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ORIGINAL ARTICLE

DIAGNOSTIC ROLE OF SERUM ADENOSINE DEAMINASE IN SMEAR-NEGATIVE PULMONARY TUBERCULOSIS

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Abstract:

Background: *Diagnosis of tuberculosis is not always easy, particularly if it is a case of sputum smear-negative pulmonary tuberculosis (SNPTB). Patients with respiratory symptoms resembling SNPTB are difficult to differentiate based on clinical features, chest X-ray, and Xpert MTB/RIF negativity. So, additional diagnostic test with high sensitivity and specificity is needed to increase the yield of the ongoing diagnostic strategy for SNPTB. Adenosine deaminase (ADA) is now being widely used for the diagnosis of TB particularly in effusion fluids due to its simplicity, low cost, and quick available results, it is not always possible to access effusion fluids and therefore, it would be helpful to take advantage of serum levels. Therefore, the purpose of the study was to assess the role of serum ADA in the diagnosis of SNPTB.* ***Methods:*** *This cross-sectional analytical study was conducted in Dhaka Medical College & Hospital, Dhaka from March 2019 to September 2021. A total of 140 patients were included in this study and divided into two groups according to selection criteria: Group I (SNPTB, n=62), and Group II (non-TB pulmonary diseases, n=78). ADA estimation was carried out using the sensitive colorimetric method described by Guisti and Galanti with a BIOSIC kit. After the collection of all the required data, analysis was done by SPSS 24.0.* ***Results:*** *The mean age of the study patients was 48.02 ± 9.60 years (23-73 years) with male predominance in both Group I and Group II (71 % and 60.3%, respectively, p>0.05). Non-TB pulmonary cases were significantly older than SNPTBpatients (52 ± 8.56 vs 43.02 ± 8.49 years, p<0.001). SNPTB patients had a significantly higher frequency of cough, fever, and weight loss compared to non-TB pulmonary cases (p<0.05). In contrast, chest pain and shortness of breath were more frequent in Group II than in Group I (p<0.05). Serum ADA was significantly higher among SNPTB patients compared to non-TB pulmonary cases (48.16 ± 12.13 vs 18.64 ± 7.85 IU/L, p<0.001). ROC analysis of serum ADA in the diagnosis of patients with SNPTB found an AUC of 0.9850 (95% CI, 0.969-1.00) which was statistically significant (p<0.001). A cut-off value of serum ADAe"33 IU/L showed sensitivity, specificity, NPV, PPV, and accuracy of 93.55%, 94.87%, 94.87%, 93.55%, and 94.29%, respectively to correctly diagnose SNPTB cases.* ***Conclusion.*** *This study finding stated that serum ADA may be a useful marker to distinguish SNPTB from non-TB respiratory diseases. However, further study with a more generalized study population is recommended.*

Keywords: *ADA, Smear-negative Pulmonary TB*

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Introduction:

Tuberculosis (TB), which is one of the important re-emerging infectious conditions is caused by Mycobacterium *tuberculosis*. Pathologically, it is characterized by the formation of granulomas.[1]About one-third of the world's population is infected with TB latently and annual new cases of TB worldwide count approximately 9 million.[2] It has become a global health concern for both developing and developed countries. TB now ranks as a leading infectious disease killer globally alongside Human Immuno-deficiency Virus (HIV) despite being preventable and curable.[3] According to WHO Global TB Report 2016, Bangladesh is regarded as one of the world's 30 high TB burden countries with an annual occurrence of 362,000 new TB cases. Due to tuberculosis, about 73,000 people die annually (World Health Organization, 2017).[4]

Tuberculosis can present as pulmonary tuberculosis (PTB) or extrapulmonary tuberculosis (EPTB). Pulmonary TB is the most common form of tuberculosis among them.[5] The main symptoms of PTB are chronic cough, low-grade fever, evening rise of temperature, hemoptysis,dyspnea, chest pain, weight loss, and unresolved pneumonia.[6] In clinical practice, rapiddiagnosis of TB can be difficult, and early pulmonary TB detection has become challenging for clinicians. Prompt detection of active pulmonary tuberculosis has become a priority for tuberculosis control, both for patient treatment and public health intervention to prevent further transmission in the community. Chest X-ray is useful but is non-specific fordiagnosis of pulmonary TB. Moreover, TB can cause symptoms and radiologic results that are indistinguishable from those of community-acquired pneumonia.[7]

