Simultaneous Measurements of Adenosine Deaminase Activity and Tuberculostearic Acid in Pleural Effusions for the Diagnosis of Tuberculous Pleuritis

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Adenosine deaminase (ADA) activity and tuberculostearic acid (TSA) levels in pleural effusions were measured in 18 patients with active tuberculous pleuritis, 16 patients suspected of having tuberculous pleuritis, 14 patients with carcinomatous pleuritis, and 19 patients suffering from pleuritis of non-malignant and non-tuberculous etiology. In the patients with active tuberculous pleuritis, ADA was elevated in 56% and TSA was positive in 78%. In 83% of these patients, either ADA was elevated or TSA was positive. ADA was elevated together with a positive TSA in 50%. In contrast, TSA was positive in only 6% and ADA was elevated in 24% of the patients with non-tuberculous pleuritis, and none of these patients showed the combination of an elevation of ADA and a positive TSA. These results suggest that simultaneous measurements of both ADA and TSA in pleural effusions are useful for the diagnosis of tuberculous pleuritis.

Introduction

Pleuritis is one of the most commonly seen thoracic diseases. From 10–30% of the cases are thought to be caused by infection with Mycobacterium tuberculosis, although there are regional differences in its incidence (1, 2). Several methods are now employed for the diagnosis of tuberculous pleuritis. These include smear and culture of pleural effusions, pleural biopsies and measurement of adenosine deaminase (ADA) activity in pleural effusions (1–8). ADA is considered to be released from activated T lymphocytes (4, 6) and is often elevated in tuberculous pleural effusions (4–8). However, its elevation is not confined to tuberculous pleuritis, and it is also observed in many other diseases (5–8).

Tuberculostearic acid (TSA) is one of the lipid components derived from Mycobacterium tuberculosis (9) and is known to be a characteristic 10-methyloctadecanoic acid in the limited species of the order Actinomycetale (10, 11). Recent studies indicate that TSA is selectively detected in clinical samples from patients with tuberculosis but not in those from patients suffering from other diseases (10–16). The sensitivity of TSA detection for the diagnosis of pulmonary tuberculosis is reported to be better than that of smear examination, but slightly less sensitive than that of culture examination for tubercle bacilli in sputum specimens (13). We have reported similar results (15, 16). However, the measurement of TSA in pleural effusions has never been reported except in our report (16).

In the present study, we measured ADA and TSA in pleural effusions and found that simultaneous measurements are valuable for the diagnosis of tuberculous pleuritis.

Materials and Methods

Subject: Pleural effusions were collected from 18 patients with tuberculous pleuritis, 16 suspected of having tuberculous pleuritis, 14 with carcinomatous pleuritis, 6 with parapneumonic pleuritis, 5 with congestive heart failure, 3 with collagen diseases, and 5 with miscellaneous diseases, all of whom were admitted to Kyushu University Hospital between 1985 and 1989. The diagnosis of tuberculous pleuritis was made by the presence of granulomatous pleuritis on histopathology and/or the demonstration of acid-fast bacilli on acid-fast stain of pleural effusions or pleural biopsy specimens. The diagnosis of other diseases was confirmed by clinical and radiographic findings.

Other methods: Adenosine deaminase (ADA) activity was measured by a method of Marinetti et al. (14) and tuberculostearic acid (TSA) was measured by the method of Shigematsu et al. (16). Pleural effusions were collected at the time of diagnosis or during the course of disease. ADA activity and TSA were measured in all the patients.

Key words: adenosine deaminase activity, tuberculostearic acid, tuberculous pleuritis
culous pleuritis was made based on the positive findings of either smear examination, culture of pleural effusions or pleural biopsy. Patients suspected to be suffering from tuberculous pleuritis included those whose clinical manifestations, including responsiveness to anti-tuberculous drugs, strongly supported the diagnosis, although the laboratory examinations were not supportive of such a diagnosis. Patients in this group, clinically were considered not to have other causes which could induce pleuritis. Control patients with non-tuberculous pleuritis were separated into two groups, carcinomatous pleuritis and others, in consideration of the severity of diseases. Measurement of ADA: ADA was measured by Giusti’s method (17). We placed the cut-off point for positive levels at 50 international u/l (37°C), in accordance with previous reports (5, 7).

Measurement of TSA
TSA analysis was performed by Larsson’s method with some slight modifications (10, 11, 15) and was measured semi-quantitatively. The details of the methods were previously reported (16). Gas chromatography/mass spectrometry analysis was carried out using an EMD-05A (ESCO, Denshi-Kagaku Co., Tokyo, Japan) attached to a 3 m glass tubular column (4 mm i.d.) packed with Chromosorb WAW DMCS 80/100 mesh coated with 2% OV–17 (Gascho-Kogyo Co., Tokyo, Japan).

Statistical analysis
The results were compared according to $\chi^2$-verification.

Results
As shown in Fig. 1, ADA was elevated in 10 (56%) and TSA was detected in 14 (78%) of 18 patients with tuberculous pleuritis. In 83% of these patients, either ADA was elevated or TSA was positive, and in 9 patients (50%) ADA was elevated together with positive TSA. On the other hand, both low ADA and negative TSA were found in only three patients. Thirteen of 18 patients with tuberculous pleuritis were diagnosed by a positive pleural biopsy. In the other 5 patients, cultures of their pleural effusions were positive and ADA was higher than 50IU/l (37°C). In all three groups of measurements, ADA, TSA, ADA and/or TSA, positive rates in patients with tuberculous pleuritis were significantly higher than those in control patients statistically (Table 1).

