

CORRELATION OF FLUID ADA & CBNAAT IN THE DIAGNOSIS OF TUBERCULOSIS IN EXUDATIVE PLEURAL EFFUSION

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Abstract

Introduction: Tuberculosis (TB) remains a major cause of morbidity and mortality throughout the world. The World Health Organization (WHO) estimated nearly 10.4 million new cases globally in 2015, with maximum burden in the developing world. Pulmonary TB is the commonest form of TB, and a definite microbiological diagnosis is possible in most instances through sputum microscopy, culture and the use of cartridge-based nucleic acid amplification tests (CBNAAT) and fluid ADA levels. The present study was undertaken to estimate the levels of pleural fluid ADA levels and CBNAAT testing in patients with exudative pleural effusion and to study the diagnostic accuracy of ADA and CBNAAT testing in suspected cases of tubercular exudative pleural effusion.

Materials and methods: After obtaining informed consent, 100 cases of pleural effusion were subjected for following Investigations-Medical history taking, full clinical examination, Sputum for AFB smear and culture, diagnostic thoracentesis under aseptic conditions. Aspirated pleural fluid was sent to biochemical tests sugar, protein, LDH, microbiological tests –AFB stain, total cytological count and differential count. Simultaneously Serum protein and serum LDH were also measured. All patients fulfilling Light's criteria for exudative pleural effusion were further subjected to ADA estimation and CBNAAT testing.

Results: We included a total of 100 patients as per inclusion and exclusion criteria aged >20 years and < 70 years, out of which 78 were males and 22 were females. Majority of the patients were in the age group of 21-30 years representing 32%. We evaluated clinical features in these subjects, it is found that 86% had cough, 78% fever, 63% chest pain, 62% loss of appetite, 56% shortness of breath (SOB) and 53% presented with weight loss. Fluid ADA levels were measured in all the patients, with cut-off values of 40 units/L, 68 patients had ADA levels >40 units /L and 22 had <40 units/L. CBNAAT testing was done in all the patients out of which in 42 patients MTB was detected and in 32 patients MTB was not detected. Similarly, out of 22 patients (ADA ≤40 units /L), none of the patients had CBNAAT positive. Diagnostic accuracy of ADA and CBNAAT showed 64.3%, 26% sensitivity, 33.8% and 100 % specificity, 93.6 and 100% PPV, 6.4% and 10.2%, NPV, 58 and 26.6% diagnostic accuracy.

Discussion and conclusion: Diagnosis of tubercular exudative pleural effusion is the real challenge faced in the day to day management of the patients who come to OPD with symptoms of suspected tuberculosis. Each and every laboratory investigation plays an key role in the diagnosis and further

antituberculosis drug therapy approach. Before we start the therapy to the patients it is highly essential to ensure the diagnosis of tubercular exudative pleural effusion. Each component i.e., history, physical examination, blood investigations, fluid analysis, ADA estimation, and CBNAAT are important in evaluation of such cases and provide some diagnostic clues. Relying solely on single diagnostic modality may be associated with low diagnostic yield, and thus each step of patient assessment must be given equal preference to improve the diagnostic yield. However, we report that ADA as better tool as compared to CBNAAT for diagnosis of tubercular exudative pleural effusion.

Key-words: tuberculosis, exudative pleural effusion, adenosine deaminase, cartridge based nucleic acid amplification test.

INTRODUCTION

Tuberculosis (TB) remains a major cause of morbidity and mortality throughout the world. The World Health Organization (WHO) estimated nearly 10.4 million new cases globally in 2015, with maximum burden in the developing world. Pulmonary TB is the commonest form of TB, and a definite microbiological diagnosis is possible in most instances through sputum microscopy, culture and the use of cartridge-based nucleic acid amplification tests (CBNAAT) and fluid ADA levels. The present study was undertaken to estimate the levels of pleural fluid ADA levels and CBNAAT testing in patients with exudative pleural effusion and to study the diagnostic accuracy of ADA and CBNAAT testing in suspected cases of tubercular exudative pleural effusion.

MATERIALS AND METHODS:

An observational study was done on 100 patients presenting with symptoms, medical history, radiological picture suggestive of pleural effusion, either admitted or attending OPD of Department of Department of respiratory medicine were enrolled in the study. We included the patients aged >20 years with a medical history suggestive of pleural effusion, Patients with pleural effusion were identified by-Clinical examination, chest x-ray, ultrasonography, and diagnostic thoracentesis for fluid analysis showing exudative nature. We excluded the patients aged <20 years, those not willing to give consent, patients with transudate pleural effusion and hemothorax.

After obtaining informed consent,100 cases of pleural effusion were subjected for following Investigations-Medical history taking, full clinical examination, Sputum for AFB smear and culture, diagnostic thoracentesis under aseptic conditions. Aspirated pleural fluid was sent to biochemical tests sugar, protein, LDH, microbiological tests –AFB stain, total cytological count and differential count. Simultaneously Serum protein and serum LDH were also measured. All patients fulfilling Light's criteria for exudative pleural effusion were further subjected to ADA estimation and CBNAAT (Cartridge based nucleic acid amplification test).