There are other different diagnostic modalities but they have some drawbacks. Cultureis the golden standard for TB diagnosis, but it may take 8 weeks. The polymerase chain reaction (PCR) test for TB diagnosis is expensive and requires skilled personnel and a lot of equipment. Finding acid-fast bacilli is the quick screening method fordiagnosis of pulmonary TB; nevertheless, the sensitivity is low.[8] Despite the best of efforts, a clinician oftenhas to face difficulties in smear-negative patients, and sometimes, it becomes very difficult to diagnose this entity. Co-morbidities like diabetes mellitus, HIV, and other immune-compromised conditions further complicate the picture leading to atypical clinical and radiological presentations.[9,10] This delay in diagnosis and subsequent treatment leads to increased transmission of TB and chances of drug resistance. Hence, in recent years, there has been a great demand to find a rapid diagnostic method for the same.[11]

Adenosine deaminase (ADA) is one such biomarker that is nowadays being studied as a diagnostic tool in the diagnosis of tuberculosis due to its simplicity, low cost, and quickly available results.[12] Adenosine deaminase (ADA) activity measurement is a biomedical method. ADA is an enzyme that contributes to purine metabolism. This enzyme helps in catalyzing the hydrolytic deamination of adenosine to inosine, and deoxyadenosine to deoxy inosine and plays an important physiological role in the regulation of the effects of these metabolites on immunological, neurological, and vascular processes. ADA is important for the proliferation and differentiation of lymphoid cells, especially T cells, and helps in the maturation of monocytes to macrophages. ADA is an index for cellular immunity and previous studies have proved its value in the diagnosis of TB, even for assessing TB effusions.[13] Activity of this enzyme increases in TB patients. In several studies, ADA levels in sputum and serum were used to diagnose tuberculosis and followed during treatment. However, some prior studies employed effusion fluids, and a very small number of studies used patients' serum. It is not always possible to access effusion liquids in patients with pulmonary and extra-pulmonary TB; therefore, it would be helpful to take advantage of serum levels.[14]This study was designed to look at the diagnostic usefulness of serum ADA in smear-negative pulmonary TB. The findings of this study will serve to provide a clear image of ADA for TB diagnosis, allowing future TB strategies to be conducted more effectively.

Methods:

This cross-sectionalanalytical study was conducted at the Department of Medicine, Dhaka Medical College & Hospital, Dhaka between March, 2019 to September, 2021. The study protocol was approved by the Ethical Review Committee (ERC) of Dhaka Medical College and Hospital. A total of 140newly diagnosed pulmonary cases admitted within the study period fulfilling the inclusion and exclusion criteria were included in this study byconvenient purposive sampling. The study subjects were divided into two groups according to selection criteria: Group I (Smear negative Pulmonary TB, n=62), and Group II (non-TB pulmonary cases viz. pneumonia, COPD, bronchiectasis, lung malignancy, n=78). The inclusion criteria for Group I(smear-negative pulmonary TB cases) were age: >18 years, at least two sputum specimens negative for Acid Fast Bacilli (AFB) but Xpert MTB/RIF positive, radiological abnormalities consistent with pulmonary TB - any cavitary lesion, consolidation involving mostly in the upper lobe, diffuse patchy opacity/consolidation involving lobe/whole lungand clinical symptoms of pulmonary TB - (any two) - cough for 3 weeks or more, hemoptysis, fever,

loss of appetite, weight loss. night sweats. The inclusion criteria for Group II(non-Tb pulmonary cases: pneumonia, COPD, bronchiectasis, lung malignancy) were: Age: > 18 years, for pneumonia – clinical symptoms of pneumonia (any two – cough with/ without sputum production, fever, dyspnea, pleuritic chest pain) and radiological evidence of consolidation with/without sputum smear positive for bacteria or culture positive; for COPD – clinical symptoms (dyspnea, chronic cough or sputum production) and radiological evidence suggestive of COPD with spirometric confirmation of post-bronchodilator FEV1/ FVC < 0.7; for bronchiectasis - chronic cough with tenacious sputum production and radiologically by the presence of bronchial airway dilation on CT chest; for lung malignancy – clinical symptoms (cough, hemoptysis, shortness of breath, chest pain, weight loss, fever) and radiological abnormalities consistent with lung malignancy (obvious mass, widening of the mediastinum, atelectasis (lung collapse), consolidation, pleural effusion) with histological confirmation by biopsy and sputum specimens negative for Acid Fast Bacilli (AFB) / Xpert MTB/RIF negative for all non-TB pulmonary cases. Patients with sputum smear-positive pulmonary TB, history of previous pulmonary/extra-pulmonary TB or ongoing anti-TB treatment, having secondary immunodeficiency states: HIV, organ transplantation, treatment with long-term corticosteroids, any malignancy in the body other than lung malignancy, presence of hepatic or renal impairment, pregnant and lactating women, having concomitant lymphoproliferative disease were excluded from the study. In those patients having a productive cough, sputum for AFB was sent to DOT corner in DMCH which was done by Light Emitting Diode (LED) fluorescence microscopy (FM). Sputum for acid-fast bacilli was performed in all patients on two sputum specimens as follows - one on-spot specimen which was collected on the spot when a patient was sent to the DOTS. Another was the early morning specimen where patients were given a sputum container to collect the second specimen on the following morning. Early morning sputum was also collected for Xpert MTB/ RIF examination which was done in the college building of DMCH. Sputum was also sent for gram stain, and culture in the Microbiology Department of DMCH. Chest-Xray P/A view was done in the Radiology Department of DMCH.Other necessary investigations - CBC with ESR, MT, Spirometry, CT scan of the chest, and CT-guided FNAC/Biopsy where appropriate were carried out.Those subjects whose at least two sputum specimens were negative for Acid Fast Bacilli (AFB) but Xpert MTB/RIF positive were considered as smear-negative pulmonary TB cases. And for those whose Xpert MTB/RIF was negative, they were considered as non-TB pulmonary cases. After the final enrollment, blood samples for serum ADA were sent to the BSMMU Microbiology Department written in routine biochemistry form. Serum ADA was analyzed using a sensitive colorimetric method described by Guisti and Galanti with a BIOSIC kit.All the final data were collected in the semi-structured and pretested case record form. During data collection, the highest standard ethical measures were ensured and maintained throughout the study. These data were analyzed statistically by the standard procedure to arrive at a definite conclusion about the research question.

