



ADENOSINE DEAMINASE AS A DIAGNOSTIC MARKER FOR TUBERCULAR PLURAL EFFUSION: A COMPARATIVE ANALYSIS

Anita Bisht^{1*}, Mohammad Shahrukh², Sumita Sharma³

^{1*}Tutor, Department of Biochemistry, Shri Ram Murti Smarak Institute of Medical Sciences, Bareilly, U.P. Email ID: bishtanita533@gmail.com

²Assistant Professor, Department of Biochemistry, Rohilkhand Medical College & Hospital, Bareilly, U.P. Email ID: mohammadshahrukh.saifi@gmail.com

³Additional Professor Department of Biochemistry, AIIMS Bilaspur, Himachal Pradesh. Email ID: drsumita17@gmail.com

***Corresponding Author: Anita Bisht**

*Tutor, Department of Biochemistry, Shri Ram Murti Smarak Institute of Medical Sciences, Bareilly, U.P. Email ID: bishtanita533@gmail.com

Abstract

Background: Tuberculosis is the single most frequent infectious cause of deaths worldwide and also the commonest cause of pleural effusion. The measurement of level of adenosine deaminase (ADA) enzyme in body fluids is a helpful diagnostic tool. The level of ADA elevates as the lymphocytic activity increases. The present study was carried out to assess the ADA activity in pleural fluid for early diagnosis and management of tubercular pleural effusion.

Objective: To determine the diagnostic value of ADA in differentiating tubercular and non-tubercular pleural effusion based on sensitivity and specificity.

Materials and Methods: The present study was conducted on 56 tubercular pleural effusion patients and 54 non-tubercular pleural effusion patients who attended the OPD of Shri Guru Ram Rai Institute of Medical and Health Sciences, Dehradun. A structured proforma was used for documenting the history, symptoms and various biochemical investigations like glucose, protein and ADA activity of these patients.

Result: The study suggested that ADA levels were significantly higher ($p\text{-value} = < 0.0001$) in cases as compared to controls. The sensitivity and specificity of ADA for the diagnosis of tubercular pleural effusion are 85.7% and 88.9% respectively.

Conclusion: ADA levels are noticeably raised in tubercular pleural effusion and showed good diagnostic accuracy. ADA estimation is simple, reliable, cost-effective and diagnostically useful investigation.

Keywords: ADA, Pleural fluid, Tuberculosis and Tubercular pleural effusion.

INTRODUCTION

Tuberculosis is the single most frequent infectious cause of death worldwide and also is one of the leading causes of pleural effusion caused by *Mycobacterium tuberculosis* [1]. Pleural effusion is defined as an excessive accumulation of fluid in the pleural space; it indicates an imbalance between pleural fluid formation and absorption. Each year approximately 1 million patients develop

pleural effusion [2]. It remains a disease with a high rate of mortality worldwide especially in developing countries [3]. Extra-pulmonary tuberculosis occurs in about 10% -20% of patients and the most common form involved is tubercular pleural effusion.

Around the world, TB accounts for 30–60% of pleural effusions [4]. Adenosine deaminase (ADA) is an enzyme involved in purine metabolism. It is present in lymphocytes in high concentration [5]. ADA contains two isoenzymes, namely ADA-1 and ADA-2. ADA-1 is a ubiquitous enzyme that may be produced by many different cell types, particularly lymphocytes and macrophages. On the other hand, ADA-2 is secreted only by monocytes and macrophages and is the predominant isoenzyme (85%) in tubercular pleural effusion. Ever since ADA was first reported in 1978, the measurement of pleural fluid ADA has consistently demonstrated a high accuracy for diagnosing tubercular pleural effusion [6-8]. The present study was planned to evaluate the diagnostic utility of ADA in distinguishing tubercular from non-tubercular pleural effusion by assessing its sensitivity and specificity.

MATERIAL AND METHODS

The study was conducted on 56 tubercular pleural effusion patients (TPE) and 54 non- TPE subjects who attended the OPD of SGRRIM&HS, Dehradun, over a period of 06 months.

