ORIGINAL ARTICLE

Rapid Diagnosis of Active Tuberculosis by Lipoarabinomannan (LAM) Test

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Abstract

Presence of antimicrobial antibodies were repidly detected in 47 out of 50 cases of active pulmonary and extra-pulmonary tuberculosis. The lipoarabinomanan (LAM) antigen binds with the optimum concentration of anti LAM antibodies from the serum. Our findings showed that the LAM test is simple, low cost, rapid and reliable test for detecting active tuberculosis.

Key Words

Active tuberculosis, LAM test.

Introduction

Tuberculosis (TB) has been a cause of significant morbidity and mortality for mankind throughout history (l). There are 20 million cases of TB worldwide with 8 million new cases every year, out of which 3 million annual deaths are due to tuberculosis (2,3). In addition there has been a dramatic increase over the last 5 years in the incidence of tuberculosis in fast developing countries. This increase can be attributed to increased incidence of Acquired Immunodeficiency syndrome AIDS (3), and other primary risk factors like poverty, malnutrition and low socioeconomic status (5). Tuberculosis is caused by the bacterium Mycobacterium-tuberculosis, and spread by bacteria present in aerosols generated by coughing. Currently the diagnosis of pulmonary tuberculosis is based largely on the microscopic detection of causative organism in patient's sputum, but this method of detection is error prone and has got only 20-50% sensitivity. Culture of M. tuberculosis on solid media is significant but much more expensive and results are seldom obtained before 4-6 weeks. Immunodiagnostic products like ELISA and PCR are also expensive and tedious. The Mantoux test, raised ESR are non specific and chest x-ray findings are inadequate (4). Also, the new problem of HIV infected TB patients has created a complication in the detection of TB by x-ray, since HIV infection can change the classical appearance of TB in chest x-rays (5,6). In addition an appreciable number of all TB cases are extrapulmonary for which diagnosis is made on the basis of clinical presentation and detection of bacilli in biopsy specimen. The treatment of TB can reduce the rate of mortality and morbidity if active state of TB infection

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can be diagnosed. Hence the present study was taken up with the aim of rapid diagnosis of active state of tubercular infection and also to evaluate the efficacy of the LAM test.

Material and Methods

Fifty cases of clinically active tubercular infection were sent to our laboratory for LAM test. The patients were (a) Pulmonary tuberculosis (25 cases-15 M;10 F), (b) Extrapulmonary tuberculosis (25 cases-15 M;10 F) within the age of 25-35 years, and with no previous episode of tubercular infection. Ten normal healthy (5 M;5 F) individuals with age of 25-35 years were included as control group.

Active antimicrobial antibodies were evaluated by the Dynagen's. Mycodot LAM test (kit supplied by Span Diagnostic Ltd. India).

Test Procedure

All the reagents and patients' sera were brought to room temperature. The concentrated wash buffer was diluted 1:5 with distilled water. The number of teeth required (sample and controls) were determined. Three drops (0.15 ml) of sample diluent were added in each numbered microtest wells. In the next row of micro test well four drops of (0.2 ml) of colloidal gold reagent was added. In each microtest well containing sample diluent 50-µl of patients' serum was incorporated. LAM antigen coated comb was inserted into respective wells and kept for incubation for 6 minutes at room temperature. The antigen coated comb was washed 8-10 times by moving the comb forward and backward in the washing solution. The comb was reinserted into microtest well containing colloidal gold signal reagent and incubated for 10 minutes at room temperature, and was again washed with washing buffer. The comb was allowed to be air dried and checked for the development of color spot on the edge of the teeth. The result for intensity of colored spot was interpreted by comparing with the intensity of positive control sera and supplied standared reference comb.

Interpretation of Results

No spot at all or very less intense spot than the weakest positive spot on test comb was considered as negative reaction. A positive reaction generated by the Mycodot test was recorded as an intense colored spot developed on the teeth of comb.

Results

Antimicrobial antibodies to Lipoarobinomannan and lipomannan of M.tuberculosis were detected in all the cases, out of 25 cases of pulmonary tuberculosis 20 cases-80% (12M and 8F) were found to be strongly positive (intense color spot), Sera of two patients (8%) developed less intensive positive color spot and 3 patients were noted negative, since no color spot developed at all. Whereas all the twenty-five (100%) (15 M and 10 F) extrapulmonary tuberculosis patients were found to be strongly positive. LAM test was negative in all the ten normal healthy control subjects.

Discussion

Tuberculosis (TB) causes millions of deaths (2,3) every year, and a serious problem of illness in developing and developed countries. Suppression of cell mediated immunity by HIV infection and exacerbration of tubercular infection in the immune compromised host (3) attributed to increased rate of mortality. Hence early diagnosis of disease is utmost requirement of the both patients and clinicians, since TB is curable with the use of drugs, if diagnosis is followed by appropriate intervention. The LAM test was noted as simple reliable, with a positivity of 100% in extrapulmonary and 88% in

pulmonary TB infection. Our results are comparable to the results of Sada *et. al.*, who have shown LAM test to be 85% sensitive and 96% specific (7). Moreover the test offers low cost, single visit, no exposure to x-rays, no error prone results of AFB smear, and no time consuming methods of culture of *M. tuberculosis* on solid media. The test has also been found negative in healthy or BCG vaccinated individuals (8). For these reasons, a test capable of detecting active TB irrespective of its focus of infection in the body would be a valuable addition to effective and efficient diagnosis, and LAM Mycodot test meets this need.

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