Prevalence of MDR TB in Suspected Pulmonary Tuberculosis Patients in A Tertiary Care Hospital Kanchipuram, Chennai,Tamilnadu, India

T.Vijayalakshmi[1],Dr. Sumathi[2],Dr. Selvaraj Stephen[2], M.Kamalraj

[1]Department of Microbiology, Sri muthukumaran Medical college Hospital and Research Institute, Chennai, Tamilnadu, India.
[2]Professor& HOD Microbiology, Sri Muthukumaran Medical College Hospital and Research Institute, Chennai, Tamilnadu, India.
[3]Professor, Department of Microbiology, Mahatma Gandhi Medical College Hospital and research Institute Puducherry, India.\
[4]Department of Microbiology, Sri muthukumaran Medical college Hospital and Research Institute, Chennai, Tamilnadu, India.

Abstract: *Pulmonary tuberculosis (TB) is caused by the bacteria Mycobacterium tuberculosis (M. tuberculosis). One gets TB by breathing in air droplets from a cough or sneeze of an infected person. To study the prevalence of drug resistance in Mycobacterium tuberculosis isolated from patients with pulmonary tuberculosis. A total of 241 clinical specimens sputum samples were collected from suspected pulmonary tuberculosis patients in Tertiary care Hospital and Research institute, Kanchipuram District. All the Samples were stained by Ziehl-Neelsen (ZN) method. All sputum AFB smear positive specimens and 178 extrapulmonary clinical specimens were cultured in Lowenstein Jensen (LJ) medium. All Mycobacterial isolates were identified by standard protocols and tested by conventional drug susceptibility method using absolute concentration method.. Out of 241 clinically suspected pulmonary tuberculosis patients, 61 (22.50%) patients were smear positive for AFB. Among these 34 (55.73%) were positive for Mycobacterium tuberculosis culture on LJ medium. Those isolates were tested for first line anti-TB drugs by conventional drug susceptibility method. Two (7.14%) M.tuberculosis isolates were resistant to rifampicin, isoniazid. Two multidrug resistant tuberculosis (MDR - TB) strains were isolated from pulmonary tuberculosis patients. The overall MDR TB percentage is reported in this hospital, is 7.14 percentage of MDR - TB isolates were reported in culture positive PTB cases.*
Keywords: *Mycobacterium tuberculosis, absolute concentration method, first line anti-TB drugs, MDR TB*

I. Introduction

Pulmonary Tuberculosis (TB) is a contagious bacterial infection that involves the lungs, but may spread to other organs**.** Pulmonary tuberculosis (TB) is caused by the bacteria *Mycobacterium tuberculosis (M. tuberculosis)*. One gets TB by breathing in air droplets from a cough or sneeze of an infected person. In the United States, most people recover from primary tuberculosis infection without further evidence of the disease. However, in some people it can get reactivated. Most people who develop symptoms of a tuberculosis infection first became infected in the past. However, in some cases, the disease may become active within weeks after the primary infection. Multi drug resistance in *Mycobacterium tuberculosis* (MTB) is a serious threat in developing countries [1]. The emergence of drug resistant tuberculosis (TB) control and is a major public health concern in several countries.

Ethiopia is one of the high TB and HIV burdened countries in sub-Saharan Africa which ranks 7th among the 22 high TB burden countries in the world (2). In 2006, an estimated 500,000 individuals throughout the world developed multidrug-resistant (MDR) TB, which, at a minimum, is refractory to treatment with isoniazid and rifampicin, the 2 firstline antibiotics for treating TB [3,4].The average prevalence of multi drug resistant tuberculosis (MDR-TB) in new cases of tuberculosis was 1.1% (range 0-14.2%). Among the previously treated cases median prevalence of resistance to any drug was 33.4 % (range 0- 93.8%). High levels of resistance have been reported in certain regions of the world, particularly in Asia and parts of Africa [5-11].

India is contributing to nearly one third of the world's tuberculosis (TB) cases and has the highest rate of new cases [12]. Prevalence of MDR-TB cases is on the rise in India, being about 1.1% to 5.3%. The mean average of MDR TB among previously treated patients varied from 8% to 67% [13] MDR-TB most commonly develops due to inappropriate treatment, or patients missing doses or failing to complete their treatment [14]. Drug resistance in mycobacteria is defined as a decrease in sensitivity to a drug to be reasonably certain that the strain concerned is different from a sample of wild strains of human type that have never come in contact with

the drugs [15]. For both guidance of therapy and surveillance of drug résistance, accurate drug susceptibility testing (DST) for *Mycobacterium tuberculosis* complex is considered highly important [16]. In 2006, an estimated 500,000 individuals throughout the world developed multidrug-resistant (MDR) TB, which, at a minimum, is refractory to treatment with isoniazid and rifampicin, the 2 firstline antibiotics for treating TB. [17,18] The present study was undertaken to find the prevalence of drug resistance in *Mycobacterium tuberculosis* isolated from sputum AFB smear positive pulmonary tuberculosis patients in a teaching Hospital of Kanchipuram district, Tamilnadu, South India.