Results:

Out of 140 patients, 62 patients had smear-negative pulmonary TB (SNPTB), and rest 78 patients were suffering from non-TB pulmonary diseases, of whom, 35 patients had pneumonia, 25 had COPD, 10 had lung carcinoma and the remaining 8 patients had bronchiectasis. SNPTB patients were assigned as Group I and non-TB pulmonary cases were assigned as Group II.

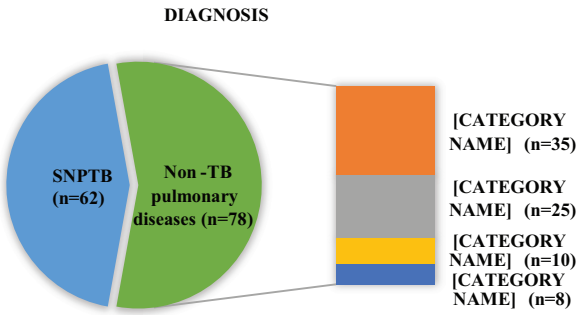

Fig. -1: *Distribution of study population according to diagnosis (n=140)*

Table I shows the baseline characteristics of the study population. Male subjects were predominant in both Group I and Group II which were 71 % and 60.3%, respectively. The mean age of the study population was 48.02±9.60 years. Patients in Group II (non-TB pulmonary cases) were significantly (p<0.00) older than Group I (SNPTB cases) with a mean age of 52±8.56 vs 43.02±8.49 years respectively.The majority patients in Group I were garment workers (38.7%) which was found statistically significant (p<0.05), while the majority patients in Group II were service holders (24.4%).Most of the study subjects in Group I and Group II were from middle-income status - 51.60% and 50% respectively, though there were no significant differences (p value=0.928) between the two groups regarding socio-economic status.

Table I

Baseline characteristics of the study population (n = 140)

Baselinecharacteristics	Group I (*n*=62)	Group II (*n*=78)	p-value
Gender			
Female	18(29 %)	30(39.7 %)	
Male	44(71%)	48(60.3%)	#[a]p>0.05
Mean Age (years)	43.02 ± 8.49	52 ± 8.56	#[b]p<0.001*
Range	41-50	51-60	
Occupation			
Housewife	7(11.3%)	18(23%)	#[a]0.7
Service Holder	11(17.7%)	19(24.4%)	#[a]0.3
Garment worker	24(38.7%)	15(19.2%)	#[a]<0.011*
Businessman	9(14.5%)	11(14.1%)	#[a]0.94
Cultivators	6(9.7%)	10(12.8%)	#[a]0.56
Unemployed	3(4.8%)	4(5.1%)	#[a]0.94
Student	2(3.2%)	1(1.3%)	#[a]0.43
Socio-economic status			
Low income	43.50%	46.20%	
Middle income	51.6%	50%	#[a]0.928
High income	4.8%	3.8%	

Group I= Smear negative pulmonary TB, Group II= Non-TB pulmonary cases
#[a]Chi-squared Test ($c^2$2), #[b]Students t-test were performed.
* Significant

Table II shows that SNPTB patients had a significantly higher frequency of cough, fever and weight loss compared to non-TB pulmonary cases (p-value<0.05). In contrast, chest pain and shortness of breath were more frequent in Group II than in Group I (p<0.05).

