ORIGINAL ARTICLE

# A study on prevalence of rifampicin resistance by Gene Xpert with clinico-radiological correlation in previously treated pulmonary tuberculosis patients.

**Mahendra Kumar[1*], Ananda Datta[2], Hemant Kumar[3]**

[1]MD, Professor, [2]MBBS, 3rd year PG resident, [3]MBBS, 3rd year PG resident, Department of Respiratory Medicine, R.N.T Medical College, Udaipur.

**ABSTRACT**

**BACKGROUND**: The drug resistant tuberculosis is a great threat to mankind. The rifampicin resistance is a good predictor of multidrug resistant tuberculosis. The Gene Xpert is an excellent diagnostic tool for rapid detection of *Mycobacterium tuberculosis* and rifampicin resistance. **AIMS & OBJECTIVES:** The aims & objectives of the study were to find out the prevalence of rifampicin resistance among smear positive and smear negative previously treated pulmonary tuberculosis patients and to correlate clinico-radiological profile with rifampicin resistance. **MATERIALS & METHODS:** The prospective analytic study was conducted in 200 presumptive TB patients with past history of anti-tuberculosis treatment for one month or more. The Gene Xpert test (CBNAAT) was performed. The results were analysed using appropriate statistical tests. **RESULTS:** Out of 200 study population, Mycobacterium tuberculosis was detected in 159 (79.5%) cases by Gene Xpert. Rifampicin resistance was detected in 36 (22.64%) out of 159 cases. The prevalence of rifampicin resistance among sputum positive previously treated pulmonary tuberculosis was 25.21% (31/123) whereas the prevalence among previously treated sputum negative pulmonary tuberculosis was 13.8% (5/36). Out of 57 relapse cases, 26 failure cases, 40 treatment after default cases and 36 other cases, rifampicin resistance was detected in 9 (15.79%), 12 (46.15%), 10 (25%) and 5 (13.88%) patients respectively. The statistically significant correlation was found between failure cases and rifampicin resistance ($P<0.05$). **CONCLUSION:** The overall prevalence of rifampicin resistance is 22.64% among previously treated pulmonary tuberculosis. The failure case is significantly associated with rifampicin resistance.

**Keywords:** Rifampicin resistance, Gene Xpert, MDR TB.

## INTRODUCTION

Tuberculosis (TB), a disease caused by Mycobacterium tuberculosis has affected mankind for over 5,000 years and still continues to be a leading cause of mortality and morbidity.[1] TB ranks as a leading cause of death from infectious diseases worldwide alongside human immunodeficiency virus (HIV).[2] Emergence of drug resistance has further worsened the situation and become a significant public health problem world over creating an obstacle to effective TB control. According to Global Tuberculosis report 2015, globally 3.3% of new and 20% of previously treated TB cases was tuberculosis (MDR-TB) in 2014.[2] MDR - TB is a man-made phenomenon. Poor estimated to have multi-drug resistant treatment, poor drugs and poor adherence lead to development of MDR-TB.[3] Culture is the gold standard for final determination of drug resistance but it is time consuming and may take up to 2-8 weeks. Thus rapid identification which is essential for earlier diagnosis, treatment initiation, improved patients outcomes and more effective public health interventions relies on nucleic acid amplification techniques.[4] The Gene Xpert MTB/RIF (Cartridge Based Nucleic Acid Amplification Test-CBNAAT) is an automated real time polymerase chain reaction (PCR) assay designed for the rapid and simultaneous detection of Mycobacterium tuberculosis and 'Rifampicin' resistance within 2 hours.[5,6,7] MDR-TB is defined as disease caused by mycobacterium tuberculosis which is resistant to at least 'Isoniazide' (H) and 'Rifampicin' (R) with or without other first line anti-tuberculosis drugs.

***Corresponding Author:***
Dr. Mahendra Kumar
Q. no.2, TB & Chest Hospital,
Bari,Udaipur-313001
Contact No: 8769163850
Email-Id: mkbainara@gmail.com

Resistance to rifampicin detected by genetic probes are very suggestive of MDR TB as in most instances, it co-exists with resistance to isoniazide and only <10% cases of rifampicin resistance is mono-resistance.[8] So rifampicin resistance is used as a surrogate marker for MDR-TB. The present study assessed the prevalence of rifampicin resistance in previously treated pulmonary tuberculosis patients using CBNAAT along with clinico-radiological correlation.

**AIMS & OBJECTIVES**

1. To find out the prevalence of 'Rifampicin' resistance by CBNAAT among smear positive and smear negative previously treated pulmonary tuberculosis patients.
2. To correlate the clinico-radiological profile with 'Rifampicin' resistance.

