

Original article:

Study of comparison of ADA activity alone vs ADA activity in combined with L/N ratio for diagnosing TB pleuritis

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Abstract:

Introduction: Pulmonary tuberculosis is the most frequent cause of death by an infectious agent worldwide. Among the extrapulmonary presentations after tuberculous lymphadenitis, pleural tuberculosis is the second most frequent. Failure to diagnose and treat pleural tuberculosis can result in progressive disease with the involvement of other organs in as many as 65% of patients.

Methods and materials : The present study was conducted at Navodaya Medical College, Raichur during the period between January 2011 and December 2011. 130 consecutive pleural fluid specimens from patients admitted to medical, surgical, gynaecologic, and paediatric wards were analysed.

In the present study, ADA activity was highest among the tuberculous group. Para-infective conditions were also seen to be associated with high ADA activities. The relative cell count or L/N ratio could be used to distinguish between these two entities⁵⁻⁸. In the cases of tuberculous pleurisy, a predominant lymphocyte count was usually found, resulting in a L/N ratio of 0.75 or greater, whereas in the case of para-infective effusions, a predominant neutrophil count was usually found (L/N ratio <0.75).

Conclusion: In conclusion, it is suggested that the combined use of adenosine deaminase activity along with lymphocyte neutrophil ratio would provide a more efficient means for diagnosing tuberculous pleuritis than the use of ADA alone.

Introduction

Pulmonary tuberculosis is the most frequent cause of death by an infectious agent worldwide. Among the extrapulmonary presentations after tuberculous lymphadenitis, pleural tuberculosis is the second most frequent. Failure to diagnose and treat pleural tuberculosis can result in progressive disease with the involvement of other organs in as many as 65% of patients.

Pleural effusions may arise secondary to pulmonary or systemic disease and their development is classically associated with an influx of inflammatory cells into the pleural space⁴. Lymphocytes predominate in malignant and tuberculous pleural effusions¹⁻⁴.

Hence this study is aimed to determine whether combined use of pleural fluid lymphocyte/neutrophil ratio and ADA activity would provide a more efficient means for diagnosing tuberculous pleurisy than the use of ADA levels alone.

Methods and materials

The present study was conducted at Navodaya Medical College, Raichur during the period between January 2011 and December 2011. 130 consecutive pleural fluid specimens from patients admitted to medical, surgical, gynaecologic, and paediatric wards were analysed.

Inclusion criteria:

All exudative pleural effusion cases

Exclusion criteria:

1. Patients with transudative pleural effusion
2. Patients with malignant pleural effusion
3. Patients with immunodeficient states like HIV/AIDS, those on chemotherapy were excluded
4. Patients having hemothoraces or empyemas too turbid for analysis were excluded

Besides a detailed history and clinical examination, the following investigations were carried out:

Determination of adenosine deaminase

There are several methods for estimating adenosine deaminase in the pleural fluid. The colorimetric method of Guisti and Galanti⁶⁸ is the method used here. It was developed by Giuseppe Guisti and Bruno Galanti in 1974 and is a simple and sensitive method.

Adenosine deaminase is an enzyme of the purine catabolism that catalyses the conversion of

adenosine to inosine. The ammonia which is produced during the reaction can be assayed by a colorimetric method. This forms the basis of the Guisti and Galanti method of adenosine deaminase estimation. This method which uses adenosine, 20mmol/l in the assay mixture ensures optimized conditions for measuring ADA activity from various sources.

Ammonia is determined by the Chaney and Marbach modification of Berthelot's reaction. Ammonia on reacting with sodium hypochlorite and phenol in alkaline solution forms an intensely blue indophenols. Sodium nitroprusside is the catalyst. The ammonia concentration is directly proportional to the absorbance of indophenols. The reaction catalysed by ADA is stopped at the end of the incubation period by the addition of phenol nitroprusside solution.

The sensitivity, specificity, positive predictive value, negative predictive value for pleural fluid ADA >50U/L alone and combined pleural fluid ADA >50U/L and lymphocyte neutrophil ratio of >0.75 were calculated and compared using student t-test.

