International Journal of Research in Medical Sciences
Quadry SA et al. Int J Res Med Sci. 2024 Sep;12(9):3176-3181
www.msjonline.org
pISSN 2320-6071 | eISSN 2320-6012
DOI: https://dx.doi.org/10.18203/2320-6012.ijrms20242591

Original Research Article

Evaluation of usefulness of pleural fluid/serum alkaline phosphatase in the diagnosis of tubercular pleural effusion

Shafiul Azam Quadry[1]*, Abir Hasan Dip[2], Uma Dhar[3], M. Shariful Alam[4], M. Tanimul Haque Rijvy[5], M. Zakir Hossain Sarkar[6], M. Sayedul Islam[6], Abdullah Al Masud[7], M. Shohidul Islam[8], M. Rowshan Arif[9]

[1]Medical Officer, Upazila Health Complex, Ashuganj, Brahmanbaria, Bangladesh
[2]Department of Cardiology, Shaheed Ziaur Rahman Medical College Hospital, Bogura, Bangladesh
[3]Department of Medicine, OSD, DGHS, Mohakhali, Dhaka, Bangladesh
[4]Medical Officer, OSD, DGHS, Mohakhali, Dhaka, Bangladesh
[5]Assistant Registrar, Jashore Medical College Hospital, Jashore, Bangladesh
[6]Department of Respiratory Medicine, National Institute of Diseases of the Chest and Hospital, Mohakhali, Dhaka, Bangladesh
[7]Medical Officer, National Institute of Diseases of the Chest and Hospital, Mohakhali, Dhaka, Bangladesh
[8]Junior Consultant, Chest Diseases Hospital, Bogura, Bangladesh
[9]Residencial Medical Officer, National Institute of Diseases of the Chest and Hospital, Mohakhali, Dhaka, Bangladesh

Received: 30 June 2024
Accepted: 12 August 2024

***Correspondence:**
Dr. Shafiul Azam Quadry,
E-mail: shafredefined@gmail.com

ABSTRACT

Background: Tuberculosis (TB) remains a common cause of pleural effusions, diagnosed by detecting Mycobacterium tuberculosis in pleural fluid or biopsy specimens through microscopy, culture, or histological demonstration of caseating granulomas and acid-fast bacilli (AFB). In high-burden settings, the diagnosis is frequently inferred. Objective was to assess the usefulness of pleural fluid/serum alkaline phosphatase in the diagnosis of tubercular pleural effusion.

Methods: This cross-sectional observational study took place at the department of respiratory medicine, NIDCH, Dhaka, from December 2018 to December 2019. Seventy new cases of pleural effusion meeting specific criteria were enrolled with informed consent obtained. Diagnostic assessments included Gene Xpert, cytology, culture and sensitivity testing, biochemical analyses, and alkaline phosphatase testing on pleural fluid and blood samples. Closed needle pleural biopsies using an Abrams needle were performed for histopathological examination. Data analysis was carried out using SPSS version.

Results: In this study of 70 patients, mean age 48.67±17.99 years (range: 17-83 years), with 42.9% aged 51-70 years, males predominated (6:1). Histopathology showed TB in 45.7%. TB patients had lower neutrophil and platelet counts, lower serum alkaline phosphatase, higher ESR, and higher pleural fluid alkaline phosphatase and P/S ALP ratio. The P/S ALP ratio had an AUC of 0.881 (95% CI 0.798-0.964), with a cut-off of 0.49, sensitivity/NPV of 93.8%, and specificity/PPV of 78.9% for diagnosing TB pleural effusion.

Conclusions: The pleural fluid/serum alkaline phosphatase ratio is a valuable diagnostic tool for tuberculous pleural effusion. Further validation through a multicenter randomized trial with a larger sample size is recommended.