Table II

Distribution of study population according to clinical features (n=140)

Clinical features	Group I (n=62) No. (%)	Group II (n=78) No. (%)	p-value#
Cough	62(100)	71(91)	0.017*
Sputum	49(79.0)	52(66.7)	0.105
Hemoptysis	4(6.5)	8(10.3)	0.424
Fever	46(74.2)	36(46.2)	0.001*
Weight loss	32(51.6)	25(32.1)	0.019*
Chest pain	7(11.3)	24(30.8)	0.006*
Shortness of breath	10(16.1)	38(48.7)	<0.001*

Group I= Smear negative pulmonary TB, Group II= Non-TB pulmonary diseases
Chi-squared Test (χ^2) was performed.
* Significant

Table III shows that maximum SNPTB patients (74.2%) had ESR in 1st hour 50-100 mm, while maximum non-TB pulmonary cases had ESR <50 mm in 1st hour which was statistically significant (p<0.001).

Table III

Distribution of study population according to ESR level (n=140)

ESR in 1st hour	Group I (n=62) No. (%)	Group II (n=78) No. (%)	p-value#
<50 mm	8(12.9)	61(78.2)	<0.001*
50-100 mm	46(74.2)	14(17.9)	
>100 mm	8(12.9)	3(3.8)	

Group I= Smear negative pulmonary TB, Group II= Non-TB pulmonary diseases
Chi-squared Test (χ^2) was performed.
* Significant

Table IV shows that maximum number of SNPTB patients (69.4%) had positive Mantoux test (MT), while all of the non-TB pulmonary patients were negative for MT, which was statistically significant (p<0.001).

Table IV

Distribution of study population according to Mantoux test (n=140)

MT test	Group I (n=62) No. (%)	Group II (n=78) No. (%)	p-value#
Positive	43(69.4)	0(0)	<0.001*
Negative	19(30.6)	78(100)	

Group I= Smear negative pulmonary TB, Group II= Non-TB pulmonary diseases

Chi-squared Test (χ^2) was performed. * Significant

Table V shows that Sputum for gram stain was positive in 14.5% of SNPTB patients and 23.1% of non-TB pulmonary disease patients without any statistical significance (p>0.05).

Table V

Distribution of study population according to sputum examination (n=140)

Sputum	Group I (n=62) No.(%)	Group II (n=78) No.(%)	p-value#
Gram Stain positive	9(14.5)	18(23.1)	0.202
Gene Xpert positive	62(100)	0(0)	1.00

Group I= Smear negative pulmonary TB, Group II= Non-TB pulmonary diseases

Chi-squared Test (χ^2) was performed.

Table VI shows that serum ADA was significantly higher among SNPTB patients compared to non-TB pulmonary patients (48.16±12.13 vs 18.64±7.85 IU/L, p<0.001).

Table VI

Comparison between the two groups according to serum ADA values (n=140)

	Serum ADA level (IU/L)		p-value#
	Mean±SD	Range (min-max)	
Group I	48.16±12.13	21-81	<0.001*
Group II	18.64±7.85	5-38	

Group I= Smear negative pulmonary TB, Group II= Non-TB pulmonary diseases

Student t-test was performed. * Significant

ROC analysis of serum ADA in the diagnosis of patients with SNPTB found an AUC of 0.9850 (95% CI 0.969-1.00) which was statistically significant (p<0.001). A cut-off value measured ≥33 IU/L showed 93.55% sensitivity and 94.87% specificity (Fig.-II and Table VII)

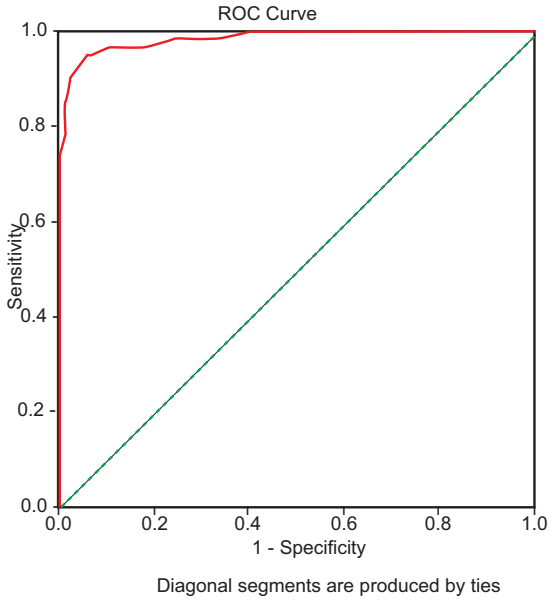

Fig.-2: *ROC analysis of serum ADA in diagnosis of patients with SNPTB (n=140)*

Table VII

Result of ROC curve

AUC	Standard error	95% CI		P value
	Lower	Upper		
0.985	0.005	0.969	1.00	<0.001*

AUC: Area under the curve; CI: Confidence Interval

* Significant

Table VIII shows that, among 62 SNPTB cases, a cut-off value of serum ADA of e”33 IU/L could detect truly 58 cases of SNPTB.

