mean value of Pleural fluid ADA and glucose among lymphocytes predominant was 65.5U/Land 85mg/dl and 58.7U/L and 59.9mg/dl among Neutrophil predominant and the difference was statistically significant at <0. 001.Similarly Pleural fluid LDH and protein were higher in Neutrophil predominant in which mean LDH and protein were 1657.5 IU/Land 5.3gm/dl and the difference were statistically significant at <0.001.

Table 4. Microbiological Characteristics of Lymphocyte- and Neutrophil-Predominant Pleural TB

Parameters	Total (n =100) (%, N)	Lymphocyte (n =79) (%, N)	Neutrophil (n = 21) (%, N)	P-value
Sputum for AFB	15.0 (15)	15.2 (12)	14.3 (3)	0.918
Pleural fluid GeneXpert (PCR)	15.0 (15)	17.7 (14)	4.8 (1)	0.139

Table 4 shows that out of 100 TB effusion patients only 15% had sputum positive AFB in which 15.2% and 14.3% belong to lymphocytes and neutrophil predominant in which 7 patients had positive GeneXpert. Similarly, Pleural GeneXpert (PCR) was 17.7% in Lymphocyte predominant and 4.8% in Neutrophil Predominant Tubercular pleural effusion. However, there was no statistically significant.

Table 5. Distribution of sputum for AFB and pleural fluid for GeneXpert

	Pleural Fluid GeneXpert		Total
	Positive	Negative	
Sputum for AFB			
Positive	7	8	15
Negative	8	77	85

Out of 100 patients, 15 patients had pleural effusion for GeneXpert positive and all patients was rifampicin sensitive (100%). The sensitivity of pleural fluid for gene xpert test was 46.6%, and specificity was 90% .

Discussion

TPF is a form of extra pulmonary tuberculosis that is difficult to diagnose clinically. Lymphocyte-based exudate and high ADA>40U/L have been considered to be a part of the diagnostic criteria in high prevalence country like Nepal. This study was conducted to find clinical and laboratory differences between lymphocyte and neutrophil-predominant Tuberculous pleural effusion and find out the role of GeneXpert for the evaluation and its applicability in Nepalese populations.

In this study 100 patient with diagnosis of exudative Tubercular pleural effusion were included. Among them, the mean age was 35.35±14.62 years(17-73yrs) with 44% were male and 56% were female. Thus male to female ratio was 1:1.27. Most of the patients were between the age group of 15-35 years which constituted the 59% and pleural effusion was more common in female. Zhao T at el showed the average age was 35.08 ± 14.55 years old[9] and TPE affects mainly younger individuals (in higher tuberculosis burden areas, where primary infection accounts for a large percentage of patients with TPE.[10] This finding was similar to our study in which the younger patient had TPE (15-35 years).[11]

In this study the most common symptom was pleuritic chest pain in 85%, fever in 84% and cough in 82%, followed by shortness of breath 58%. This finding was similar to the study done by Zhai at el. pe´rez-Rodriguez E at el, Bansal P et at and Ferreiro L et al.[12-15]

Similarly, in this study, 39% was the current smoker and 16% was alcohol consumer. The gender disaggregated analysis indicated that more male (52.3% and 27.27) than female (28.57 and 7.1 %) were smoker and alcohol consumer and the association was statistically significant at p<0.01. This study was supported by Tewatia P et al which suggest that smoker and alcohol consumer had an independent association with TPE.[16]

In our study Pleural fluid ADA and glucose was higher in lymphocytes predominant and was statistically significant at <0. 001. Similarly, Pleural fluid LDH and protein were higher in Neutrophil predominant and the difference were statistically significant at <0.001. This study was supported by Zhao T et al. The biochemical fluid characteristics of Neutrophil predominant effusions (i.e., high LDH, low glucose) reflected an intense degree of pleural inflammation.[9,17]

In this study,15% had sputum positive AFB in which 15.2% and 14.3% belong to lymphocytes and neutrophil predominant. Similarly, Pleural GeneXpert (PCR) was 17.7% in Lymphocyte predominant and 4.8% in Neutrophil Predominant Tubercular pleural effusion. However, there was no statistically significant. similar result was seen on a study by Vorster MJ et al showing the sensitivity ranging from 0-30% in sputum AFB.[18]

In our study, among 100 patients with tubercular pleural effusion (ADA >40IU/L) pleural fluid gene xpert came positive in 15 cases (15%). Similar study was conducted in different centres. A study conducted by John k Lusiba for the role of gene xpert in histolologically or culture proven cases showed that Xpert MTB/Rif test was positive in 25 of the 87 pleural TB confirmed participants and the sensitivity and specificity were 28.7% (25/87) and 96.6% (28/29) respectively while the positive and negative predictive values were 96.1% and 31.1% respectively. [19]

Conclusion:

In pleural effusion, the positivity of gene x-pert for pleural fluid was higher among the sputum positive patients. The prevalence of Neutrophil-predominant pleural effusion was common in tubercular pleural effusion.

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