Keywords: Alkaline phosphatase, Tuberculous pleural effusion

INTRODUCTION

Tuberculosis, a specific infection caused by *Mycobacterium tuberculosis*, is the leading cause of morbidity and mortality, mostly confined to developing and under developed countries.[1] Bangladesh is one of the most endemic regions in the world for *Mycobacterium tuberculosis*. Worldwide, TB is the 13th leading cause of death and the second leading infectious killer after COVID-19. In 2020 an estimated 10 million people fell ill with tuberculosis worldwide, 5.6 million men, 3.3 million women and 1.1 million children, a total of 1.5 million people died from tuberculosis in the same year. The incidence rate of TB in Bangladesh is 221 per 100000 populations per year, making it one of the 30 high burden countries for TB.[2] Tuberculosis spread by airborne droplets or droplet nuclei from a person with pulmonary TB. Transmission often occurs indoors, where droplets and droplet nuclei can stay in the air for a long time. Although there are effective therapies for tuberculosis, epidemiological data show a rise in the incidence, especially since AIDS incidence rose steeply.[3]

Tuberculous pleural disease now more commonly represents reactivation rather than primary infection and is present in approximately 7% cases of active pulmonary TB[1]. It is also the second most common cause of extra-pulmonary tuberculosis after lymphatic involvement, where tuberculous pleural effusion makes up 15-20% of all extra-pulmonary TB.[4] Among the exudative pleural effusions, tuberculosis makes up 54.57%, followed by malignancy (28.17%), empyema (10.56%) and para-pneumonic effusion (5.28%).[5] But diagnosing the etiology of pleural effusions with certainty is still a challenging task for physicians.

It is crucially important to differentiate tuberculous pleural effusion and para-pneumonic effusion from malignant pleural effusion, as misdiagnosis and delayed treatment can result in significant mortality and morbidity. Like other forms of extra-pulmonary tuberculosis, diagnosis of tuberculous pleural effusion is usually done by an invasive procedure (pleural biopsy), which requires an expert hand and is often not possible outside a tertiary care hospital. Moreover, standard criterion [i.e. positive Ziehl-Neelsen (ZN) stain or Lowenstein-Jensen (LJ) culture of pleural fluid or tissue specimen] is seldom met due to the pauci-bacillary nature of this condition. A recent study found that only 31% of the patients with tuberculous pleural effusion have a positive microbiological test result. An increased pleural fluid adenosine deaminase (ADA) level is thus frequently used to diagnose tuberculous pleural effusion.[6-8]

Various other parameters have also been used to differentiate tuberculous from non- tuberculous pleural effusion. These includes pleural fluid lysozyme, PF-gamma interferon, PF-alpha antitrypsin, PF-protease inhibitors, PF-PCR, PF-CA-125, PF-interleukins, all of which have been reported to be elevated in tuberculous pleural effusion.[3,9,10] However high cost of most techniques and lack of availability of sophisticated tools and reagents have precluded their implementation on routine basis, especially in low-income countries. None of them has been yet validated as a standard tool for the diagnosis of tuberculous pleural effusion. Alkaline phosphatase is an endogenous plasma membrane derived enzyme of uncertain physiological function that hydrolyses synthetic phosphate esters at pH 9. It is present in serum in six different forms in the body.

These different forms are due to the difference in the carbohydrate content (sialic acid residues). These activities arise from liver, bone (osteoblast), mucosa of small intestine, proximal convoluted tubules of kidney, placenta etc. The exact metabolic function of the enzyme is not understood but it appears that alkaline phosphatase is associated with lipid transport mechanism in the intestine and with calcification process in bones. Alkaline phosphatase is one of the biochemical markers found in pleural fluid. Previous studies have successfully utilized pleural fluid alkaline phosphatase to differentiate exudative from transudative effusion.[3,11-13]

Objective

The aim of the study was to see the role of pleural fluid/serum alkaline phosphatase in differentiating tuberculous from non-tuberculous pleural effusion.

METHODS

This was a cross-sectional observational study conducted in the department of respiratory medicine, National Institute of Diseases of the Chest and Hospital (NIDCH), Mohakhali, Dhaka from December 2018 to December 2019. All new patients presenting with unilateral exudative pleural effusion aged between 15 to 85 years, were admitted to the inpatient Department of Respiratory Medicine of NIDCH, Mohakhali, Dhaka. were included in the study. However, patients with bone and joint disease, pregnant women, patients with jaundice/ hepatitis/chronic liver disease, renal failure/ CKD, known cases of transudative or bilateral pleural effusion, and obvious hemothorax secondary to trauma were excluded from the study. Finally, 70 samples were taken. Chest x-ray, P/A view, serum total protein, serum alkaline phosphatase, pleural fluid for alkaline phosphate, protein, glucose, ADA, pleural biopsy for histopathology was sent for study. A pre-tested data collection sheet. Statistical analysis was conducted using the Statistical Package for the Social Sciences (SPSS) version 23.

