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RESEARCH ARTICLE

Ziehl-Neelsen (ZN) Stained Method: Presence and Absence of Acid Fast Bacilli (AFB) of Pulmonary and Non Pulmonary Tuberculosis Patients Under Went Anti-Tuberculosis Treatment

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ABSTRACT:
Tuberculosis continues to be a major health problem worldwide despite the fact that the causative agent was discovered more than 100 years ago. The impact of tuberculosis (TB) can be devastating, especially in developing countries suffering from high burden increasing every year. A prospective study was conducted at MGM Hospitals, Warangal. Between November 2012 to October 2013, Among 116 suspected smear positive pulmonary TB patients (52%), Fourteen cases were non pulmonary tuberculosis; therefore a total of 42 smear-negative cases were enrolled in this study. The diagnosis of suspected TB cases was based on national guidelines for the microscopic test of TB . Spot-morning-spot sputum samples from symptomatic patients were collected in standardized sputum containers and all the sputum tests were done using light microscope examination of Ziehl-Neelsen (ZN) stained smears at the point of care for the presence and absence of acid fast bacilli (AFB).Patients with positive sputum smears were placed on anti-tuberculosis treatment as recommended by Revised National Tuberculosis Control Programme (RNTCP). The results reveals that, 64 (55%) males and 52 (45%) females. Of these, 64(55%) were identified based on Chest X-ray abnormalities, 52 (45%) were based on persistent Cough. All participants of the survey were first screened by symptoms and Chest X-ray. Those who had symptoms of consistent cough or haemoptysis, or Chest X-ray abnormalities were then screened by smear and culture. In study 32% Patients had smoking tendency and 68% patients were found to be non-smokers. Chi square test shows that there was significant difference in success rate between smokers and nonsmokers patients (X2=9.384, df =1,CI=95%, p value =0.002. Our article concludes that, newly diagnostic technology can accurately detect TB in patients and improve the outcome and appropriate counseling for individual can help to eradicate the tuberculosis from the universe.

KEYWORDS: India, Rural, Urban, Tuberculosis and Microbacterium

INTRODUCTION:

Tuberculosis (TB) is an airborne infection and therefore difficult to control globally. It is an infectious disease and must be to aid control and prevention. TB is caused by mycobacteria. The mycobacteria known to cause TB in humans are *Mycobacterium tuberculosis*, *Mycobacterium bovis* and *Mycobacterium africanum*. As they are closely related they are regarded as one species, collectively termed the *M. tuberculosis* complex (TB complex) [1]. TB can affect any part of the body. If it

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affects multiple sites of the body, including the lungs, it is termed miliary TB [2]. TB affecting other organs but not the lungs is termed extra-pulmonary. The current methods used in the diagnosis of tuberculosis (TB) have not greatly impacted on the diagnostic process in the paediatric population and this has resulted into delayed diagnosis or misdiagnosis and subsequent over or under treatment. [3-5] Eventhough sputum culture is considered the gold standard diagnostic tool for PTB, it is only able to detect approximately 30-40% of cases with probable tuberculosis in children [6,7]. While smear microscopy is inexpensive, simple to perform, and has a quick turnaround time, the proportion of positive samples among children is minimal due to the low bacillary burden observed in childhood tuberculosis. Newer tools such as the Nucleic Acid Amplification Tests have a slightly higher sensitivity of 40 -60%. This study was aimed to determine outcomes of tuberculosis at DOT therapy.

MATERIALS AND METHODS:

Study Site

Study design, specimen and population This survey was conducted in November 2012 to October 2013 at MGM Hospital located at Warangal, India. Consecutive patients with enlarged lymph nodes who were not responding to a two-week course of broad spectrum antibiotics and who were clinically and cytologically diagnosed as TB lymphadenitis were included in this study. Pyogenic abscesses were excluded based on the clinical and cytomorphological features.. Among 116 suspected smear positive pulmonary TB patients (52%), Fourteen cases were non pulmonary tuberculosis; therefore a total of 42 smear-negative cases were enrolled in this study. The diagnosis of suspected TB cases was based on national guidelines for the microscopic test of TB [8]. Spot-morning-spot sputum samples from symptomatic patients were collected in standardized sputum containers and all the sputum tests were done using light microscope examination of Ziehl-Neelsen (ZN) stained smears at the point of care for the presence and absence of acid fast bacilli (AFB).Patients with positive sputum smears were placed on anti-tuberculosis treatment as recommended by Revised National Tuberculosis Control Programme (RNTCP). If their cough did not improve after this treatment, a second cycle of three consecutive clinical samples were collected as described above and examined for AFB. Chest X-ray radiographic results of the patients, as evaluated by clinicians, were categorized into minimal/moderately advanced/far advanced/having cavities. As per the WHO and Government of India collaborative guidelines, grade of sputum-smear positivity was classified into scanty/single/double/triple positive status based on microscopic findings [9-11].

Cases were classified as new, relapse, treatment after default and failure case. New case is patient who has never had treatment for TB or has taken anti- TB drugs for less than one month. Treatment after default is patient, who has received treatment for TB for a month or more from any source and returns for treatment after having defaulted consecutively for two months or more and found to be smear-positive. Failure is patient who is smear-positive at 5 months or more after initiation of treatment. Relapse case is patient who was declared cured or treatment completed by a physician and who reports back to the health facility and is now found to be sputum smear positive. Patient who died during the course of treatment regardless of cause is declared as mortality case. The following information was obtained from DOTS Centers: age, sex, weight, type of case, category of treatment, other medical illness, and cause of death. Data was entered into Microsoft Office excel 2007 and were analyzed using graph pad prism version 5.

