Annals of Clinical Case Reports

Research Article
Published: 31 Oct, 2024

Tuberculous Pleurisy: The Role of the ADA Enzyme in Diagnosis and Treatment Outcomes

Abdugapparov F, Mamatov L and Ongarbayev D*

Department of Phthisiology and Pulmonology, Tashkent Medical Academy, Uzbekistan

Abstract

Purpose: To study the role of the ADA enzyme in diagnosis and treatment outcomes.

Methods: The study conducted here was a cohort study that relied on secondary data sourced from medical charts and TB forms of patients. Specifically, the study focused on individuals who were referred to the inpatient department of the Republican Specialized Scientific Practical Medical Center of Phthisiology and Pulmonology (RSSPMCPP) in Tashkent, Uzbekistan, between the years 2021 and 2022.

Results: From 2021 to 2022, the pleural fluid and blood serum of 80 patients were examined in the clinics of the Republican Specialized Physiology and Pulmonology Scientific and Practical Medical Center and the Tashkent Clinical Hospital of Physiology and Pulmonology. All of them were divided into 2 groups - (1) with TP, (2) with pleurisy of non-TB etiology. Group 1 patients had higher ADA in pleural fluid and blood serum than those in group 2. It was observed that the amount of ADA in pleural fluid was 3.2 times higher in group 1 than in group 2 (46,6 to 14,4 respectively), and 5.5 times higher in blood serum (25,3 to 4,5 respectively), respectively. The majority of patients examined had a successful treatment outcome (n=50, 91%), which was more common among people aged 40 years and younger (90.9%) compared to the older group (89.2%), with differences there was practically nothing between them. Gender groups (90.4% and 87.5% among men and women, respectively). Age or gender was not significantly associated with the risk of poor treatment outcome. Six (8.7%) patients had evidence of resistance to at least rifampicin, and the presence of drug-resistant tuberculosis significantly increased the risk of treatment failure (RR 3.97; 95% CI: 1.13-13.93, P value 0.031). Hepatitis was the only comorbidity significantly associated with the risk of treatment failure (RR 4.8; 95% CI: 1.44-15.98, P value 0.011).

Conclusion: Due to the varying sensitivity and specificity of different diagnostic approaches, the diagnosis of pleural effusion remains challenging. Multidisciplinary approaches are required to maximize diagnostic accuracy and minimize the likelihood of misdiagnosis of TP.

Keywords: Tuberculosis; Pleural effusion; ADA; Diagnosis; Pleural fluid aspiration; Pleural biopsy; Treatment outcome

OPEN ACCESS

Correspondence:
Fazlkhan Abdugapparov, Department of Phthisiology and Pulmonology, Tashkent Medical Academy, Farabi 2, Tashkent 100109, Uzbekistan,

Received Date: *03 Oct 2024*
Accepted Date: *26 Oct 2024*
Published Date: *31 Oct 2024*

Citation:
Abdugapparov F, Mamatov L, Ongarbayev D. Tuberculous Pleurisy: The Role of the ADA Enzyme in Diagnosis and Treatment Outcomes. Ann Clin Case Rep. 2024; 9: 2694.
ISSN: 2474-1655.

Introduction

A common extrapulmonary manifestation of tuberculosis is pleurisy [1,2]. Tuberculous Pleurisy (TP) develops when mycobacteria secrete an antigenic protein into the pleural cavity. This causes an incompletely understood slow type of sensitization reaction, and fluid accumulates in the pleural cavity. Difficulties usually lie not in the diagnosis of pleurisy itself, but in determining its etiology for timely etiotropic treatment. The fact is that, in addition to tuberculosis, the presence of pleural fluid can be caused by pneumonia, malignant tumors, heart failure, cirrhosis of the liver, nephrotic syndrome, infectious non-tuberculous lung disease, and diffuse connective tissue diseases [3-5]. Immune status can also influence the incidence of tuberculous pleurisy. Because the main mechanism is a delayed hypersensitivity reaction, one might hypothesize that the immunocompromised hosts are less likely develop tuberculous pleurisy than the immunocompetent host. However, incidence of tuberculous pleurisy is higher in Human Immunodeficiency Virus (HIV) - infected patients than in non-infected patients [6]. On the other hand, higher incidence is not observed in renal transplant and dialysis patients [7,8].