RESULTS

This cross-sectional observational study was conducted to assess the usefulness of pleural fluid/serum alkaline phosphatase in diagnosis of tuberculous pleural effusion. The results are as follows:

Mean age of the patients was 48.67±17.99 years ranged from 17 to 83 years. Maximum patients were in 51-70 years' age group (42.9%). Males were predominant than female (6:1). Among the study subjects 60.0% were smoker.

Table 1: Demographic profile of the study subject (n=70).

	Frequency	Percentage
Age (years)		
<30	15	21.4
31-50	18	25.7
51-70	30	42.9
>70	7	10.0
Mean±SD (min-max)	48.67±17.99 (17-83)	
Gender		
Male	60	85.7
Female	10	14.3
Smoker	42	60.0

As per histopathology findings of pleural biopsy, 45.7% of the study subjects had TB.

Table 2: Distribution of patients according to the diagnosis (n=70).

	Frequency	Percentage
TB	32	45.7
Non-TB	38	54.3

Neutrophil, platelet count, and serum alkaline phosphatase was found significantly lower in TB patient than non-TB. Lymphocyte and ESR was found significantly higher in TB patients than non-TB patients (Table 3).

WBC, neutrophils and histiocytes of pleural fluid was found significantly lower in TB patients than non-TB whereas lymphocytes, protein, alkaline phosphatase and ADA of pleural fluid were found significantly higher in TB patients than non-TB. Pleural and serum alkaline phosphatase ratio was found significantly higher in TB patients than non-TB (Table 4).

Table 3: Comparison of lab investigation of serum between TB and non-TB patients (n=70).

	TB (n=32) mean±SD	Non-TB (n=38) mean±SD	P value
Total count of WBC (106/l)	9681±1153	10150±1655	0.182
Neutrophil (%)	49.38±7.68	54.53±9.26	0.015
Lymphocyte (%)	46.19±6.51	40.13±8.09	0.001
Monocytes (%)	2.25±1.19	2.76±1.75	0.164
Eosinophils (%)	1.47±0.72	2.16±1.94	0.061
Basophils (%)	0.34±0.55	0.42±0.76	0.632
Platelet count (/mm^3)	200106±4253	229857±56061	0.011
ESR (mm in 1st hour)	88.91±7.63	62.42±10.13	<0.001
Serum total protein (gm/dl)	5.52±1.50	5.23±1.62	0.440
Serum alkaline phosphatase (IU/l)	112.53±17.80	124.00±27.13	0.044

Unpaired t test was done.

Table 4: Comparison of lab investigation of pleural fluid between TB and non-TB patients (n=70).

	TB (n=32)	Non-TB (n=38)	P value
	Mean±SD	**Mean±SD**	
WBC (/mm^3)	1067.19±225.29	1399.21±688.45	0.011
Neutrophils	14.28±7.04	19.00±11.15	0.042
Lymphocytes	64.84±11.22	49.97±15.44	<0.001
Eosinophils	0.78±1.52	0.84±1.03	0.843
Histiocytes	21.03±8.88	30.45±9.93	<0.001
Protein (gm/dl)	5.33±0.29	3.39±0.27	<0.001
Alkaline Phosphatase (U/l)	79.91±16.45	51.29±17.77	<0.001
ADA (U/l)	71.06±10.21	30.21±11.93	<0.001
PIS alkaline phosphatase ratio	0.70±0.13	0.43±0.17	<0.001

Unpaired t test was done.