RESULTS:

Of total 116 cases initiated on anti-TB treatment, out of 116 patients of pulmonary tuberculosis 102 (88%) and non pulmonary tuberculosis 14 (12%). 36(31%) patients with age between 21-30 years, 24(21%) patients with age between 31-40 years and 40 (34%) patients with age >50 years were found between November 2012 to October 2013. Rural residents had about a two times higher risk of smear –positive PTB than that of their urban counter-parts. This could be due to lack of awareness and poor housing conditions among the rural inhabitants than the urban dwellers. A study from Tigray showed that rural residents were less knowledgeable about PTB than their urban counterparts [12]. These results are in agreement with previous studies in medical literature.

Table 1. The distribution of new tuberculosis patients in Warangal by selected demographic characteristics

Patients characteristic	Age (in years)	Number of Patients	Percentage (%)
Age group	≤20	16	14
	21-30	36	31
	31-40	24	21
	41-50	20	17
	≥50	20	17
Gender	Male	64	55
	Female	52	45
Occupation	Employed	64	55
	Unemployed	22	19
	Students	30	26
Residential area	Rural	68	59
	Urban	48	41

In this study, we found that a greater number of males diagnosed with pulmonary. In this study, we found that a greater number of males diagnosed with pulmonary TB than females (55% and 45%, respectively). This similar

the assam and new jalpaiguri district (North Bengal), India.[13] Of these, 64(55%) were identified based on Chest X-ray abnormalities, 52 (45%) were based on persistent Cough. All participants of the survey were first screened by symptoms and Chest X-ray. Those who had symptoms of consistent cough or haemoptysis, or Chest X-ray abnormalities were then screened by smear and culture. In study 32% Patients had smoking tendency and 68% patients were found to be non-smokers. Chisquare test shows that there was significant difference in success rate between smokers and nonsmokers patients (X^2=9.384, df =1,CI=95%, p value =0.002. However, mathematical model predicted that smoking would produce an excess of 18 million tuberculosis cases and 40 million deaths from tuberculosis between 2010 and 2050, if smoking trends continued along current trajectories. Smoking was also expected to delay the millennium development goal target to reduce tuberculosis mortality by half from 1990 to 2015[8].Therefore, efforts should be geared towards reducing tobacco use among TB patients due to its negative impact on TB treatment outcomes. Development of TB disease is a two-step process: infection by transmission and progression to active disease. The risk of infection is dependent on exogenous factors. The intimacy and duration of contact with active TB cases the degree of infectiousness of the case, and the shared environment in which the contact takes place are important risk factors [14-18].

Table 2. Clinical and microbiological characteristics of prevalent TB cases.

Microbiological characteristics	Total (n)	Culture positive (n)	Culture – negative (n)
Smear-positive	68	58	60
Smear-negative	48	42	06

N= number of patients

Most people in our study had resided in shelters or rented house, where they slept in good environments, conditions that could not enhance transmission of tuberculosis bacilli. All patients with smear positive induced sputum were treated with anti-tuberculous treatment (ATT) as per national control program. Control of TB in the community depends on early diagnosis and treatment. In regions with a high prevalence of TB, an early diagnosis is considered as one performed between 2 to 3 weeks after the onset of clinical symptoms and a late diagnosis is that performed 4 weeks after this onset [19]. Adherence to anti TB is an important factor for sputum conversion and cure and could have introduced bias in our study. In the study setting, anti-TB therapy is administered as daily direct observed therapy (DOT) throughout the duration of treatment by either a health care worker at health facility (facility DOT) or a community treatment supporter (community DOT) according to patient's preference [20]. Adherence to anti-TB in Tanzania has been shown to be as high as 99% when urine samples were tested for metabolites of the drugs among TB patients [21].

Table 3 illustrates proportion of the patients in successful and unsuccessful treatment outcome categories.

Treatment outcome	Number of TB Patients (n)	Percentage (%)
Cured	58	50
Completed	52	45
Failure	04	3.4
Defaulter	02	0.1

Out of 116 (45%) successfully treated patients, only 50% patients were classified as cured. In terms of unsuccessfully treated patients, 04 (3.4%), while 02 (0.1%) defaulted from the treatment. A relatively small proportion of the patients (n = 04) were transferred to other health care facilities. Fantahun B studied, the disease risk was high for illiterate inmates compared to those inmates with educational status above basic literacy (''above read and write''). One of the main explanations might be the low level of health care seeking behaviors in illiterate patients. Thus, health education for illiterate inmates is important to reduce TB transmission in prisons. The present study is contrast to above finding, here 65% were literate. Despite all efforts, TB remains a major global health problem. Vaccination with Bacille Calmette – Guerin (BCG) confers good protection against disseminated childhood TB but provides variable protection against pulmonary disease, especially in adolescents and adults. The duration of the BCG protection remains debatable, although there is good evidence that the declines with time[22]. Unfortunately, the traditional methods of TB diagnosis are not only time consuming but also unreliable. Sputum smear examination has a sensitivity of only 50%, whereas sputum culture can confirm pulmonary TB in around 80% of cases [23]. Although sputum smear positive case 52% were seen in our study population.

CONCLUSION:

Our article concludes that, newly diagnostic technology can accurately detect TB in patients and improve the outcome and appropriate counseling for individual can help to improve medication adherence as well as eradicate tuberculosis from the universe. In future, to create awareness about the tuberculosis complication, prevention in rural and urban community of developing countries.

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