Observations:

Table No.1 : Showing distribution of ADA alone and response to treatment

ADA alone (U/L)	Improved	Not Improved	Total
≥50	38	8	46
<50	24	20	44
total	62	28	90

Table No.2 : Showing distribution of L/N ratio and response to treatment

L/N ratio	Improved	Not Improved	Total
≥0.75	62	8	70
<0.75	0	20	20
Total	62	28	90

Table No.3 : Showing combined use of ADA and L/N ratio and response to treatment

ADA and L/N ratio	Improved	Not Improved	Total
≥50U/L and ≥0.75	38	3	41
<50U/L and <0.75	0	15	15
Total	38	18	56

Table No.4 : Comparison of ADA activity alone vs ADA activity in combined with L/N ratio as a means for diagnosing TB pleuritis:

Criteria used to diagnose TB						
ADA level	L/N ratio	Sensitivity%	Specificity%	PPV%	NPV%	Efficiency%
≥50U/L	-	61	71	83	45	64
≥50U/L	≥0.75	100	83	93	100	95
-	≥0.75	100	71.4	88.6	100	91.1

Discussion

The origin of the increased ADA activity found in tuberculous effusions is uncertain. Many investigators have attributed the origin of these high levels to the fact that tuberculous pleurisy is a T-cell mediated response^{5,6}. However, insignificant or inconclusive results have been obtained for studies attempting to show a correlation between number of lymphocytes or lymphocyte populations and ADA levels. Other investigators have suggested a monocyte-macrophage origin of ADA^{7,8}.

Blood ADA levels are also increased in certain T-cell lymphoproliferative disorders and this can be expected to spill over to the pleura in these conditions^{7,2}. In the para-infective effusions, ADA probably originates from lymphocytes or neutrophils^{9,10}.

In the present study, ADA activity was highest among the tuberculous group. Para-infective conditions were also seen to be associated with high ADA activities. The relative cell count or L/N ratio could be used to distinguish between these two entities⁵⁻⁸. In the cases of tuberculous pleurisy, a predominant lymphocyte count was usually found, resulting in a L/N ratio of 0.75 or greater,

whereas in the case of para-infective effusions, a predominant neutrophil count was usually found (L/N ratio <0.75).

As already mentioned, malignant effusions may also be associated with high lymphocyte counts^{6,8}. The distinction between malignant and tuberculous effusions can usually be made on the grounds of ADA activity. In general, malignant effusions have lower ADA levels than those found in TB. However, effusions secondary to lymphomas and leukemias were generally associated with higher ADA activities than non-hematologic malignancies, and could be confused with tuberculous effusions on the grounds of ADA and L/N ratios.

Another source of false-positives could be rheumatoid pleuritis. Rheumatoid pleurisy appears to be a unique entity in that it could not be differentiated from pleural TB on the basis of ADA activity alone. In addition to studying the ADA activity in these patients, Ocana et al¹¹⁻¹³ also determined differential counts on these effusions.

TB pleurisy is traditionally diagnosed by either identification of *M tuberculosis* in pleural fluid and/or biopsy specimen cultures or from the presence of granulomas in the pleural biopsy tissue.

Pleural fluid cultures have a sensitivity 20-30%⁸¹, pleural biopsy specimens 50-80%¹⁴, depending upon the clinician's proficiency. Because of the long culture periods required, clinical and therapeutic decisions are often made before these laboratory results become available. Polymerase chain reaction, having a sensitivity of 78% for active disease¹⁵, has not been found to be an efficient alternative. Use of ADA level especially in conjunction with the L/N ratio, is therefore a

valuable diagnostic tool in this regard, as it provides a rapid and accurate means of detecting TB pleurisy.

Conclusion

In conclusion, it is suggested that the combined use of adenosine deaminase activity along with lymphocyte neutrophil ratio would provide a more efficient means for diagnosing tuberculous pleuritis than the use of ADA alone.

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