ORIGINAL RESEARCH

Evaluation of pattern of drug resistance in sputum positive smear cases of pulmonary tuberculosis

[1]Dr. Manmeet Pratap Singh, [2]Dr. Mohammad Vasil Mannan

[1,2]Assistant Professor, Department of Chest and Tb, National Capital Region Institute of Medical Sciences College, Nalpur, Uttar Pradesh, India

Corresponding author
Dr. Mohammad Vasil Mannan
Assistant Professor, Department of Chest and Tb, National Capital Region Institute of Medical Sciences College, Nalpur, Uttar Pradesh, India

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ABSTRACT

Background: The present study was undertaken for assessing pattern of drug resistance in sputum positive smear cases of pulmonary tuberculosis. **Materials & methods:** A total of 50 patients were included in this study. Informed consent was taken for getting the detailed history including previous anti tuberculosis treatment, for performing systemic physical examination and for required investigations. Two Sputum samples from each patient were collected in sterile containers and immediately sent for AFB smear microscopy. All sputum smears positive cases with previous history of treatment with first line antituberculosis drugs were instructed to collect sputum sample in a sterile, leak proof falcon tube. The samples were processed for Line Probe Assay. **Results:** H: Sensitive, R: Sensitive [isoniazid (H) and rifampicin (R)] was found to be present in 48 percent of the patients, while R: Sensitive, H: Resistant was found to be present in 24 percent of the patients. H: Sensitive, R: Resistant was found to be present in 8 percent of the patients while H: Sensitive, R: Sensitive was found to be present in 20 percent of the patients. Non-significant results were obtained while assessing the correlation of radiographic findings and H & R sensitivity. **Conclusion:** Line probe assay is a rapid and accurate tool for identification of drug resistance in pulmonary tuberculosis patients.

Key words: Tuberculosis, Resistance, Pulmonary

INTRODUCTION

Mycobacterium tuberculosis (MTB) is the causative agent of tuberculosis (TB), an ancient human disease which mainly affects the lungs and hence pulmonary tuberculosis is the most common presentation. Evidence of TB has been found in human thousands of years ago and there have been references to this ancient scourge.[1, 2]

The Mycobacterium tuberculosis possesses a distinct complex cell wall. It contains long chain fatty acids called mycolic acid which provides a strong lipid barrier and essential for viability of mycobacteria.In the past few decades, there has been a collaborative global effort to eradicate TB. Pulmonary tuberculosis (PTB) refers to any bacteriologically confirmed or clinically diagnosed case of TB involving the lung parenchyma or the tracheobronchial tree. A patient with both pulmonary and extrapulmonary TB should be classified as a case of PTB. Extrapulmonary tuberculosis (EPTB) refers to any bacteriologically confirmed or clinically diagnosed case of TB involving organs other than the lungs, e.g. pleura, lymph nodes, abdomen, genitourinary tract, skin, joints and bones, meninges.[3, 4]

Treatment completed is a TB patient who completed treatment without evidence of failure BUT with no record to show that sputum smear or culture results in the last month of treatment and on at least one previous occasion were negative, either because tests were not done or because results are unavailable.A patient is confirmed to have drug resistant TB, only when the results are from a RNTCP quality-assured Culture & DST Laboratory and by a RNTCP-endorsed testing method. Drug-resistant tuberculosis (MDR-TB) has emerged as an important global health concern and is on the rise throughout the world.The emergence of multidrug and extensively drug-resistant tuberculosis (MDR-TB and XDR-TB, respectively) is a major threat to global tuberculosis control. Line probe assays (LPAs) are rapid molecular diagnostics that can detect M. tuberculosis and drug resistance.[4, 5]

Hence; under the light of above-mentioned data, the

present study was undertaken for assessingpattern of drug resistance in sputum positive smear cases of pulmonary tuberculosis.

MATERIALS & METHODS

The present study was undertaken for assessing pattern of drug resistance in sputum positive smear cases of pulmonary tuberculosis. Patients with potential symptoms suggestive of pulmonary tuberculosis and history of treatment with antituberculosis drugs reported to Chest and T.B department were enrolled. The study was conducted on sputum smear positive previously treated pulmonary TB patients and follow up sputum smear positive new pulmonary TB patients. A total of 50 patients were included in this study. Informed consent was taken for getting the detailed history including previous anti tuberculosis treatment, for performing systemic physical examination and for required investigations. Two Sputum samples from each patient were collected in sterile containers and immediately sent for AFB smear microscopy. All sputum smear positive cases with previous history of treatment with first line antituberculosis drugs were instructed to collect sputum sample in a sterile, leak proof falcon tube. The samples were processed for Line Probe Assay. All the data were compiled in Microsoft excel sheet and were analysed by SPSS software.

RESULTS

Mean age of the patients of the present study was 41.76 years. 45 percent of the patients belonged to the age group of 31 to 50 years. 68 percent of the patients were males while the remaining 32 percent were females. Fever and cough was found to be present in 92 and 96 percent of the patients. Shortness of breath and weight loss was found to be present in 44 percent and 60 percent of the patients. H: Sensitive, R: Sensitive was found to be present in 48 percent of the patients, while R: Sensitive, H: Resistant was found to be present in 24 percent of the patients. H: Sensitive, R: Resistant was found to be present in 8 percent of the patients while H: Sensitive, R: Sensitive was found to be present in 20 percent of the patients.Non-significant results were obtained while assessing the correlation of radiographic findings and H & R sensitivity.