The differential diagnosis of TP usually includes invasive procedures such as pleural biopsy and thoracoscopy [6,9-13]. These manipulations require special skills of medical personnel and can worsen the patient's condition. The high cost and long time needed to obtain results further

reduce the effectiveness of the pleural biopsy and bacteriological method, which is considered the "gold standard" of diagnosis [14]. The difficulty of diagnosing TP is complemented by the relatively low sensitivity of traditional methods. Acid-fast bacteria are detected in 20-30% of pleural fluid examinations and 50% to 80% of pleural biopsy specimens. The sensitivity does not exceed 78% even when using the polymerase chain reaction to detect mycobacteria [8]. At the same time, it is known that there are very sensitive biochemical markers in the pleural fluid, the determination of their concentration can significantly facilitate the differential diagnosis of TP [2]. The ADA enzyme is present in the cytoplasm of all mammalian tissue cells. It is involved in purine metabolism and catalyzes the deamination of adenosine and 2-deoxyadenosine to inosine and deoxynosine, respectively. There are several isoenzyme forms of ADA, the most important of which are ADA1 and ADA2. ADA1 isoenzyme is present in all cells of the body, but in the highest concentration in lymphocytes and monocytes. ADA2 isoenzyme is present only in monocytes and macrophages.

Aim of the Study

Study the role of the ADA enzyme in diagnosis and treatment outcomes.

Materials and Methods

Study design

The study conducted here was a cohort study that relied on secondary data sourced from medical charts and TB forms of patients. Specifically, the study focused on individuals who were referred to the inpatient department of the Republican Specialized Scientific Practical Medical Center of Phthisiology and Pulmonology (RSSPMCPP) in Tashkent, Uzbekistan, between the years 2021 and 2022. The main objective of the study was to investigate TB pleural effusion in this patient population.

Study setting

In Uzbekistan, the commendable task of delivering comprehensive care for Tuberculosis (TB) patients is entrusted to the National TB Program, which operates in seamless coordination with the Republican Specialized Scientific-Practical Medical Center for Phthisiology and Pulmonology (RSSPMCPP). This collaboration enables the efficient implementation of various crucial activities pertaining to TB diagnosis and treatment, all of which are delivered to patients without any financial burden.

The provision of top-notch TB care spans across the entirety of Uzbekistan's robust healthcare system, catering to patients at every level of medical assistance. At the primary level, individuals seeking care for presumptive TB can easily access a network of primary healthcare facilities, commonly known as polyclinics. These polyclinics serve as the initial point of contact for patients, offering a gateway into the comprehensive TB care services available. The delivery of Tuberculosis (TB) care in Uzbekistan follows a well-structured framework involving multiple levels of care. At the district level, TB clinics act as the second level of care, where patients undergo initial examinations, including a range of laboratory tests such as microscopy, Xpert MTB/RIF assays, and X-rays. Based on the results, patients may be referred to higher levels of care for additional laboratory examinations, such as mycobacterial culture and the Mycobacteria Growth Indicator Tube (MGIT). Additionally, patients requiring inpatient care are also directed to the third level of TB care. For more complex cases and patients with advanced co-morbidities, the fourth level of TB care is provided at the RSSPMCPP. This level of care is dedicated to managing patients with complicated diagnoses and ensuring the highest standards of treatment. Patients with presumptive TP typically enter the first and second levels of TB care, where they undergo initial assessments and examinations. Depending on the need for a more precise diagnosis and treatment, they may be further referred to the third and fourth levels of care. If sputum smear-positive, these patients are classified as pulmonary TB cases and receive inpatient TB care tailored specifically for pulmonary TB. It is important to note that the national TB treatment protocols implemented in Uzbekistan strictly adhere to the guidelines provided by the World Health Organization (WHO). This adherence ensures the highest quality of care and standardization in the management of TB cases throughout the country [8]. The same standard treatment regimens are used for patients with both pulmonary TB and EPTB.

Study population

The study included all patients with presumptive TB Pleural Effusion (TPE) who were admitted to the inpatient department of RSSPMCPP between 2021 and 2022. The final diagnosis was determined through a comprehensive approach, involving the assessment of clinical symptoms, radiological evaluations (including chest X-ray, ultrasound, and chest CT scan), and the examination of pleural fluid samples. Various tests were performed on the pleural fluid, such as bacteriological examinations (including smear microscopy for AFB, Xpert MTB/RIF, and mycobacterial culture), as well as cytological and biochemical examinations. In addition, histological evaluation of the pleural biopsy was conducted to aid in the final diagnosis.