Table VIII

Cross tabulation of serum ADA between the two groups based on derived cut-off value (33 IU/L) (n=140)

Serum ADA (IU/L)	Smear negative Pulmonary TB		Total
	Yes	No	
≥33 IU/L	True positive (TP)	False positive (FP)	TP+FP
	58	4	62
<33 IU/L	False negative (FN)	True negative (TN)	FN+TN
	4	74	78
	TP+FN	FP+TN	
	62	78	140

A cut-off value of serum ADAe" 33 IU/L showed sensitivity, specificity, PPV, NPV, PLR, NLR, and accuracy 93.55%, 94.87%, 93.55%, 94.87%, 17.98%, 0.07%, and 94.29%, respectively (fig.-6). The cut-off value for serum ADA e" 33 IU/L has been derived from Karumuri et al. (2010).

Positive likelihood ratio: An individual having serum ADA value e" 33.IU/L is 17.98times more likely to have SNPTB compared to individuals having serum ADA d" 33 IU/L.(PLR > 1 indicates a test has diagnostic value).

Diagnostic accuracy: Serum ADA ≥33 IU/L can detect 94 individuals correctly with SNPTB among 100 individuals.

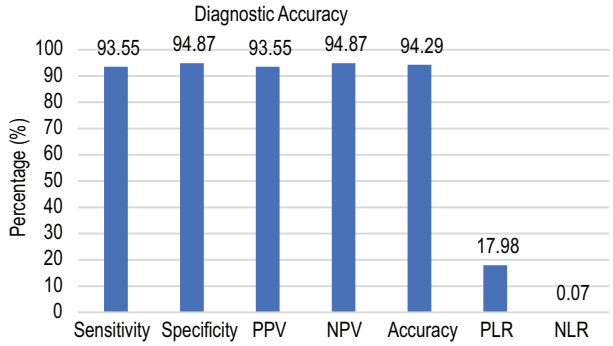

Fig.-3: Diagnostic accuracy of Serum ADA to distinguish smear-negative pulmonary TB from non-TB pulmonary diseases (n=140)

Discussions:

In this study, the majority respondentsin Group I were aged between 41-50 years (48.4%) followed by (35.5%) in the 31-40 years age group with a mean age of 43.02±8.49 years. Among patients in Group II, the majority were in the age group of 51-60 years (42.3%) followed by (35.9%) greater than 60 years with a mean age of 52±8.56 years. In line with my study findings, previous studies also found almost similar age distribution with male preponderance among pulmonary tuberculosis patients (Mohankudo et al. 2019; Malempati and Medooru 2018; Pandey et al. 2016; Shah 2015).[13,15,16,17]

The majority of the participants were male in this study. This might be because of more exposure of economically productive males to the external environment. In a previous study, the differences in sociodemographic characteristics in two periods, from the very beginning of the 21st century and 10 years after, were examined. In both observed periods, male people suffered from tuberculosis more frequently. (Smiljic et al. 2018).[18] In the low socioeconomic background of our country, females are given less attention, and access to healthcare facilities is limited.

The majority of the patients were garment workers in Group 1 (38.70%) and service holders in Group II (24.40%). Most of them belonged to middle-incomefamilies in our study, about 51.60% and 50% respectively in Group I and Group II. This might be due to more exposure to the external environment, working in overcrowded places, inadequate nutrition, alteration in immune function, poor ventilation, and poor hygiene habits.

Cough was the predominant clinical feature in 100% of patients of Group I, followed by 79% sputum production, 74.2% fever and 51% weight loss. Sajith et al. (2015) found in their study that cough with expectoration was prevalent in 96.5% of TB patients followed by weight loss (80.7%), fever (73.7%), and loss of appetite (54.4%).[19]

Maximum patients in Group I (74.2%) had ESR in 1st hour of 50-100 mm, while maximum non-TB pulmonary cases had ESR <50 mm in 1st hour which was statistically significant ($p < 0.001$). Mandal and Chavan (2016) found in their study that, ESR was elevated in 87% and normal in 26% of pulmonary TB patients.[20] About 69.4% of patients in Group I had Mantoux test positive with 100% negative in non-TB pulmonary cases in this study. Karumuri, et al. (2010) also found in their study that about 75% of pulmonary TB patients had positive Mantoux test.[21]

Sputum for gram stain was positive in 14.5% of SNPTB patients and 23.1% in non-TB pulmonary cases without any statistical significance ($p > 0.05$). Sputum for Xpert MTB/RIF was 100% positive in Group I.