Table 1: Clinical symptoms

Clinical symptoms	Number of patients	Percentage of patients
Fever	46	92
Cough	48	96
Shortness of breath	22	44
Hemoptysis	2	4
Chest pain	4	8
Weight loss	30	60
Night sweats	15	30

Table 2: Pattern of H and R resistance

H and R pattern	Number of patients	Percentage	p- value
H: Sensitive R: Sensitive	24	48	0.001 (Significant)
H: Resistant R: Sensitive	12	24	
H: Sensitive R: Resistant	4	8	
H: Resistant R: Resistant	10	20	
Total	50	100	

DISCUSSION

Tuberculosis is caused by infection of lung with small aerobic non-motile bacillus Mycobacterium tuberculosis (MTB). It spreads through the air when people who have an active MTB infection cough, sneeze, or otherwise transmit their saliva through the air.Multidrug-resistant tuberculosis (MDR-TB) is defined as disease due to Mycobacterium tuberculosis that is resistant to isoniazid (H) and rifampicin (R) with or without resistance to other drugs. Rifampicin-resistant TB (RR-TB) defined as resistance to rifampicin detected using genotypic or phenotypic methods with or without resistance to other first-line anti-TB drugs.[6-9]Hence; the present study was undertaken for assessing the pattern of isoniazid and rifampicin resistance in pulmonary tuberculosis patients using line probe assay.

Mean age of the patients of the present study was 41.76 years. 45 percent of the patients belonged to the age group of 31 to 50 years. 68 percent of the patients were males while the remaining 32 percent were females. Fever and cough was found to be present in 92 and 96 percent of the patients. Shortness of breath and weight loss was found to be present in 44 percent

and 60 percent of the patients. H: Sensitive, R: Sensitive was found to be present in 48 percent of the patients, while R: Sensitive, H: Resistant was found to be present in 24 percent of the patients. Tripathi R et al (2017) conducted a study to assess multidrug-resistant tuberculosis detection and characterization of mutations in mycobacterium tuberculosis by genotype MTBDRplus. The detection of rpoB gene mutations for rifampicin (RIF) and katG and inhA genes for isoniazid (INH), respectively, was performed on 663 samples by LPA. A total of 663 sputum samples from MDR suspects were received of which 321 (50.8%) were found to be MDR. Missing of WT8 along with mutation in codon S531 L was the most common pattern for RIF-resistant isolates (80.8%) and missing WT along with mutation in codon S315T1 of k atG gene was the most common pattern for INH-resistant isolates (91.3%).The MDR-TB in Eastern Uttar Pradesh, India, was found to be 50.8%.[10]

In the present study, H: Sensitive, R: Resistant was found to be present in 8 percent of the patients while H: Sensitive, R: Sensitive was found to be present in 20 percent of the patients. Non-significant results were obtained while assessing the correlation of radiographic findings and H & R sensitivity.Bekele S et al assessed the prevalence of drug resistance with the line probe assay GenoType MTBDRplus in a set of 161 M. tuberculosis strains that were selected from four common lineages and sub-lineages previously identified in Ethiopia. Most of the tested M. tuberculosis isolates had been genotyped by established Spoligotyping and MIRU-VNTR typing methods. The proportion of MDR-TB among the isolates was 3.1%. Mono-resistance was 1.2% to rifampicin and 4.3% to isoniazid, and resistance to either of the two first line drugs was 8.7%. Strains of Lineage 4 had the highest resistance rate (13.6%) followed by Lineage 3 (4.9%). None of the isolates representing Lineages 1 and Lineage 7 were drug resistant. Multidrug resistance among pulmonary TB and TB lymphadenitis clinical isolates was 2.8 and 3.7%, respectively. Drug resistance of strains carrying the most prevalent spoligotype in Ethiopia - SIT149 - was further explored. Stratification by MIRU-VNTR identified one genotype with a high rate of drug resistance against Rifampicin and Isoniazid and circulation of a potential MDR-TB clone is proposed. Although the strain selection was not fully randomized, the overall M. tuberculosis drug resistance rate in this strain set was 8.7% while the rate of MDR was 3.1%.[11] Sharma S et al (2018) detected multidrug resistance and extensively drug resistance among smear-negative extrapulmonary tuberculosis cases in a reference laboratory. An attempt was made to recover M. tuberculosis (MTB) from such EP specimens using rapid liquid culture (MGIT 960). Molecular Line probe assay (LPA) was used to determine the resistant for first line drugs, and second line drug resistant was determined using liquid culture. Culture positivity was found in 21.3% (133/623) of specimens; of these 95.48% (127/133) were found to be MTB and 4.51% (6/133) of specimens were found to be non-tubercular mycobacteria. Among MTB detected 18.9% of specimens were multi drug resistant, 3.90% were Rifampicin mono-resistant and 13.30% were Isoniazid resistant. Second line DST (N=29) was performed for Kanamycin and Ofloxacin; of which 3.4 % was found to be resistant to both drugs, 3.4 % was found to be resistant to Ofloxacin and 93.1% were sensitive to both drugs. Large percentage of drug resistance was observed in the study.[12]

CONCLUSION

Line probe assay is a rapid and accurate tool for identification of drug resistance in pulmonary tuberculosis patients.

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