Data collection and validation

The data from the patients' medical charts and TB forms were recorded in standardized electronic records. These records were created using the EpiData application (version 3.1 EpiData Association, Odense, Denmark). To ensure accuracy, the data underwent thorough verification and error-checking processes, including cross-tabulation and analysis of extreme values. Any inconsistencies or discrepancies identified during this process were resolved by referring back to the source documents.

Data variables

To meet the study objectives, several demographic, socio-economic, and medical history-related variables were taken into consideration. These included age, sex, place of residence, tobacco use, alcohol use, co-morbidities (diabetes mellitus, Human Immunodeficiency Virus (HIV), Hepatitis C Virus (HCV)), known diagnosis of TPE, drug-resistance profile, treatment information and outcome, details on sputum and pleural fluid examination (microscopy, Xpert MTB/RIF, mycobacterial culture, cytology, ADA), and histology.

Definitions

Diabetes status was identified based on clinical history or blood glucose measurement followed by an endocrinologist's evaluation. All patients were tested for HIV and HCV antibodies at baseline and positive rapid test results were further confirmed. Data on smoking and alcohol use were obtained based on patient self-reports. Tuberculosis treatment outcomes were classified according to the WHO recommended definitions: successful outcome (including cure and treatment completion) and unsuccessful treatment outcome

comprising failure, loss to follow-up, and death.

Analysis and statistics

Descriptive statistics were used to describe patient demographic and clinical characteristics. These included: frequencies, proportions, measures of central tendency (mean), and variation (standard deviation). The level of significance was set at $p<0.05$. Risk Ratios (RR) as a measure of association between predictors and treatment outcome (unfavorable *vs.* favorable) were calculated using binomial log-linear regression and presented in respective tables. Along with the parameter estimates, 95% Confidence Intervals (CI) and p values were calculated as well.

Results and Discussion

From 2021 to 2022, the pleural fluid and blood serum of 80 patients were examined in the clinics of the Republican Specialized Physiology and Pulmonology Scientific and Practical Medical Center and the Tashkent Clinical Hospital of Physiology and Pulmonology. All of them were divided into 2 groups - (1) with TP, (2) with pleurisy of non-TB etiology. The age of the examined patients was from 20 to 86 years. The characteristics of the groups are presented in Table 1. Get the material. Pleural fluid samples were obtained by thoracentesis. About 40 ml of pleural fluid was obtained from each examined person. A part of the liquid was taken to calculate the cell content, cytological examination, staining of acid-resistant bacteria, and determination of the amount of protein. Another part of the liquid was centrifuged at 1500 rpm for 10-15 minutes, the resulting supernatant was separated and stored at -20°C and used for direct study of ADA. In parallel, a pleural biopsy was performed, during which samples were taken for research, tissues used for pathohistological and microbiological examination. Tuberculosis was diagnosed when any of the following analysis results were met: Mycobacterium tuberculosis was detected in pleural fluid or pleural biopsy, granuloma and acid-fast bacteria were detected in pleural tissue, or tuberculosis was detected in pleural tissue with granuloma and no acid-fast bacteria were found. When effective treatment is observed.

The diagnosis of pleurisy with tumor etiology was made on the basis of cytological examination of pleural fluid or histological analysis of pleural biopsy. Examination of ADA enzyme in blood serum and pleural fluid. ADA enzyme activity was determined by the method described by Giusti G. and Galanti B. This method is based on the Bertolet reaction (in the presence of ammonia separated from adenosine) and is related to the formation of a colored indophenol complex and the subsequent spectrophotometric estimation of its concentration. Results are expressed in international activity units (IU). A unit of ADA activity is the amount of enzyme required to release 1 mmol of ammonia in 1 minute under standard assay conditions.

Group 1 patients had higher ADA in pleural fluid and blood serum than those in group 2 (Table 2). It was observed that the amount of ADA in pleural fluid was 3.2 times higher in group 1 than in group 2, and 5.5 times higher in blood serum, respectively. We investigated the diagnostic value of ADA levels in pleural fluid and serum (Table 3).