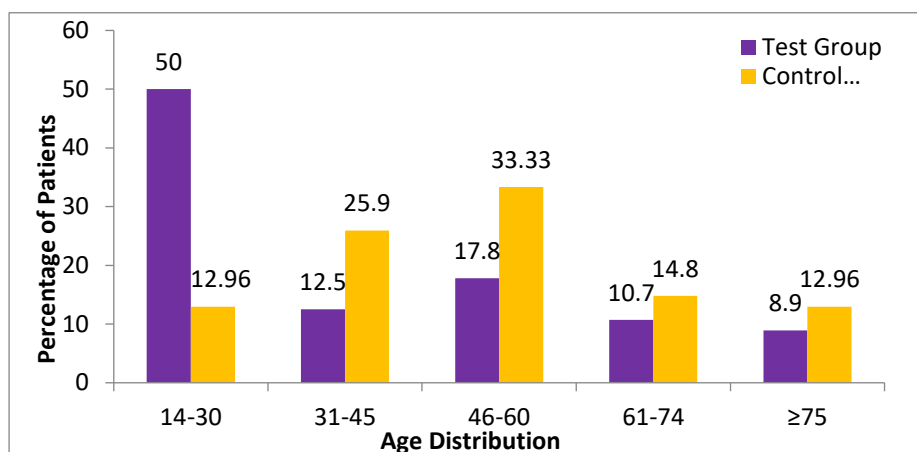
Classification of pleural effusions as tubercular and non-tubercular was based on clinical and biochemical data available at the time of diagnosis. Exclusion criteria was age below 14 years. Glucose and protein were estimated by fully automated Vitros5600 integrated system and ADA was estimated by semi auto-analyzer method with standard protocols. Mean, standard deviation (SD) was calculated by descriptive analysis tool and p-value was calculated by student t- test.

RESULT

110 subjects (56 cases and 54 controls) were involved in the study. Pleural aspirate samples were examined for ADA activity, glucose and protein levels in test and control groups. These patients were examined for different ADA levels in test group (mean $\pm$ SD= 103.29 $\pm$ 61.26) & in control group (mean $\pm$ SD= 25 $\pm$ 18.2). Graph chart 1 shows different ADA levels in test and control group, Graph chart 2 shows comparison between ADA activity, glucose and protein levels in test group and control group. (** <0.01 highly significant, * <0.05 significant). Age distribution in study group showed peak level in age between 14-30 years represented by 28 patients (50%) and the least number of patients were in the age group $\geq$ 75 years represented by 5 patients (8.92%). Age distribution in control group showed peak level in age between 46-60 years represented by 18 patients (33.33%) and the least were in the group 14-30 years and $\geq$ 75 years represented by 7 patients (12.96%).

AGE (IN YEARS)	NO. OF PATIENTS			
	STUDY GROUP		CONTROL GROUP	
	Number	(%)	Number	(%)
14-30	28	50	7	12.96
31-45	7	12.5	14	25.92
46-60	10	17.85	18	33.33
61-74	6	10.71	8	14.81
$\geq$ 75	5	8.92	7	12.96
Total	56	100	54	100

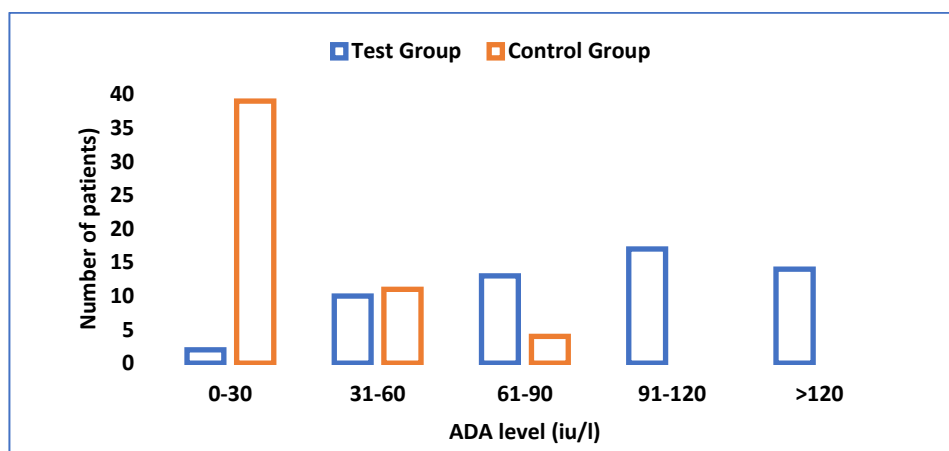
Table-1: Distribution of study group and control group according to age.



