DIAGNOSTIC SIGNIFICANCE OF ADENOSINE DEAMINASE IN PLEURAL TUBERCULOSIS

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Background: Tuberculosis (TB) is a major cause of pleural effusion, which in TB usually has lymphocytic and exudative characteristics. Analysis of adenosine deaminase (ADA) activity is a very useful diagnostic approach to achieve a more rapid and precise diagnosis in cases of Pleural TB (pTB). **Methods**: Fifty male and fifty female patients presenting with tuberculous pleural effusion was included in the study. The patients were taken from the medical ward of Sir Ganga Ram Hospital between September 2001 and September 2002. Activity of Adenosine Deaminase (ADA) was estimated by the technique of Sodium dodecyl sulphate electrophoresis (SDS-EF) using 10% polyacrylamide gel. **Results:** Mean age of males was 45.72±19.22 years and of female was 43.74±16.09 years. Mean protein level was 3.39±0.24 g/dl in males, and it was 3.02±0.26 g/dl in females. Mean specific gravity both in males and females was 1.020±0.01. The results show an increased level of enzyme ADA in patients as compared to normal subjects. **Conclusion:** Estimation of ADA activity may provide basis for rapid and efficient diagnosis of pleural TB in different clinical settings. However study should be extended to larger number of patients to reach a better conclusion.

Keywords: Adenosine deaminase, Pleural TB, Electrophoresis

INTRODUCTION

TB is no longer the scourge it once was, but it remains an important cause of morbidity and mortality worldwide. Recent estimates are that 8–10 million new tuberculosis (TB) cases occur each year in the world and 2–3 million die. In developing countries, TB is one of the common opportunistic infections in people who are seropositive for HIV.[1] Fuelled by increasing poverty, homelessness, immigration, drug abuse, declining prevention programs, and the HIV epidemic, its incidence in the developing and developed countries has increased dramatically. Strict distinction between 'adult' and 'childhood' patterns of TB should be avoided.[2]

Pleural effusion develops because of excessive filtration or defective absorption of accumulated fluid. Effusion may be a primary manifestation or a secondary complication of many disorders. Tuberculosis (TB) is a major cause of pleural effusion, which in TB usually has lymphocytic and exudative characteristics.[3] Exudates are due to pleural inflammation (pleurisy), with an increased permeability of the pleural surface to proteinaceous fluid. Inflammation or injury increases pleural membrane permeability to proteins and various types of cells and leads to the formation of exudative effusion. In general, exudates have protein concentration higher than 3 g/dL or a specific gravity of 1.020 on a refractometer.[4,5] Lymphatic obstruction may also contribute to accumulation of pleural fluid.[6]

Diagnosis can be made in a majority of patients from the clinical features, pleural fluid examination (including cytology, biochemistry and bacteriology), and pleural biopsy. Adenosine deaminase estimation in pleural fluid is occasionally useful.[3]

Adenosine Deaminase (ADA) is an endogenous tissue enzyme which is released into the serum in patients with many different types of malignancies and infections, including viral hepatitis, infectious mononucleosis, typhoid fever, and tuberculosis. In pleural fluid, elevated ADA levels are very commonly associated with tuberculosis. In CSF, ADA is elevated in cases of tuberculous meningitis.[7,8] It is reported that it is a pleural fluid marker for tuberculosis. The analysis of PCR and ADA activity, however, is a very useful diagnostic approach to achieve a more rapid and precise diagnosis in the cases of pleural TB (pTB).[8] It is found that Pleural fluid ADA levels in TB effusions were significantly higher than the non-TB effusions.[9] The molecular forms of ADA were studied[10] that in pleural effusions using the technique of SDS-PAGE. Analysis of the Adenosine deaminase in pleural fluid pinpoints many pulmonary and systemic diseases are known to cause pleural effusions.

Present study tried to find out the diagnostic significance of Adenosine deaminase in pleural tuberculosis using the technique of gel electrophoresis.