In the study, presence of first or more than one of the following criteria was adopted to label a case as tuberculosis

1. Bacteriological confirmation of the presence of Mycobacterium tuberculosis in pleural fluid or sputum by ZN stain and CBNAAT testing.
 2. Exudative (according to Light's criteria), lymphocytic pleural effusion with ADA>40 units/L.
 3. Definite clinical and radiological improvement in 2 months of administration of exclusive ATT.
- All cases of malignancy diagnosed on cytology by pleural fluid, showing the malignant cells.

RESULTS:

Table 1: Shows age wise and gender wise distribution of study subjects

	Number	Percentage
21-30 years	32	32
31-40 years	26	26
41-50 years	18	18
51-60 years	12	12
61-70 years	10	10
Males: Females	78/22	78%/22%

Table 2: Shows clinical features in study population

	Number	Percentage
Cough	86	86
Fever	78	78
Chest pain	63	63
Loss of appetite	62	62
SOB	56	56
Weight loss	53	53

Table 3: Shows pleural fluid ADA levels

ADA≤40 U/L	22	22
ADA>40 U/L	78	78
Total	100	100

Table 4: Shows pleural fluid ADA levels and CBNAAT status in study subjects

	No of cases	MTB CBNAAT positive	MTB CBNAAT Negative
ADA≤40 U/L	22	0	22
ADA>40 U/L	78	42	36
Total	100	42	58

Table 5: Shows Diagnostic accuracy of ADA and CBNAAT

	ADA	CBNAAT
Sensitivity	64.3	26
Specificity	33.8	100
Positive predictive value	93.6	100
Negative predictive value	6.4	10.2
Diagnostic accuracy	58	26.6

DISCUSSION AND CONCLUSION:

Tuberculosis (TB) remains a major cause of morbidity and mortality throughout the world. The World Health Organization (WHO) estimated nearly 10.4 million new cases globally in 2015, with maximum burden in the developing world [1]. Pulmonary TB is the commonest form of TB, and a definite microbiological diagnosis is possible in most instances through sputum microscopy, culture and the use of cartridge-based nucleic acid amplification tests (CBNAAT).

Adenosine deaminase (ADA), an enzyme produced from lymphocytes and involved in purine metabolism, has been extensively studied as a biochemical marker in pleural fluid during investigation for TPE. The test is simple, cheap, rapid, minimally invasive, and can be performed in most

laboratories [3]. Although pleural fluid ADA is not a perfect discriminator, its level is considerably elevated in patients with TPE. High ADA levels can sometimes be observed in pleural fluid from patients of empyema, malignancy, or rheumatoid pleurisy [4-6]. Therefore, presence of raised pleural fluid ADA is considered a useful marker for diagnosis of TPE, especially in patients with exudative and lymphocytic pleural effusion in high TB burden settings. These patients can empirically be started on anti-tuberculous therapy if no other investigation can provide a definite diagnosis. Cartridge-based nucleic acid amplification test: CBNAAT is based upon the principle of Polymerase Chain Reaction (PCR) which helps in rapid detection of Mycobacterium tuberculosis. For CBNAAT, 1ml sputum sample was taken from the patients. Manual procedure: Addition of sample treatment reagent to liquefy and inactivate the bacteria in the sputum, transfer of 2ml of liquefied sputum into the cartridge and loading of the cartridge into the device for the assay. All further steps are automated. It targets rpoB gene and helps in identification of rifampicin resistance as well. This method is highly specific as this method uses 3 primers and 5 molecular probes which specifically target rpoB gene of Mycobacterium tuberculosis.

We included a total of 100 patients as per inclusion and exclusion criteria aged >20 years and <70 years, out of which 78 were males and 22 were females. Majority of the patients were in the age group of 21-30 years representing 32%. We evaluated clinical features in these subjects, it is found that 86% had cough, 78% fever, 63% chest pain, 62% loss of appetite, 56% shortness of breath (SOB) and 53% presented with weight loss. Fluid ADA levels were measured in all the patients, with cut-off values of 40 units /L, 68 patients had ADA levels >40 units/L and 22 had <40 units /L. CBNAAT testing was done in all the patients out of which in 42 patients MTB was detected and in 32 patients MTB was not detected. Similarly, out of 22 patients (ADA ≤40 units /L), none of the patients had CBNAAT positive. Diagnostic accuracy of ADA and CBNAAT showed 64.3%, 26% sensitivity, 33.8% and 100 % specificity, 93.6 and 100% PPV, 6.4% and 10.2%, NPV, 58 and 26.6% diagnostic accuracy. The findings of our study were in agreement with the studies conducted in the past [7-14].

CONCLUSION:

Diagnosis of tubercular exudative pleural effusion is the real challenge faced in the day to day management of the patients who come to OPD with symptoms of suspected tuberculosis. Each laboratory investigation plays a key role in the diagnosis and further antituberculosis drug therapy approach. Before we start the therapy to the patients it is highly essential to ensure the diagnosis of tubercular exudative pleural effusion. Each component i.e., history, physical examination, blood investigations, fluid analysis, ADA estimation, and CBNAAT are important in evaluation of such cases and provide some diagnostic clues. Relying solely on single diagnostic modality may be associated with low diagnostic yield, and thus each step of patient assessment must be given equal preference so as to improve the diagnostic yield. However, we report that ADA as better tool as compared to CBNAAT for diagnosis of tubercular exudative pleural effusion.

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