In patients suspected to have tuberculous pleuritis, ADA and TSA were each positive in 38%. On the other hand, TSA was detected in only 2 and ADA was elevated in only 8 of 33 patients with pleuritis due to etiologies other than infection with Mycobacterium tuberculosis.

![Fig. 1. The results of measurements of ADA and TSA in pleural effusions. A closed circle refers to a TSA positive case and an open circle refers to a TSA negative case.](image-url)
The sensitivity and specificity of ADA measurements alone for the diagnosis of tuberculous pleuritis were 56% and 76%, respectively, and those of TSA measurements alone were 78% and 94%, respectively. However, with simultaneous measurements of ADA and TSA, these were 83% and 76%, respectively (Table 1). There was no statistically significant difference in the sensitivity among ADA, TSA, and ADA and/or TSA (p > 0.1). Regarding specificity, TSA analysis showed a statistically significant difference from the other measurements (p < 0.05).

Discussion

It is sometimes difficult to make an appropriate diagnosis of tuberculous pleuritis in the patient who shows negative findings in smear and culture of pleural effusions and pleural biopsies. A tuberculin reaction does not give significant information for a diagnosis of tuberculosis in the Japanese population because of the great prevalence of BCG vaccination (1-3). The diagnostic rate for tuberculous pleuritis has been reported to be 20-30% by smear and culture of pleural effusions (1-3, 7) and 40-80% by pleural biopsies (1-3, 7), which indicates that many patients must be diagnosed by a favorable response to administration of antituberculous agents (1, 2).

Adenosine deaminase (ADA) activity in pleural effusions is now known to be a useful diagnostic marker for tuberculous pleuritis. ADA is reported to be elevated in many patients with tuberculous pleuritis (4-8). However, the present results show a much lower sensitivity of ADA for the diagnosis of tuberculous pleuritis than the results reported by previous investigators (4-8). One of the reasons for the lesser sensitivity in our results may be that most of our specimens were collected from patients diagnosed only by a positive pleural biopsy. In fact, ADA measured in pleural effusions collected from patients with positive cultures was always greater than 50 IU/l (37°C). Recently, some investigators have reported that ADA is not of great diagnostic value for tuberculous pleuritis (18, 19). In our data, two patients suffering from tuberculous pleuritis showed an ADA levels of much lower than 50 IU/l (37°C). One was an 82-year-old man with hypoproteinemia and the other suffered from chronic renal failure and was on hemodialysis therapy.

The present findings indicate that ADA is less specific than TSA for the diagnosis of tuberculous pleuritis (Table 1). Elevation of ADA is not confined to tuberculous pleuritis alone, but is also observed in many other diseases (5-8). The underlying diseases of patients who show an ADA elevation of more than 50 IU/l (37°C) were bronchogenic carcinoma, adult T cell leukemia, ovarian cancer, pneumonia, aspergillosis, collagen vascular disease, and empyema. Measurement of ADA is also considered to be strongly affected by contamination with hemolysed blood (8), the effect of which may be partially responsible in the present findings.

Tuberculostearic acid (TSA) is one of the lipid components of Mycobacterium tuberculosis (9) and is characteristic of a limited species of the order Actinomycetales (10, 11). Measurements of TSA in the sputum and spinal fluid have proved to be useful for the diagnosis of pulmonary tuberculosis (10, 11, 13, 15) and tuberculous meningitis (12, 14). The present findings also show that the sensitivity of TSA measurement for the diagnosis of pulmonary tuberculosis is as good as that of culture examinations (16).

In the present study, we performed simultaneous measurements of ADA and TSA in pleural effusions. Measurement of TSA in pleural effusions has been reported only from our laboratory (16). Our results indicate that tuberculous pleuritis can be highly suspected when pleural fluid ADA and TSA are both positive, and that tuberculous pleuritis can be almost ruled out when both ADA and TSA are negative. As we, clinically experience many patients who are diagnosed as tuberculous pleuritis by the response to anti-tuberculous drugs because of no definite findings for the diagnosis, we measured ADA and TSA in pleural effusions of such patients. The data showed a higher positive rate than that of control patients (p < 0.01), suggesting that simultaneous measurements of ADA and TSA are potentially useful for clinical application as a rapid diagnostic method for tuberculous pleuritis (Fig. 1). We could not verify that simultaneous measurements of ADA and TSA were more useful than the measurement of ADA or TSA alone. However, because many patients with an ADA of lower than 50 IU/l (37°C)
have positive TSA (Fig. 1), and because the specificity of TSA analysis is significantly higher than that of ADA in tuberculous pleuritis (Table 1), it is suggested that simultaneous measurements are complementary to the measurement of ADA alone for the diagnosis of tuberculous pleuritis.

Acknowledgments: The authors are grateful to Miss Hisako Tanabe for her technical assistance and to Miss Eriko Minami for her assistance with preparation of the data.

References


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