The presented work presents the results of the first study carried out in the Republic of Uzbekistan to evaluate the use of ADA activity in pleural fluid and blood serum for the purpose of differential diagnosis of TP. They indicate the uniqueness of these tests. The increase in total ADA activity in pleural fluid and blood serum is

Table 1: Checked patients characteristics.

Groups	Number of patients examined	Male	Female	Age range	Average age
Tuberculous pleurisy (group 1)	50	42	8	20-83	42
Non-tuberculous pleurisy (group 2)	30	19	11	27-86	60

Table 2: Pleura liquid and blood ADA enzyme in serum activity.

Indicators being determined	Controlled groups	
	Group 1	Group 2
ADA IU/l in pleural fluid	46.6 (3-69)	14.4 (4-38)
ADA IU/l in blood serum	25.3 (0-37)	4.5 (0-19)

Note: $p<0.05$

Table 3: Tuberculosis in pleurisy Indicators of diagnostic value of ADA detection.

Indicators	ADA in pleura liquid IU/l	ADA in blood serum IU/l
Threshold result of the indicator	30	20
True positive results the number	47	21
False positive results the number	3	0
True negative results the number	27	30
False negative results the number	3	9
Sensitivity %	93	100
Specificity %	96	70

mainly due to the isoenzyme form of ADA2. A similar phenomenon was observed by other researchers. The test for total ADA activity in pleural fluid and blood serum appears to have high sensitivity and specificity compared with results obtained in other countries.

An analysis of literature data showed that among European countries, the threshold values of ADA in the pleural fluid of patients with tuberculous pleurisy range from 41 to 70 IU/l, and the sensitivity of the test ranges from 79% to 100% [3]. Even greater fluctuations, according to various laboratories, are characteristic of the threshold level of IFN-g (from 12 to 240 pg/ml) [14]. The reasons for such large differences are the use of different sets of reagents for enzyme immunoassay or radioimmunoassay, the incidence of tuberculosis in the population and the characteristics of the population itself [11]. Taking into account the above data, when choosing a test priority, issues of efficiency come to the fore. As studies show, the determination of total ADA activity in pleural fluid is not only clinical, but also economically effective, since the method is simple to perform and does not require expensive equipment and reagents. The result can be obtained within 2 hours. This method should, first of all, be recommended for widespread implementation in practical medicine. The majority of patients examined had a successful treatment outcome (n=50, 91%), which was more common among people aged 40 years and younger (90.9%) compared to the older group (89.2%), with differences there was practically nothing between them. Gender groups (90.4% and 87.5% among men and women, respectively). Age or gender was not significantly associated with the risk of poor treatment outcome. Six (8.7%) patients had evidence of resistance to at least rifampicin, and the presence of drug-resistant tuberculosis significantly increased the risk of treatment failure (RR 3.97; 95% CI: 1.13-13.93, P value 0.031). Hepatitis was the only comorbidity significantly associated with the risk of treatment failure (RR 4.8; 95% CI: 1.44-15.98, P value 0.011). Among the different diagnostic methods, there was no significant association between the diagnosis and treatment outcome of TP with bacteriological or histological

Table 4: Predictors of treatment outcome for patients with tuberculous pleurisy admitted for treatment at the Republican Scientific and Practical Medical Center, Tashkent, Uzbekistan, 2021-2022.

Characteristics	Total		Successful treatment result		Unsuccessful treatment outcome		R.R.	95% CI	P value
	N	%	N	(%)	N	(%)			
Age group									
<40 years	22	-44	20	-90.9	2	-9.1	1		
40 years and older	28	-56	25	-89.2	3	-10.8	1.37	(0.50–3.70)	0.54
Sex									
male	42	-84	38	-90.4	4	-9.6	1		
female	8	-16	7	-87.5	1	-2.5	1.11	(0.41–3.04)	0.836
Drug resistance									
Sensitive / not confirmed	44	-88	40	-90.9	4	-9.1	**1**		
Verified RR/MDR	6	-12	4	-66.7	2	-33.3	**3.97**	**(1.13-13.93)**	**0.031**
Hepatitis									
Yes	5	-10	3	-60	2	-40	**4.8**	**(1.44-15.98)**	**0.011**
No	45	-90	41	-91.1	4	-8.9	**1**		
Bacteriologically confirmed tuberculosis									
Yes	10	-20	9	-90	1	-10	0.96	(0.23–4.01)	0.958
No	40	-80	37	-92.5	3	-7.5	1		
Cytologically confirmed tuberculosis									
Yes	31	-62	29	-93.5	2	-6.5	**1**		
No	19	-38	16	-84.2	3	-15.8	**4.52**	**(1.05-19.47)**	**0.043**
Histologically confirmed tuberculosis									
Yes	28	-56	24	-85.7	4	-14.3	1		
No	22	-44	20	-90.9	2	-9.1	0.44	(0.14–1.42)	0.169