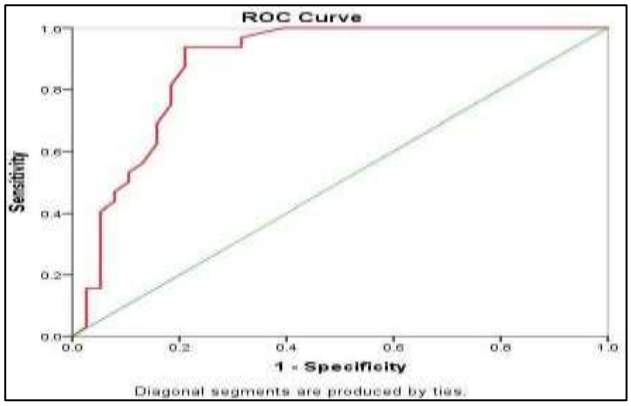

Figure 1: Receiver operating characteristic (ROC) curve of P/S alkaline phosphatase ratio for the differentiation of TB from non-TB.

The area under the curve was 0.881 (95% CI 0.798-0.964), Youden index for the optimal cut-off point (P/S alkaline phosphatase ratio >0.49), was 0.727.

Table 5: Diagnostic efficacy parameters for the use of PIS alkaline phosphatase ratio in the differentiation of TB from non-TB for different cut-off point (n=70).

PIS ALP ratio	Sensitivity	Specificity	Youden index
0.40	1.000	0.605	0.605
0.43	0.938	0.684	0.622
0.44	0.938	0.737	0.674
0.45	0.938	0.763	0.701
0.49	0.938	0.789	0.727
0.51	0.929	0.789	0.720
0.54	0.906	0.789	0.696
0.57	0.875	0.789	0.664
0.59	0.813	0.816	0.628
0.63	0.750	0.816	0.566

The optimal cut-off point of P/S alkaline phosphatase ratio was 0.49 in differentiating TB from non-TB according to Youden index (0.727).

Table 6: Association of P/S APL ratio with histopathological findings (N=70)

P/S APL ratio	Histopathology		P value
	TB	Non-TB	
0.49	30 (93.8)	8 (21.1)	<0.001
<0.49	2 (6.3)	30 (78.9)	

Chi-square test was done

Among 32 histopathology diagnosed TB patients 30 had P/S APL ratio 0.49 and 2 had P/S APL ratio<0.49, they were true positive and false negative respectively. Among 38 histopathology diagnosed non-TB cases 30 had P/S APL ratio <0.49 and 8 had P/S APL ratio 0.49, they were true negative and false positive respectively.

Table 7 shows diagnostic efficacy parameters for the use of P/S APL ratio in differentiating TB from non-TB pleural effusions at a cut-off point 0.49. Sensitivity, specificity, PPV and NPV of P/S APL ratio were 0.938, 0.789, 0.789 and 0.938 respectively in differentiating TB from non-TB pleural effusions at a cut-off point 0.49.

Table 7: Diagnostic efficacy parameters for the use of P/S alkaline phosphatase ratio in diagnosing TB pleural effusion at a cut-off point 0.49 N=70).

		95% CI	
		Min	Max
Sensitivity	0.938	0.815	0.989
Specificity	0.789	0.686	0.833
Positive predictive value	0.789	0.686	0.833
Negative predictive value	0.938	0.815	0.989
Accuracy	0.857	0.745	0.904

DISCUSSION

This cross-section observational study was conducted in the department of respiratory medicine, NIDCH, Mohakhali, Dhaka over a period of one year (From December 2018 to December 2019). A total of 70 patients were considered for the study as per inclusion and exclusion criteria. Mean age of the patients was 48.67±17.99 years which ranged from 17 to 83 years. Maximum patients were in 51-70 years' age group (42.9%). Among the study subjects 60.0% were smoker. As per histopathology test 45.7% of the study subjects had TB. Neutrophil, platelet count and serum alkaline phosphatase was found significantly lower in TB patient than non-TB. Lymphoctye count and ESR was found significantly higher in TB patients than non-TB. Mean serum alkaline phosphatase was 112.53±17.80 IU/l in TB patients and 124.00±27.13 IU/l in non-TB patients, difference was statistically significant. But one the previous study found no significant difference in serum alkaline phosphatase (140.36±43.21 IU/l versus 140.60±32.80 IU/l; p=0.981) between tuberculous and non-tuberculous pleural effusion.[3]

