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Original Research Article

STUDY OF DIAGNOSTIC VALUE OF PLEURAL FLUID ADA AND CBNAAT IN PULMONARY TUBERCULOSIS

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Abstract

Introduction: 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, 60 cases of pleural effusion were subjected for following Investigations-Medical history taking, full clinical examination, Sputum for AFB smear and culture, diagnostic thoracocentesis under aseptic conditions. Aspirated pleural fluid was sent to biochemical tests sugar, protein, LDH, microbiological tests –AFB stain, total cytological count and differential count. 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 60 patients as per inclusion and exclusion criteria aged >30 years and <70 years, out of which 48 were males and 12 were females. Majority of the patients were in the age group of 31-40 years representing. Fluid ADA levels were measured in all the patients, with cut-off values of 40 U/L. 44 patients had ADA levels >40U/L and 16 had <40 U/L.
CBNAAT testing was done in all the patients, out of 44 patients MTB CBNAAT was positive in 33 cases and in 11 cases it was negative. Similarly, out of 16 patients (ADA <=40 U/L), none of the patients had CBNAAT positive.

Diagnostic accuracy of ADA and CBNAAT showed 65.3%, 27% sensitivity, 35.6% and 100 % specificity, 94 and 100% PPV, 7% and 10.4%, NPV, 59 and 28% 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. 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:

Source and type of study: An observational study was done on 60 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 who were willing to give voluntary consent for the study.

Inclusion criteria: We included the patients aged >30 years with a medical history suggestive of pleural effusion, Patients with pleural effusion were identified by-Clinical examination, chest x-ray, ultrasonography, and diagnostic thoracocentesis for fluid analysis showing exudative nature.

Exclusion criteria: We excluded the patients aged <30 years, those not willing to give consent, patients with transudate pleural effusion and hemothorax.

Methodology: After obtaining informed consent, 60 cases of pleural effusion were subjected for following Investigations-Medical history taking, full clinical examination, Sputum for AFB smear and culture, diagnostic thoracocentesis 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 tuberculous

1. Bacteriological confirmation of the presence of Mycobacterium tuberculosis in pleural fluid or sputum by z-n stain and CBNAAT testing.
2. Exudative (according to Light's criteria), lymphocytic pleural effusion with ADA>40IU/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
31-40 years	24	40%
41-50 years	16	26.6%
51-60 years	12	20%
61-70 years	8	13.33%
Males: Females	48/12	

Table 2: Shows clinical features in study population		
	Number	Percentage
Cough	60	100%
Fever	60	100%
Chest pain	42	70%
Loss of appetite	40	66.66%
SOB	38	63.33%
Weight loss	39	65%

Table 3: Shows pleural fluid ADA levels and CBNAAT status in study subjects			
	No of cases	MTB CBNAAT positive	MTB CBNAAT Negative
ADA<=40 U/L	16	0	16
ADA>40 U/L	44	33	11
Total	100	33	28

Table 4: Shows Diagnostic accuracy of ADA and CBNAAT		
	ADA	CBNAAT
Sensitivity	65.3	27
Specificity	35.6	100
Positive predictive value	94	100
Negative predictive value	7	10.4
Diagnostic accuracy	59	28

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 U/L, 68 patients had ADA levels >40U/L and 22 had <40 U/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 U/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.

REFERENCES:

1. Langer AJ, Iqbal SA, Pratt R, et al. Tuberculosis statistics in United States. Report, United States Department of Health and Human Services, Public Health Services, Centres for Disease Control, Atlanta, GA, 1989, 1991.

2. Komanapalli SK, Prasad U, Atla B, Vasundhara N, Yendluri D. Role of CB-NAAT in diagnosing extra pulmonary tuberculosis in correlation with FNA in a tertiary care center. Int J Res Med Sci. 2018;6(12):4039-45.

3. TB Statistics India. Available from: https://tbfacts.org/tb-statistics-india/. Last accessed on 17th July 2021.

4. World Health Organisation (WHO). Global TB Report 2019. https://www.who.int/ tb/publications/global_report/en/Last accessed on 17th July 2021.

5. Solovic I, Jonsson J, Korzeniewska-Koseła M, Chiotan DI, Pace-Asciak A, Slump E, et al. Challenges in diagnosing extrapulmonary tuberculosis in the European Union, 2011. Euro Surveill. 2013;18(12):20432.

6. Sumi S, Radhakrishnan VV. Evaluation of Immunohistochemistry with a panel of antibodies against recombinant mycobacterial antigens for the diagnosis of tuberculous lymphadenitis. International Journal of Medicine and Medical Sciences. 2009;1(5):215-19.

7. Tuon FF, Higashino HR, Lopes MI, Litvoc MN, Atomiya AN, Antonangelo L, et al. Adenosine deaminase and tuberculous meningitis-a systematic review with meta-analysis. Scand J Infect Dis. 2010;42(3):198-207.

8. Riquelme A, Calvo M, Salech F, Valderrama S, Pattillo A, Arellano M, et al. Value of adenosine deaminase (ADA) in ascitic fluid for the diagnosis of tuberculous peritonitis: A meta-analysis. Journal of Clinical Gastroenterology. 2006;40(8):705-10.

9. Tuon FF, Litvoc MN, Lopes MI. Adenosine deaminase and tuberculous pericarditis: A systematic review with meta-analysis. Acta Trop. 2006;99:67-74.

10. National Tuberculosis Elimination Programme. Central TB Division. Available from: https://tbcindia.gov.in/WriteReadData/l892s/India%20TB%20Report%20 2020.pdf. Last

accessed on 17th July 2021.

11. World Health Organization: WHO report 2010: Global tuberculosis control. Geneva, Switzerland: WHO; 2010.

12. Lee JY. Diagnosis and treatment of extrapulmonary tuberculosis. Tuberculosis and Respiratory Diseases. 2015;78(2):47-55.

13. ADA Diazyme method. Available from: https://www.diazyme.com/adenosine- deaminase-ada. Last accessed on 16th February 2022.

14. Rachow A, Zumla A, Heinrich N, Rojas-Ponce G, Mtafya B, Reither K, et al. Rapid and accurate detection of Mycobacterium tuberculosis in sputum samples by Cepheid Xpert MTB/RIF assay-a clinical validation study. PLoS One. 2011;6(6):e20458.