studies, however, we found an increased risk of unfavorable treatment outcome among patients whose diagnosis of TP was confirmed only by cytology (RR 4.52; 95% CI: 1. 05-19.47, P value 0.043) (Table 4).

Conclusion

Diagnosis of tuberculous exudative pleurisy is a difficult problem. Among non-conventional tests, ADA and IFN-γ have the best sensitivity and specificity, but they are biomarkers of pleural inflammation and do not confirm the etiological agent. There are limited data for other new tests and biomarkers. An important place should be given to the phthisiatric alertness of doctors in the general medical network. The ADA method is a complementary examination method, is simple to perform, and does not require expensive equipment and reagents. The result can be obtained within 2 hours. Due to the varying sensitivity and specificity of different diagnostic approaches, the diagnosis of pleural effusion remains challenging. Multidisciplinary approaches are required to maximize diagnostic accuracy and minimize the likelihood of misdiagnosis of TP. Patients with co-infections/conditions and patients with a drug resistance profile should be monitored more closely for successful completion of anti-TB treatment.

References

1. Abdugapparov F. Diagnostic procedures, diagnoses, and treatment outcomes of patients with presumptive tuberculosis pleural effusion in Uzbekistan. Int J Environ Res Public Health. 2021;18(11):5769.
2. Khodzhaeva M. Peculiarities of the Course of Tuberculosis When Combined with HIV Infection, Complicated by Visceral Mycosis. Интернаука. 2020;24:58-62.
3. Light RW. Pleural Diseases, 6th ed.; Lippincott Williams & Wilkins: Philadelphia, PA, USA, 2013.
4. Massavirov S. Risk Factors for Unfavorable Treatment Outcomes among the Human Immunodeficiency Virus-Associated Tuberculosis Population in Tashkent City, Uzbekistan: 2013–2017. Int J Environ Res Public Health. 2021;18(9):4623.
5. Mirzaboyev S. Zamonaviy Tibbiyot Sharoitida Birlamchi Aniqlangan Tuberkulyozning Turli Shakillarini Barvaqt Aniqlashda Raqamli Rentgen Diagnostikasining Ahamiyati. 2023.
6. Ongarbayev DO. Effectiveness of Diagnostics and Treatment of Tuberculosis in Patients with COVID-19. World Bulletin Public Health. 2023;20:29-33.
7. Steingart KR, Henry M, Laal S, Hopewell PC, Ramsay A, Menzies D, et al. A systematic review of commercial serological antibody detection tests for the diagnosis of extrapulmonary tuberculosis. Postgrad Med J. 2007;83:705-12.
8. Vorster MJ, Allwood BW, Diacon AH, Koegelenberg CFN. Tuberculous pleural effusions: Advances and controversies. J Thorac Dis. 2015;7(6):981-91.
9. World Health Organization. Global Tuberculosis Report 2020; World Health Organization: Geneva, Switzerland, 2020.
10. Abdugapparov F. The results of clinical and laboratory studies in patients with disseminated pulmonary tuberculosis: дис. Toshkent. 2023.
11. Abdugapparov F. Diagnosis and treatment results of patients with presumed tuberculous pleuritis in Uzbekistan // Bulletin. 2021;53.
12. Ongarbaev D, Abdugapparov F, Mamatov L. IIV Infection Sil Epidemiologysiga Taysiri: dis. – "Innovative approaches to the diagnosis, treatment and prevention of tuberculosis and nonspecific respiratory

pathology in adults and children". 2021.

13. Parpieva NN. Methods for detecting multidrug resistance in patients with tuberculosis: dis. Toshkent. 2023.
14. Khakimov AA. Assessment of the prevalence of bronchopulmonary and cardiovascular pathology in patients with newly diagnosed tuberculosis: dis. Toshkent. 2022.