While studying pleural fluid, WBC, neutrophils and histiocytes of pleural fluid was found significantly lower in TB patients than non-TB cases whereas lymphocytes, protein, alkaline phosphatase and ADA of pleural fluid were found significantly higher in TB patients than non-TB. Pleural fluid/serum alkaline phosphatase was found significantly higher in TB patients than non-TB. Mean pleural alkaline phosphatase was 79.91±16.45 IU/l in TB patients and 51.29±17.77 IUIL in non-TB patients. Mean ADA was 71.06±10.21 in TB patients and 30.21±11.93 in non-TB patients. Mean pleural fluid/serum alkaline phosphatase ratio was 0.70±0.13 in TB patients and 0.43±0.17 in non-TB patients. In one of the previous study, patients with tuberculous pleural effusion also had a significantly (p<0.0001;) higher mean pleural fluid/serum alkaline

phosphates ratio than with non-tuberculous pleural effusion.[3] The optimum cut-off level was determined by selecting points of test values that provided the greatest Youden Index (sensitivity + specificity- 1). Optimal cut-off point of pleural fluid/serum alkaline phosphatase ratio was 0.49, in differentiating TB from non-TB in this study. In this study, among 32 histopathology diagnosed TB patients, 30 had pleural fluid/serum alkaline phosphatase ratio 0.49 and 2 had pleural fluid/serum Alkaline phosphatase ratio <0.49, they were true positive and false negative respectively. Among 38 histopathology diagnosed non-TB cases 30 had pleural fluid/serum Alkaline phosphatase ratio <0.49 and 8 had pleural fluid/serum alkaline phosphatase ratio 0.49, they were true negative and false positive respectively.

In one of the previous studies, pleural fluid/serum alkaline phosphatase ratio greater than 0.51 was observed in 27 out of 30 cases of tuberculous pleural effusion and four out of 30 cases of non-tuberculous pleural effusion.[3] The value of pleural fluid alkaline phosphatase and pleural fluid/serum alkaline phosphatase ratio was higher in patients with tuberculous pleural effusion as well as it was found that the sensitivity and specificity of pleural fluid/serum alkaline phosphatase ratio of 93.8% and 78.9% respectively for diagnosing tuberculous pleural effusion which was provided by ROC curve analysis. A similar finding was observed in a previous study which revealed a sensitivity of 90.0% and specificity 86.6%.[3] Although prior studies have attempted to use alkaline phosphatase to distinguish tuberculous from other kinds of pleural effusion, none of them have been successful in clearly distinguishing tuberculous from nontuberculous pleural effusion.[13-15] Pleural fluid alkaline phosphatase was considerably higher in malignant pleural effusions compared to tuberculous, nontuberculous, and effusions owing to other causes in one of the studies.[15] Furthermore, while comparing tuberculous pleural effusion to neoplastic effusion, other exudates, and transudates, one of the studies found that pleural fluid alkaline phosphatase and pleural fluid/serum alkaline phosphatase ratio were considerably higher in tuberculous pleural effusion than in neoplastic effusion, other exudates, and transudates.[14] However, in distinguishing exudates from transudates, a previously conducted study found that alkaline phosphatase did not distinguish tuberculous from other sources of effusion, such as malignancy, parapneumonic effusion, and nonspecific effusion.[13] In light of the aforementioned debate, the use of pleural fluid/serum alkaline phosphatase ratio was investigated in distinguishing tuberculous from nontuberculous pleural effusion, and discovered that pleural fluid/serum alkaline phosphatase ratio is a valuable biochemical marker for the diagnosis of tuberculous pleural effusion.

This study's small sample size and single-center design limit the generalizability of its findings. The cross-sectional nature restricts causal inferences, and the selection criteria may have introduced bias. Reliance on specific diagnostic tests could affect comparability with other studies. Additionally, the lack of long-term follow-up data prevents assessment of patient outcomes and treatment effectiveness, and potential confounding variables may not have been fully accounted for.