II.   Materials And Methods
A total of 241 clinical specimens sputum samples were collected from suspected pulmonary tuberculosis patients in a tertiary care hospital, Kanchipuram district, Tamilnadu, South India. Out of 241 clinically suspected pulmonary tuberculosis patients, 61 (22.50%) patients were smear positive for AFB were categorized according to the Revised National Tuberculosis Control Programme (RNTCP) guidelines such as; those who had never received anti-TB drugs previously, patients who had history of treatment for pulmonary tuberculosis in the past and patients with history of treatment for pulmonary tuberculosis for more than one month during illness. The study was approved by the Institutional Ethical Committee (IEC) of the Mahatma Medical College Hospital and Research Centre Pondicherry and informed consent was obtained from each patient.

Sample Collection
According to the WHO guidelines [19] two consecutive sputum samples namely one "spot" and another one "early morning" sputum sample were collected from 241 patients who had clinical evidence of pulmonary tuberculosis (PTB). specimens collected in sterile universal container.

Specimens Processing
All pulmonary specimens were processed and stained by Ziehl-Neelsen (Z-N) method and examined for Acid Fast Bacilli (AFB). Direct AFB smear positive sputum samples and pus samples were processed for culture by Petroff`s (4% NaOH) method. The samples were centrifuged and the deposit was decontaminated with 5 percent H2SO4. Fine needle aspiration (FNA) samples were directly inoculated on to a pair of L-J medium. Once growth appeared, it was tested by Ziehl-Neelsen (Z-N) staining for Acid-Fast Bacilli (AFB). Mycobacterial species identified by standard conventional protocol [20] such as; based on slow growth rate, absence of pigmentation, Niacin test positivity and absence of growth on L-J medium with ρ-nitrobenzoic acid, semi-quantitative Catalase test and Catalase test at 68°C /PH 7.0.

Culture For Primary Isolation And Drug Susceptibility For *M. Tuberculosiss.*
Preparation of Lowenstein-Jensen (L-J) Medium: Lowenstein-Jensen (L-J) medium was prepared for primary isolation of M. tuberculosis from clinical specimens. Approximately 15 ml of medium was poured into sterilized 25 ml Bijou (McCartney) Bottle, secured with sterilized cap and kept at 85°C in slanting position for 45 min. Once the medium solidified, it was kept at 115°C for 10 min, cooled, labeled and stored at 2-8°C. Every batch of L-J medium was checked for quality control before specimen inoculation.

III.   Results And Discussion
Among 241 clinically suspected pulmonary tuberculosis patients**,** 61 (22.50%) patients were smear positive for AFB.among which 43(70.49%) were male and 18 (29.51%) were female. Maximum number of patients belonged to the age group of >60 (Table 1) out of 61 AFB smear positive 34 specimens yielded mycobacterial growth. All 34 M. tuberculosis strains were slow growers, positive for niacin test, failed to grow on PNB containing L-J media and negative for Catalase test at 68°C /PH 7.0. They were weakly positive for semi-quantitative Catalase test. In this study, one NTM isolate was a slow grower, produced visible colonies on PNB containing media on 21st day, Catalase positive, niacin negative and was reported to be photochromogens.

Table 1. Age group distribution of sputum AFB smear positive pulmonary tuberculosis patients (N = 61)

S.no	Age group	No.of. Male	No.of.Female
1	18-40	12	4
2	41-60	14	11
3	>60	17	3
4	Total	43	18

In this study revealed resistance to anti Tb drugs from 34 culture positive samples Out of 61 sputum AFB positive newcases 17 were *mycobacterium tuberculosis* culture positive of these 2 were MDR TB

IV.   Conclusion

AFB staining by Ziehl-Neelsen (ZN) method helps in Diagnosis of Pulmonary tuberculosis. Early screening of patients and drug susceptibility test of culture positive and MTB cases,will   help in initiating treatment of MDR TB.This will help in preventing spread in the community. In this study two patients had MDR TB and this high lights the necessity for performing drug sensitivity test. This will ensure success of the RNTCP programme.

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