In this study, serum ADA was significantly higher among Group I patients compared to Group II (48.16 ± 12.13 vs 18.64 ± 7.85 IU/L, $p<0.001$). Similarly, Chander and Shrestha (2012) also found in their study that the mean serum ADA among smear-negative TB cases was (42.26 ± 21.22 U/L) andhealthy control was (18.88 ± 6.67 U/L) with statistical significance ($p<0.0001$).[22]Alaarag et al. (2016) found mean ADA of (42.26 ± 21.22 U/L) and (23.31 ± 8.22 U/L) in smear-negative pulmonary TB and non-TB pulmonary cases respectively in their study with statistical significance ($p<0.001$).[23]Karumuri, et al. (2010) in their study found a higher mean ADA of (41.6 ± 6.4 U/L) in smear-negative TB cases compared to healthy controls (15.5 ± 0.5 U/L) with statistical significance ($p<0.001$).[21] Saini et al. (2018) reported a mean ADA of (39.478 ± 32.22 U/L) in sputum-negative TB cases and (11.819± 8.0235 U/L) in control groups with statistical significance ($p<0.00$).[1] In a study by Shah (2015), the serum ADA level in smear-negative pulmonary TB subjects was (35.12 ± 12.1 U/L) which was statistically significant ($p<0.001$) as compared to that of in healthy

subjects (14.603 ± 4.69 U/L).[17] Agarwal et al. (2019) also reported that serum ADA levelin smear-negative pulmonary TB subjects was found to be highly significant (38.48 ±10.56 vs 15.30 ± 0.23 U/L, p<0.001) as compared to that of healthy subjects.[24]

In the present study, ROC analysis of serum ADA in the diagnosis of patients with smear-negative pulmonary TB cases found an AUC of 0.9850 (95% CI 0.969-1.00) which was statistically significant (p<0.001). A cut-off value of serum ADA ≥33 IU/L showed sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and diagnostic accuracy of 93.55%, 94.87%, 93.55%, 94.87% and 94.29% respectively to correctly diagnose smear-negative pulmonary TB cases. Karumuri et al. (2010) also found 33 U/L as the cut-off value for serum ADA in diagnosing pulmonary tuberculosis in their study with a sensitivity 98.06% and specificity of 95.35%.[21] Similarly, Kanchan et al. (2014) evaluated the usefulness of ADA with a cut-off value of 33.3 U/L in serum to diagnose pulmonary tuberculosis patients efficiently with sensitivity, specificity, positive predictive value, negative predictive value of 96.69%, 96.69 %, 96.69% and 96.69% respectively.[25] Besides, an exact similar cut-off point (serum ADA e" 33 U/L) was also reported by Jhamaria et al. (1988) in diagnosing TB patients from patients with non-tubercular diseases with a specificity 100% and sensitivity of 98%.[26]Alaarag et al. (2016) found the sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) of serum ADA to be 95%, 86.7%, 90.5%, and 92.9% respectively, at 30 U/L cut-off point.[23] Pandey et al. (2016) found a cut-off value of serum ADA for TB diagnosis of 30 U/L, with sensitivity, specificity, positive predictive value (PPV), negative predictive value(NPV) of 81.3%, 100%, 81.33%, and 100% respectively.[13] Chander and Shrestha (2012) showed that at a serum ADA level of 30 U/L as a cut-off value, serum ADA had sensitivity, specificity, positive predictive value (PPV), negative predictive value of 83.10%, 91.25%, 94% and 69.52% respectively.[22]

However, according to the cut-off point that has been used in different research, sensitivity, specificity, PPV, and NPV have been reported differently, and therefore the outcomes of different studies must be interpreted cautiously. Since there is no program in order to standardize the ADA results, determining a cut-off point for ADA must be dependent on the type of method and defined separately for each area. Studies have shown that in areas in which tuberculosis is endemic, test sensitivity is of high importance.[27,28] In my research project, the sensitivity for truly diagnosing smear-negative pulmonary TB cases from non-TB pulmonary cases was also high (93.55%). Besides, the negative predictive value of this test was also high (94.87%) and this gives it a place as a widely usable screening test to exclude smear-negative pulmonary TB. Therefore, the determination of serum ADA should be done routinely, particularly if the diagnosis of tuberculosis is in doubt, and also to differentiate smear-negative pulmonary tuberculosis from non-tubercular pulmonary diseases.

