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Diagnosis of Tuberculosis : Experience with Use of an Immunochromatographic Assay in a Nigerian Hospital.

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***Aim*: To evaluate an immunochromatographic assay method in the serodiagnosis of tuberculosis.**

***Patients and Methods:* Thirty participants, 25 patients and five normal individuals were studied between July and October 2001. 40μL of whole blood or 20μL of serum was used in the test for each kit. Using sputum smear microscopy as gold standard, the new method was compared with x-ray diagnosis**

***Results*: The sensitivity, specificity and efficiency of the new immunodiagnostic method were 89.5%, 77.7% and 85.7% while that of X-ray were 75%, 50% and 66.6% respectively.**

***Conclusion:* The new immunodiagnostic method gave better results in sensitivity, specificity (as well as in other performance indices) than X-ray diagnosis. The new test is rapid, reliable and field operable.**

Keywords Tuberculosis, Nigeria, Immunodiagnosis

INTRODUCTION

It is estimated that 2000 million persons world wide may be suffering from pulmonary tuberculosis (TB) caused by the bacterium *Mycobacterium tuberculosis* [1]. The TB problems have been exacerbated by two unheralded events .One is emergence of strains of the bacterium resistant to major tuberculostatic agents [2,3]. The other is the surge of the HIV/AIDS pandemic. [4] These two events have catapulted TB as one of the leading causes of death even in developing societies [3] Diagnosis of TB and monitoring patients receiving anti-tuberculosis drugs are pivotal to control of the disease. The gold standard for tuberculosis diagnosis is isolation from culture of *M. tuberculosis* .This procedure is time consuming, expensive and requires well trained personnel which is not readily available in developing countries. Sputum smear microscopy is the main recommended and used method for TB diagnosis in developing countries. X-ray is used to diagnose chest infections in general and is not specific for TB . Again, this diagnostic technology is not readily available under field conditions in the developing world.

Some culture filtrate antigens from the TB bacterium have been shown to evoke strong immune response and thus possess good immuno-diagnostic potential for tuberculosis [5,6] .This procedure , consequently tackles the problems of diagnosis or misdiagnosis especial in children who cannot express sputum, a situation that adversely affect control of tuberculosis even with multiple drug therapy and hence the public health of the community. Therefore, there is a compelling need to diagnose TB accurately, cheaply and rapidly . Because of difficulties and errors in diagnosis by culture , X-ray or smear microscopy for acid for bacilli (AFB), we investigated the applicability of immuno diagnosis of TB, using venous or capillary blood (finger prick) with a qualitative immuno chromatographic assay utilizing indirect solid-phase antigen immunoassay technology called smart check TB (SCTB) Kit. In the absence of TB culture facility , we have compared the utility of this assay with X-ray methodology in the diagnosis of TB, using AFB smear microscopy as our "Gold standard". To our knowledge , this is the first time the test is used in this part of the World .

PATIENTS AND METHODS

Between July and October 2001, 25 patients attending chest and other clinics at the University of Nigeria Teaching Hospital (UNTH) Enugu and five normal individuals, were investigated with SCTB kit. The various diagnostic units (immunodiagnosis , radiology and microscopy) worked independent of each other not knowing which case was positive or negative in other tests. The following informed consenting categories of 30 individuals were studied (see table 1).

Tables 1 categories of cases studied

Cases	No
X-ray +ve Sputum-ve	10
X-ray –ve Sputum +ve	10
Patients tread with multiple drugs	5
Normal Individuals	5

In the test, 40μl of blood or 20μl of serum or plasma was added to the upper part of samples window of the kit . Then 100μl of the developer solution was added to the lower part of the sample window. The tests were read within two to five minutes as recommended for positive, negative or invalid results . Positive result is indicated by the presence of two distinct bands in the kit, negative if there is only one band and invalid if there is no band at all. In reporting smear microscopy, 300 fields were visualized before reporting as follows.[7]

No of acid fast bacilli	Result
None in 300 fields	Negative
1-10 in 100 fields	+
1-10 in 10 fields ++	
1-10 per field	+++
> 10 per field	++++

The data from the 30 participants were analyzed statistically with Epi info version 6 software of the computer. The following statistical performance indices recognized for assessing diagnostic test were obtained for the SCTB and X-ray diagnosis. These are, sensitivity, specificity , efficiency, positive predictive value, percentage reduction of misdiagnosis and overall measure of reliability. Using (a) as true positive ,(b) as false positive , (c) for false negative and (d) for true negative in the data , the statistical indices were calculated thus: Sensitivity =a/a+c, specificity =d/b+d, efficiency =a+d/a+b+d+c, positive predicative value =a/a+b, negative predictive value =d/c+d, percentage reduction of misdiagnosis =100x(d-b)/[(a+c)+(a+b)] all as percentages, and overall measure of reliability =[(axd)-(bxc)]/[(a+c)(a+b)].

RESULTS

Table 2 summarizes the results . The sensitivity , specificity and efficiency for SCTB are 89.5% , 77.7% and 85.7% respectively.

Table 2. Performance indices of SCTB test and X-ray diagnosis compared to smear positive microscopy.

PERFORMANCE INDICES	X-RAY	SCTB TEST
Sensitivity	75%	89.5%
Specificity	50%	77.7%
Efficiency	66.6%	85.7%
Positive predictive value	75%	89.5%
Negative predictive value	50%	77.7%
Overall measure of reliability	0.13%	0.23%
Percentage reduction of misdiagnosis	0%	13.2%

DISCUSSION

The result shows that SCTB has a sensitivity of 89.5%, while X-ray has 75%. SCTB is more specific in diagnosing TB and also an enviable efficiency of 85.7% when compared to X-ray methodology of 66.6%. It is appropriate for more tests to be conducted in other parts of Africa with SCTB to ascertain its suitability as a diagnostic tool for TB.

We recognize the limitations of sputum smear examination, as it cannot specifically identify the ATB as pathogene . Therefore a definitive diagnosis of TB requires culture and speciation of the organism . However TB can be open, with patients actively excreting *M. tuberculosis* or abacillary (Pauci bacillary) in whom the currently available methods are unable to directly demonstrate the bacilli in the sputum by smear or culture.[8] Other direct and better diagnostic method include radiometric cultivation and use of nucleic acid probes to identify the species.[9] It is hard to find centres in sub-Saharan Africa with facilities for culture, and more difficult to get radiometric culture and nucleic acid probe technology. Hence indirect methods of diagnosis of TB applying immunodiagnosis like SCTB has great promise for the future. SCTB shows good prospects in the diagnosis of tuberculosis. Its use does not require highly qualified technical staff or wide observer variation and interpretation of radiographs as in X-ray diagnosis .[10] Nigeria has one of the poorest medical facilities in the world,[11] and is in the zone with a high incidence of TB [12] . We are of the opinion that a rapid, reliable and field operable method of diagnosis such as SCTB is extremely desirable.

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