CONCLUSION

In this study, pleural fluid/serum alkaline phosphatase, with a cut-off of 0.49, had a sensitivity of 93.8%, specificity of 78.9%, positive predictive value of 78.9%, and negative predictive value of 93.8%, with an accuracy of 85.7% in successfully detecting tuberculous pleural effusion. In light of the findings of this study, pleural fluid/serum alkaline phosphatase may be a new and valuable laboratory indicator for early screening and diagnosis of tuberculous pleural effusion.

Funding: No funding sources
Conflict of interest: None declared
Ethical approval: The study was approved by the Institutional Ethics Committee

REFERENCES

1. Prasad BS. Tuberculous pleural effusion diagnosis by detection of alkaline phosphatase in serum and pleural fluid. Sch J Appl Med Sci. 2016;4(12C):4414-8.
2. World Health Organization. Global tuberculosis report. 2018. Available from: http://www.who.int/iris/handle/10665/274453. Accessed on 5 July 2019.
3. Jadhav AA, Bardapurkar JS, Jain A. Alkaline phosphatase: Distinguishing between tuberculous and nontuberculous pleural effusion. Lung India. 2009;26(3):77.
4. Quddus MA, Uddin MJ, Bhuiyan MM. Evaluation of extrapulmonary tuberculosis in Bangladeshi patients. Mymensingh Med J. 2014;23(4):758-63.
5. Maji A, Maikap MK, Jash D, Saha K, Kundu A, Saha D, et al. Role of common investigations in etiological evaluation of exudative pleural effusions. J Clin Diagn Res. 2013;7(10):2223.
6. Sahn SA, Heffner JE. Pleural fluid analysis. In: Light RW, Lee YCG, eds. Textbook of pleural diseases. 2nd edn. London: Hodder Arnold; 2008:209-226.
7. Lin MT, Wang JY, Yu CJ, Lee LN, Yang PC. TAMI Group. *Mycobacterium tuberculosis* and polymorphonuclear pleural effusion: incidence and clinical pointers. Respir Med. 2009;103(6):820-6.
8. Kim CH, Lee SY, Lee YD, Yoo SS, Lee SY, Cha SI, et al. Atypical pleural fluid profiles in tuberculous pleural effusion: sequential changes compared with parapneumonic and malignant pleural effusions. Intern Med. 2016;55(13):1713-9.
9. Ocana I, Martinez-Vazquez JM, Segura RM, Fernandez-De-Sevilla T, Capdevila JA. Adenosine deaminase in pleural fluids: test for diagnosis of

tuberculous pleural effusion. Chest. 1983;84(1):51-3.
10. Villegas MV, Labrada LA, Saravia NG. Evaluation of polymerase chain reaction, adenosine deaminase, and interferon-y in pleural fluid for the differential diagnosis of pleural tuberculosis. Chest. 2000;118(5):1355-64.
11. Gazquez I, Porcel JM, Vives M, de Vera MV, Rudio M. Pleural alkaline phosphatase in separation of transudative and exudative pleural effusions. Chest. 1997;112(2):569-70.
12. Tahaoglu K, Kizkin Ö, El R. Alkaline phosphatase: distinguishing between pleural exudates and transudates. Chest. 1994;105(6):1912-3.
13. Lone MA, Wahid A, Saleem SM, Koul P, Dhobi GN, Shahnawaz A. Alkaline phosphatase in pleural effusions. Indian J Chest Dis Allied Sci. 2003;45(3):161-4.
14. Metinta M, Alata S, Alata F, Colak B, Ozdemir N, Erginel S. Comparative analysis of biochemical parameters for differentiation of pleural exudates from transudates: Light's criteria, cholesterol, bilirubin, albumin gradient, alkaline phosphatase, creatine kinase, and uric acid. Clin Chim Acta. 1997;264(2):149-62.
15. Carrion F, Perpifia M. Use of pleural alkaline phosphatase content to diagnose tuberculous effusions. Chest. 1993;104(2):646-7.

Cite this article as: Quadry SA, Dip AH, Dhar U, Alam MS, Rijvy MTH, Sarkar MZH, et al. Evaluation of usefulness of pleural fluid/serum alkaline phosphatase in the diagnosis of tubercular pleural effusion. Int J Res Med Sci 2024;12:3176-81.