Conclusion:

In this study, serum ADA was significantly higher in smear-negative pulmonary TB (SNPTB) patients than in non-TB pulmonary cases, with a remarkable diagnostic accuracy. These results correspond with the findings of previous studies with slight variations. Hence, the present study suggests to use of serum ADA estimation as the biochemical marker in the diagnosis of SNPTB highlighting it as a simple, rapid, cheaper, and accurate diagnostic test.

Limitations of the study:

Although the results of this study support the hypothesis,there are some facts to be considered which might haveaffected the result of the current study. It was a single-center study. The study population was relativelysmall.

Data Availability: The datasets analyzed during the current study are not publicly available due to the continuation of analyses but are available from the corresponding author upon reasonable request.

Conflict of Interest: The authors stated that there is no conflict of interest in this study

Funding: This research received no external funding.

Ethical consideration: The study was approved by the Ethical Review Committee of Dhaka Medical College & Hospital, Dhaka, Bangladesh. Informed consent was obtained from each participant or caregiver of the patients.

Author Contributions: All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis, and interpretation, or all these areas; took part in drafting, revising, or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

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References:

1. Saini V, Lokhande B, Jaswal S, Aggarwal D, Garg K & Kaur J. Role of serum adenosine deaminase in pulmonary tuberculosis. Indian Journal of Tuberculosis. 2018;65(1):1-12. https://doi.org/10.1016/j.ijtb.2017.08.001. PMid:29332644
2. Das AC. Epidemic situation of tuberculosis in Bangladesh: An overview. South East Asia Journal of Public Health.2017;6(2):61-62. https://doi.org/10.3329/seajph.v6i2.31837
3. Ryu YJ. Diagnosis of pulmonary tuberculosis: Recent advances and diagnostic algorithms. Tuberculosis And Respiratory Diseases.2015;78:64-71. https://doi.org/10.4046/trd.2015.78. 2.64. PMid:25861338 PMCid:PMC4388902
4. World Health Organization (WHO), World Tuberculosis Day 2017. https://www.who.int/bangladesh/news/detail/23-03-2017-world-tuberculosis-day-2017 [accessed 08.08.2021]
5. Barua R & Hossain M.Adenosine Deaminase in diagnosis of tuberculosis: A Review. Anwer Khan Modern Medical College Journal.2014;5(2):43-48. https://doi.org/10.3329/akmmcj.v5i2.21132
6. Kanchan SK, Santosh VG, Vishal SM, Leela AG, Niyogi Guha NN, et al. Study of adenosine deaminase levels in patients of pulmonary tuberculosis with and without Pleural Effusion. IOSR Journal of Dental and Medical Sciences.2014;13(1):30-37. https://doi.org/10.9790/0853-13193037
7. Ryu YJ.Diagnosis of pulmonary tuberculosis: Recent advances and diagnostic algorithms. Tuberculosis And Respiratory Diseases. 2015;78:64-71. https://doi.org/10.4046/trd.2015.78.2.64. PMid:25861338 PMCid:PMC4388902
8. Salmanzadeh S, Tavakkol H, Bavieh K & Alavi SM.Diagnostic value of serum adenosine deaminase (ADA) level for pulmonary tuberculosis.Jundishapur Journal of Microbiology.2015;8(3):1-5. https://doi.org/10.5812/jjm.21760. PMid:25861440 PMCid:PMC4385252
9. Jeon CY and Murray MB. Diabetes mellitus increases the risk of active tuberculosis: a systematic review of 13 observational studies. PLoS Med, 2008;5(7):152. https://doi.org/10.1371/journal.pmed.0050152. PMid:18630984 PMCid:PMC2459204
10. McShane H. Co-infection with HIV and TB: double trouble. International Journal of STD & AIDS. 2005;16(2):95-100. https://doi.org/10.1258/0956462053057576. PMid:15807935
11. Rajeswari R, Chandrasekaran V, Suhadev M, Sivasubramaniam S, Sudha G & Renu G. Factors associated with patient and health system delays in the diagnosis of tuberculosis in South India. The International Journal of Tuberculosis And Lung Disease. 2002;6(9):789-95
12. Bhoumik SK, Rahman MM, Ibrahim M, Hiron MM & Ahamad M.Evaluation of adenosine deaminase (ADA) activity for diagnosis of tubercular pleural effusion. Bangladesh Journal of Medical Biochemistry.2013; 6(2):40-48. https://doi.org/10.3329/bjmb.v6i2.17642
13. PandeyR, Tamrakar D, Jaiswal S, Sharma A, Koju S, Duwal SR, et al.Serum adenosine deaminase: A novel biomarker tool for the diagnosis of tuberculosis. Biosciences Biotechnology Research Asia.2016;13(1): 551-556. https://doi.org/10.13005/bbra/2068
14. Stavanovic G, Pelemis M, Pavlovic M, Lavandinivic L, Dakic Z, Milosevic I, et al. Significance of adenosine deaminase serum concentrations in the diagnosis of extra-pulmonary tuberculosis. Journal of IMAB.2011;17:130-134. https://doi.org/10.5272/jimab.2011171.130
15. Mohankudo S, Manjhi R, Dutta P, Pothal, S, Jagaty SK &Chhotray P.Role of serum ADA in diagnosis of pulmonary tuberculosis. International Journal of Medical Science and Advanced Clinical Research.2019;2(4):57-62.
16. Malempati UD &Medooru KK. Evaluation of adenosine deaminase activity in serum and pleural fluid of pulmonary tuberculosis patients with pleural effusion. International Journal of Research in Medical Sciences.2018;6(10):3358. https://doi.org/10.18203/2320-6012.ijrms20184046
17. ShahVK.Study of serum adenosine deaminase (ADA) level in the diagnosis of extrapulmonary and smear-negative tuberculosis. International Journal of Scientific Research, 2015;4(2277):4-6.
18. Smiljiæ S, Stanisavljeviæ D, Radoviæ B, Mijoviæ M, Saviæ S, Ristiæ S, et al. The sociodemographic characteristics and risk factors for tuberculosis morbidity between two decades at the beginning of the 21st century in the north of Kosovo, Serbia. VojnosanitetskiPregled.2018;75(5):461-467. https://doi.org/10.2298/VSP160323335S
19. SajithM, Thomas A, Kothia JJ, Chandrakar B, Bargaje, MD, et al. Socio-demographic characteristics of tuberculosis patients in a tertiary care hospital. International Journal of Medical and Health Research.2015;1(3):25-28
20. Mandal SK and Chavan L. Erythrocyte sedimentation rate values in cases of active tuberculosis without HIV co-infection. Journal of Medical Science and Clinical Research. 2016;4(10):13156-13159. https://doi.org/10.18535/jmscr/v4i10.58
21. Karumuri SR, Kumar H, Rudresh BM, Srinivas T & Bhat K.A Comparative study and evaluation of serum adenosine deaminase activity in the diagnosis of pulmonary tuberculosis. Biomedical Research.2010; 21:189-194.
22. Chander A & Shrestha CD. Diagnostic value of serum adenosine deaminase levels in sputum smear-negative