**MATERIALS & METHODS**

This was a prospective analytic study conducted in 200 presumptive TB patients with past history of anti-tuberculosis treatment (ATT) for one month or more attended the department of Respiratory Medicine, R.N.T. Medical College, Udaipur from Nov. 2014 to Oct. 2015.

**Inclusion criteria**

1. All smear positive previously treated pulmonary TB cases at diagnosis.
2. All smear negative previously treated pulmonary TB cases at diagnosis.

**Exclusion criteria**

1. Patient who are still taking ATT from any source and whose follow is positive.
2. Patients below 10 years of age.
3. Moribund and severely ill patients.
4. Patients with active hemoptysis.
5. Patient with HIV co-infection.

The Operational Research Committee of State TB division has given approval to conduct this study at our institution. We also got approval from the ethical committee of this medical college. Prior Informed and written consents were taken from the study population. The demographic profile, socioeconomic status, occupation, personal history, family history of TB, clinical history and previous ATT history of each patient were carefully assessed and documented. Thorough physical examination was performed in each patient and was documented. All patients were subjected to chest radiograph. Early morning and spot sputum samples in sterile broad mouth containers were sent for AFB examination under RNTCP. Sputum was induced by nebulization with hypertonic saline (3%) in patients who had less or no expectoration. Early morning, deep coughed sputum specimens were also sent in falcon tubes with filled Annexure-I for CBNAAT. Reports of tests were collected electronically on same day. The statistical analysis was done using Statistical Package for Social Sciences ver. 16 (SPSS-16).

**RESULTS**

Out of 200 study population, Mycobacterium tuberculosis was detected in 159(79.5%) cases by Gene Xpert. 123 (77.35%) were sputum positive and 36 (22.65%) were sputum negative. The overall prevalence of rifampicin resistance among previously treated pulmonary tuberculosis was 22.64% (36/159). Among the 36 rifampicin resistant cases, 31 (86.11%) were sputum positive and 5(13.89%) were sputum negative. The prevalence of rifampicin resistance among sputum positive previously treated pulmonary tuberculosis was 25.21% (31/123) whereas the prevalence of rifampicin resistance among previously treated sputum negative pulmonary tuberculosis was 13.8% (5/36).

**Table1: Distribution Of Cases & Rifampicin Resistance (By Sputum Microscopy & Gene Xpert).**

| Sputum microscopy result | MTB detected by Gene Xpert | | |
|---|---|---|---|
| | Rifampicin sensitive | Rifampicin resistance | Total |
| Sputum positive (n=125) | 92(74.79%) | 31(25.21%) | 123(100%) |
| Sputum negative (n=75) | 31(86.11%) | 5(13.89%) | 36(100%) |

Among the resistant cases, male to female ratio was 2.6:1 and maximum number of patients was in the age group of 31-40 years.

**Table2: Age And Sex Distribution Of Rifampicin Resistant Patients.**

| Age | Male | Female | Total |
|---|---|---|---|
| 11-20 | 2 (5.56%) | 1 (2.78%) | 3 (8.33%) |
| 21-30 | 6 (16.67%) | 3 (8.33%) | 9 (25%) |
| 31-40 | 9 (25%) | 3 (8.33%) | 12(33.34%) |

| | | | |
|---|---|---|---|
| **41-50** | 5 (13.88%) | 2 (5.56%) | 7 (19.44%) |
| **51-60** | 4 (11.11%) | 0 (0%) | 4 (11.11%) |
| **>60** | 0 (0%) | 1 (2.78%) | 1 (2.78%) |
| **Total** | 26(72.22%) | 10(27.78%) | 36 (100%) |

Out of 57 relapse cases, 26 failure cases, 40 treatment after default cases and 36 other cases, rifampicin resistance was detected in 9 (15.79%), 12 (46.15%), 10 (25%) and 5 (13.88%) patients respectively.

**Table3: Relation Of Rifampicin Resistance With Type Of Cases.**

| Type of case | Rifampicin Sensitive | Rifampicin Resistance | Total | P value |
|---|---|---|---|---|
| Relapse | 48(84.21%) | 9(15.79%) | 57(100%) | 0.139 |
| Failure | 14(53.85%) | 12(46.15%) | 26(100%) | 0.003 |
| Treatment after default | 30(75 %) | 10(25%) | 40(100%) | 0.718 |

There is statistically significant relation between failure cases and rifampicin resistance ($P<0.05$). Diabetes mellitus was the only co-morbidity found in 8.33% of resistant cases. Out of 36 resistant cases, 31 (86.11%) patients were underweight and 5 (13.89%) had normal weight. Among resistant patients, 2 (61.11%) had far advanced disease, 11 (30.55%) had moderately advanced disease and 3 (8.33%) had minimal disease in their chest radiographs.