MATERIALS AND METHOD

We studied ADA level in pleural fluid of 50 male and 50 female presenting to Sir Ganga Ram Hospital, Lahore with tuberculous pleural effusion between September 2001 and September 2002. Determination of ADA activity was carried out by using the

technique of Sodium dodecyl sulphate electrophoresis (SDS-EF) using 10% polyacrylamide gel. Comparisons between groups were done using student's *t*-test.

RESULTS

The age of the subjects was in the range of 42–46 years. Mean age of males was 45.72±19.22 years and of female was 43.74±16.09 years. Mean protein level was 3.39±0.24 g/dl in males, and it was 3.02±0.26 g/dl in females. Mean specific gravity both in males and females was 1.020±0.01. By comparing the level of fluid protein with the standard level of normal subjects it is observed that although the level of fluid protein having a lower level in patients but this shows no significant difference (Table-1).

Table-1: Age, level of pleural fluid protein and specific gravity in patients

Parameters	Male (n=50)	Female (n=50)
Age (years)	45.72±19.22	43.74±16.09
Protein (gm/dl)	3.39±0.24	3.02±0.26
Sp. Gravity	1.020±0.01	1.020±0.01

Level of Adenosine deaminase was determined by applying the sample of both patients and of normal subjects on polyacrylamide gel (for gel electrophoresis). It is observed that a 44 KDa band with density 0.0372 was observed in male patient. On the other hand, in normal control subjects the same band having a density of 0.0159 was observed. This shows an increased level of enzyme ADA in patients as compared to normal subjects (Figure-1).

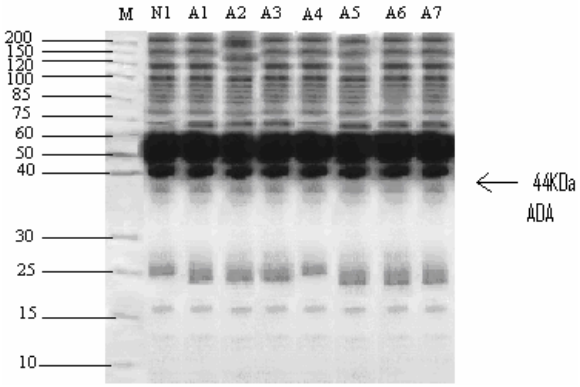

Figure-1: Serum protein profile of patients and control subjects

N= normal, A= patients, M= markers

DISCUSSION

The combination of PCR and ADA activity determination allowed the selective increase of sensitivity and specificity for probable and confirmed cases compared to individual methods. Positive and negative predictive values for these individual or combined methods were maintained over a wide range of prevalence of pleural TB in the patient population presenting with pleural effusions.[11]

It is observed that the fluid protein is having a low value in our subjects compared to normal standard protein (>3 g/dl). However in another study[12] the pleural fluid total protein concentration was reported between 5.1–5.5 g/dl.

Electophoretic profile of pleural fluid of male patients and normal subjects was carried out to find out the level or density of ADA. The density of 44 KDa ADA in our study was 0.0372. On the other, in the fluid of normal subjects (fluid having a protein of 5–6 gm/dl) the 44 KDa ADA have a density of 0.0159. According to a study[13], adenosine deaminase exists in its smallest molecular form (ADA-S) of <42 kDa in primate and rodent brain, intestine and liver, human erythrocytes, avian liver and in bovine spleen and intestine. The enzyme is a monomeric protein of molecular weight 44,000.[14] It is highlighted that pleural cells become activated and produce cytokines as a response to mycobacteria. Intra-macrophage and direct cytotoxic elimination of mycobacteria, granuloma formation, and fibrosis are the main facets of this reaction. It has been shown that the most useful diagnostic test is the level of adenosine deaminase in the pleural fluid. Elevation of these compounds in lymphocytic pleural effusions is virtually diagnostic of tuberculous pleurisy.[15]

CONCLUSION

Estimation of ADA activity may provide the basis for rapid and efficient diagnosis of pleural TB in different clinical settings. However, study should be carried out on large number of patients to reach a better conclusion.

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