Graph Chart-1: Percentage of patients according to age in study group and control group.

ADA ACTIVITY(IU/L)	STUDY GROUP		CONTROL GROUP	
	Number	Percentage (%)	Number	Percentage (%)
0-30	2	3.57	39	72.22
31-60	10	17.85	11	20.37
61-90	13	23.21	4	7.40
91-120	17	30.35	0	0
>120	14	25	0	0

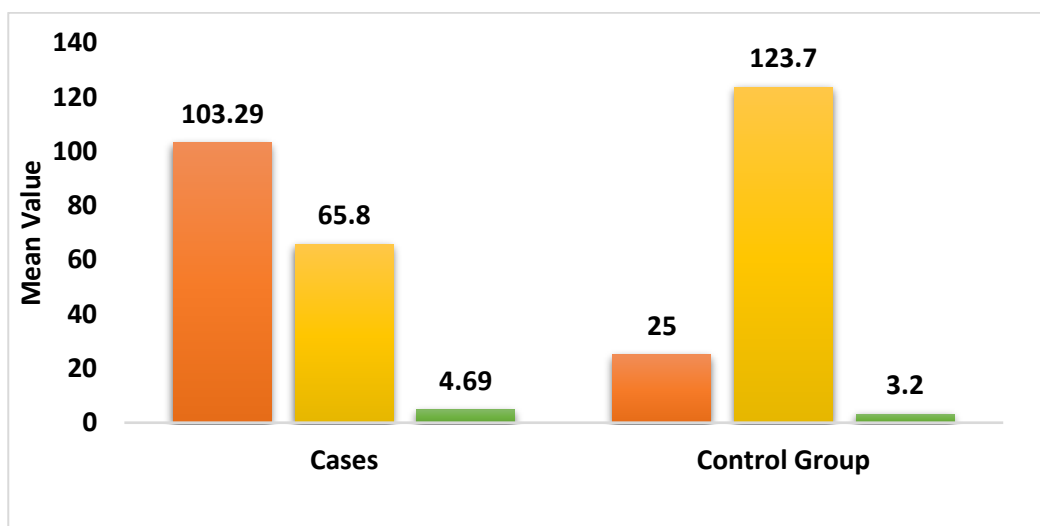
Table-2: Distribution of the subjects in study group and control group according to different ADA levels