**Table4: Relation Of Radiological Extent Of Disease With Rifampicin Result.**

| Radiological extent | Rifampicin Sensitive | Rifampicin Resistance |
|---|---|---|
| Minimal disease | 10 (8.13%) | 3 (8.33%) |
| Moderately advanced disease | 36 (29.26%) | 11 (30.55%) |
| Far advanced disease | 77 (62.6%) | 22 (61.11%) |
| **Total** | 123 (100%) | 36 (100%) |

## DISCUSSION

According to Global Tuberculosis report 2015, globally 3.3% of new and 20% of previously treated TB cases was estimated to have multi-drug resistant tuberculosis (MDR-TB) in 2014. Estimated 4,80,000 people developed MDR-TB in 2014, out of these 71,000 were reported from India. 2.2% of new cases and 15% of retreatment cases were notified to have MDR TB in India in the year 2014.[2] In our study, we found prevalence of rifampicin resistance among previously treated pulmonary tuberculosis was 22.64%. The reported prevalence of rifampicin resistance were 33.7% from New Delhi by Jain et al. in 1992[9], 37.3% from Gujarat by Trivedi et al.[10] in 1998, and 37.47% from Gujarat by Shah et al.[11] in 2002 and 28.2% by Malhotra et al.[12] in 2002 from Jaipur. Low prevalence in our study in comparison to the previous literature may be due of effective implementation of various components of RNTCP. Since drug-resistance is a dynamic phenomenon, it is important to monitor the trend of drug-resistance periodically. There is an extremely high burden of Multidrug resistant Tuberculosis among the BRICS countries which may be attributed to its demographic and socio-economic profile like poverty, lack of knowledge attitude and practice, overcrowding, malnutrition, care during illness and lack of social security. Adequate information on prevalence of MDR-TB, epidemiological factors and their interactions are essential prerequisites to redirect health resources in formulating a National Treatment policy which would control the transmission pattern of MDR-TB as well as ensure better patient management and its prevention. No single factor is fully attributable for emergence of MDR-TB.[13] Demographic characteristic analysis showed that among the resistant cases, male to female ratio was 2.6:1. Manna et al.[13] reported male to female ratio was 2.4:1. The higher rates of TB among male is due to a higher risk of exposure. Furthermore, the stigma attached to a positive diagnosis leads many women to forego seeking necessary medical attention. In the present study, maximum number of patients was in the age group of 31-40 years. Comparable results were also reported by Rasaki et al.[14] and Sharma SK et al. (2011)[15] who found age group of 31-40 years had the highest drug resistant TB. In our study, 15.79% of relapse, 46.15% of failure, 25% of treatment after default cases had rifampicin resistance. Mekonnen F et al. (2015)[16] reported 3.8% prevalence of 'R' Resistance in relapse cases and 40% prevalence in default cases using Gene Xpert. Abdulla K et al. (2015)[17] concluded that the patients with history of treatment failure or defaulter were more likely to have isolates with rifampicin resistance as seen in our study. Regarding the radiological extension of disease, out of 36

resistant patients, 8.33% had minimal disease, 30.56% had moderately advanced disease and 61.11% had far advanced disease. Cavity was the most common radiological lesion. Zahirifard et al. (2003)[20] found cavitary lesions in 80% MDR-TB patients. Abd El-Azim et al. (2003)[21] revealed that 58% of patients with MDR-TB had far advanced lesion in chest X-ray followed by minimal lesion in 26% and moderately advanced lesion in 16% of cases. In a study by Mukharjee P et al. (2015),[19] 43.02% MDR-TB patients had moderately advanced disease, 38.95% had far advanced disease and 18.02% had minimal disease in chest radiograph. They also reported cavitary lesion as the common radiographic presentation. The limited drug penetration into the cavity that harbors a large mycobacterial load and a greater number of acid fast bacilli in the moderately advanced or far advanced disease is believed to contribute to the drug resistance. Consistant observation was found in this study. In our study, we found that there was a statistically significant association between rifampicin resistance and failure cases only.

## CONCLUSION

The Gene Xpert is a novel technique that gives result in less than two hours. This can help in appropriate treatment decision on the same day especially in high TB burden countries like India to reduce the transmission of MDR-TB infection in the population. The prevalence of rifampicin resistance detected by this technique in our study is slightly higher than WHO reported data on MDR-TB. The failure cases are significantly associated with rifampicin resistance. So there must be a high index of suspicion for MDR-TB in failure cases.The sputum negative cases that have been treated previously should also be suspected for DR-TB.

## LIMITATION

As the study was limited exclusively to the patients attending our department, the results of our study were not representative of whole community. Other constraint in our study was that repeat CBNAAT was not performed in patients whose sputum microscopy and initial CBNAAT was negative due to resource limitations.

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