pulmonary tuberculosis patients in the Nepalese population. Asian Pacific Journal of Tropical Biomedicine.2012;2(3 SUPPL.):S1896-S1899. https://doi.org/10.1016/S2221-1691(12)60517-6

23. Alaarag AH, Mohammad OI & Farag NM.Diagnostic utility of serum adenosine deaminase level in the diagnosis of pulmonary tuberculosis. Egyptian Journal of Bronchology.2016;10(2):133-139. https://doi.org/10.4103/1687-8426.184369

24. Aggarwal AN, Agarwal R, Sehgal IS, Dhooria S. Adenosine deaminase for diagnosis of tuberculous pleural effusion: A systematic review and meta-analysis. PLoS One. 2019;14(3):e0213728. https://doi.org/10.1371/journal.pone.0213728. PMid:30913213 PMCid:PMC6435228

25. Kanchan SK, Santosh VG, Vishal SM, Leela AG, Niyogi Guha NN, Joshi A, et al. Study of adenosine deaminase levels in patients of pulmonary tuberculosis with and without Pleural Effusion. IOSR Journal of Dental and Medical Sciences.2014;13(1):30-37. https://doi.org/10.9790/0853-13193037

26. Jhamaria JP, Jenaw RK, Luh SK, Mathur DK, Parihar HL, Sharma SK, et al. Serum adenosine deaminase (ADA) in differential diagnosis of pulmonary tuberculosis and common non-tubercular respiratory diseases. Indian Journal of Tuberculosis, 1988;35:25-27.

27. Mohammad TZ, Mashayekhpour S, Mohammadi F, Mansouri D &Masjedi MR.Diagnostic value of adenosine deaminase isoenzyme (ADA2) and total ADA in tuberculous pleural effusion. Tanaffos.2005;4:37-42.

28. Binesh F, Jalali H, Zare MR, Behravan F, Tafti AD, Behnaz F, et al. Diagnostic value of sputum adenosine deaminase (ADA) level in pulmonary tuberculosis. Germs.2016;6(2):60-65. https://doi.org/10.11599/germs.2016.1090. PMid:27482515 PMCid:PMC4956162.