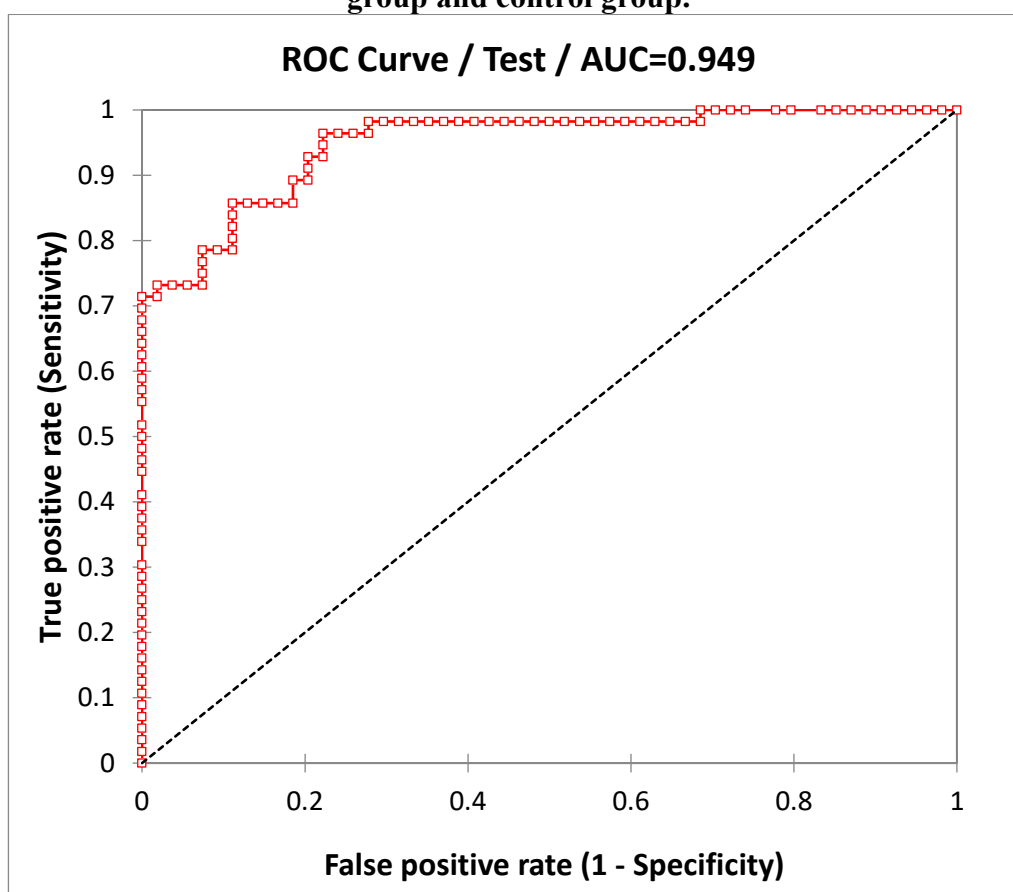
Graph Chart-2: Distribution of the subjects in study group and control group according to different ADA levels.

Parameter	Study Group (n= 56)	Control Group (n= 54)	t-value	p-value	Level of significance
	Mean ± SD				
ADA (IU/L)	103.29 ± 61.26	25.0 ± 18.2	9.0142	0.0001	HS
Glucose (mg/dl)	65.80 ± 29.27	123.7 ± 79.9	5.0815	0.0001	HS
Protein (g/dl)	4.69 ± 0.97	3.2 ± 1.0	7.9326	0.0001	HS

Table-3: Comparison of pleural fluid adenosine deaminase activity, glucose and protein levels in the study and control group.



Graph-3: Comparison of Pleural Fluid ADA activity, Glucose and Protein levels in the test group and control group.



Graph Chart-4: ROC curve.

INTERPRETATION OF ROC CURVE

The receiver operating characteristic (ROC) curve for ADA was performed for the pleural effusion patients. The area under curve (AUC) was 0.949 (95% CI 0.917-0.981). ADA level of 45.82 IU/L had a sensitivity of 85.7% and specificity of 88.9% for the diagnosis of tubercular pleural effusion.

DISCUSSION

In the developing countries like India the commonest cause of pleural effusion is TB. Untreated tubercular pleural effusion can develop into active TB. So, it is important to make rapid and accurate diagnosis for TPE and initiation of treatment. Definitive diagnosis of TB is a difficult task, as in more than 50% of the cases, pleura is the only site of infection. Pleural effusion is a building

up of fluid in the pleural space which is an indicator of an underlying disease process that may be pulmonary or non-pulmonary in origin. Based on the mechanism of fluid formation pleural effusions are transudates or exudates. Transudates are ultrafiltrate of plasma, whereas exudates are the result of inflammation of pleura or decreased lymphatic drainage. TB is the major cause of pleural effusion and such pleural effusions are usually exudates. Hence in the present study exudative pleural effusion cases are considered as TPE and cases other than exudative effusion or transudative effusion as non TPE [9].

ADA (IU/L):The study observed higher levels of ADA activity in TPE patients. According to the study done by Salam et al, mean $\pm$ SD of ADA activity in TPE cases was 55 ± 14.2 IU/L [10]. In a study by Masood et al, the mean $\pm$ SD of ADA activity in TPE cases was 72.17 ± 68.13 IU/L [11]. Suleman et al, showed the mean $\pm$ SD of ADA activity in TPE cases was 52.16 ± 2.4 IU/L [5]. Gayathri devi et al, showed the mean $\pm$ SD of ADA activity in TPE cases was 62.61 ± 24.50 IU/L [12]. Suchita D. Modi et al, showed the mean $\pm$ SD of ADA activity in TPE cases was 85.09 ± 30.20 IU/L [1]. Study done by Dave et al, mean $\pm$ SD of ADA activity in TPE cases was 54.97 ± 23.51 IU/L [13].

ADA is an enzyme catalyzing the conversion of the adenosine and deoxyadenosine to the inosine and deoxy-inosine in the purine degradation pathway. Its quantity increases in the immature and non-differentiated T-lymphocytes following mitogenic and antigenic stimulation. [14] While the increase of ADA activity in the malignant pleural effusion has been associated with the predominance of CD-8, prominent rise observed in TPE has been tried to be explained with the presence of gradually increasing CD-4 blastogenesis after the mycobacterial antigenic stimulation. [15]

PROTEIN (g/dl):

In the present study the range of protein in the TPE cases was 2.3 - 7 g / d * l and the mean $\pm$ SD was In the non-TPE cases the range of the protein was g/dl and the mean $\pm$ SD was 3.2 plus/minus 1 g/dl a study done by Gupta et al, [16] the mean $\pm$ SD of protein in the TPE cases was 4.3 ± 0.9 g/dl and in the non-TPE cases it was 4.2 ± 1.1 g/dl. Another study done by Priyadarshini K.S et al, [17] the range of protein in the TPE cases was 3.5-6.4 g/dl and the mean $\pm$ SD was 4.63 ± 0.6 g/dl.

The above studies showed that the range and mean $\pm$ SD of protein activity in the TPE was very similar with the present study.

GLUCOSE (mg/dl):

In the present study the range of glucose in the TPE cases was 10-111 mg/dl and the mean $\pm$ SD was 65.80 ± 29.27 mg/dl. In the non-TPE cases the range of protein was 20-442 mg/dl and the mean $\pm$ SD was 123.7 ± 79.9 mg/dl. In a study done by Valdes et al, [18] the range of glucose in the TPE cases was 9-116 mg/dl and the mean $\pm$ SD was 70.0 ± 12.1 mg/dl. In the non-TPE cases the range of glucose was 79-95 mg/dl and the mean $\pm$ SD was 87.7 ± 5.0 mg/dl. Another study done by Priyadarshini K.S et al, [17] the range of glucose in TPE cases was 50-100 mg/dl and the mean $\pm$ SD was 67.3 ± 14.12 mg/dl. Both the above studies showed results very similar with the present study.

CONCLUSION

Adenosine deaminase levels in non-tubercular exudative pleural effusion rarely exceeded the cut-off set for tubercular disease. We conclude that the test performance of ADA in diagnosing TPE is reasonably good, and it should be a useful tool for diagnosing TPE. Estimation of ADA activity is simple, reliable and cost-effective. The study showed that estimation of ADA activity is highly sensitive and specific method to routinely differentiate between tubercular and non-tubercular pleural effusion.

Conflict of interest: Nil

References:

1. Smith I. Mycobacterium tuberculosis pathogenesis and molecular determinants of virulence. Clin Microbiol Rev 2003; 16:463-96.
2. Modi SD, Agrawal AK, Bhake AS, Agrawal VR. Role of adenosine deaminase in pleural fluid in tubercular pleural effusion. J Datta Meghe Inst Med Sci Univ 2018;13:163-7.
3. Vorster MJ, Allwood BW, Diacon AH, Koegelenberg CF. Tuberculous pleural effusions: Advances and controversies. J Thorac Dis 2015;7:981-91.
4. Vereza Hernando HR, Masa Jimenez JF, Dominguez Juncal L, Perez Garcia-Buela J, Martin Egaña MT, Fontan Bueso J. Meaning and diagnostic value of determining the lysozyme level of pleural fluid. Chest 1987;91(3):342-5.
5. Suleman A, Kamal M, Abbasi MA, Anwar SA, Khan H. Diagnostic utility of pleural fluid adenosine deaminase level in tuberculous pleural effusion. Journal of Ayub Medical College Abbottabad. 2016 Jun 1;28(2):245-8.
6. Bielsa S, Palma R, Pardina M, Esquerda A, Light RW, Porcel JM. Comparison of polymorphonuclear and lymphocyte-rich tuberculous pleural effusions. Int J Tuberc Lung Dis. 2013 Jan 1;17(1):85-9.
7. Piras MA, Gakis C, Budroni M, Andreoni G. Adenosine deaminase activity in pleural effusions: an aid to differential diagnosis. Br Med J 1978;2(6154):1751-2.
8. Rousta F, Sokouti M, Rouy SB, Parsay S. Diagnostic Value of Pleural Fluid Adenosine Deaminase In Patients with Pleural Tuberculosis. Journal of Genetic Engineering and Biotechnology Research. 2022 Nov 24;4(3):393-8.
9. Lee SJ, Kim HS, Lee SH, Lee TW, Lee HR, Cho YJ, Jeong YY, Kim HC, Lee JD, Hwang YS. Factors influencing pleural adenosine deaminase level in patients with tuberculous pleurisy. Am J Med Sci. 2014 Nov;348(5):362-5. doi: 10.1097/MAJ.0000000000000260. PMID: 24762755.
10. Salam HM, Abdel-Hamid HM, Mostafa YM. Adenosine deaminase activity in serum and pleural fluid of patients with tuberculosis and those without tuberculosis. The Egyptian Journal of Chest Diseases and Tuberculosis. 2022 Apr 1;71(2):228.
11. Masood KI, Shahid S, Jabeen K, Farooqi J, Kerawala SR, Irfan M. Diagnostic accuracy of different cut-off values of adenosine deaminase levels in tuberculous pleural effusion. Journal of Pakistan Medical Association. 2023;73.
12. HJ, G.D., 2019. Utility of pleural fluid adenosine deaminase in diagnosing tubercular pleural effusion: a prospective observational study in a tertiary care hospital
13. Dave V, Patel SJ, Shah A. Pleural Fluid Adenosine Deaminase for Diagnosis of Tuberculous Pleural Effusion. Int J Res Med. 2018;7(2):41-9
14. Light RW. Pleural diseases. Current opinion in pulmonary medicine. 2003 Jul 1;9(4):251-3.
15. Baganha MF, Pêgo A, Lima MA, Gaspar EV, Cordeiro AR. Serum and pleural adenosine deaminase: correlation with lymphocytic populations. Chest. 1990 Mar 1;97(3):605-10.
16. Ahmed S, Fatema R, Saleh AA, Sattar M, Miah MR. Diagnostic significance of pleural fluid adenosine Deaminase activity in tuberculous pleurisy. Ibrahim Medical College journal. 2011;5(1):1-5.
17. Darshini KP, Reena R, Nussrath A. Diagnostic utility of adenosine deaminase activity in tubercular pleural effusion. Journal of Evolution of Medical and Dental Sciences. 2013 Jun 17;2(24):4358-63.
18. Valdés L, Álvarez D, San José E, Penela P, Valle JM, García-Pazos JM, Suárez J, Pose A. Tuberculous pleurisy: a study of 254 patients. Archives of internal medicine. 1998 Oct 12;158(18):2017-21.