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A Comparative Study of Genexpert MTB/RIF Assay with Conventional AFB Smear for the Diagnosis of Suspected Cases of Pulmonary And Extrapulmonary Tuberculosis From a Tertiary Care Hospital, Greater Noida

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Abstract

Introduction: *Tuberculosis (TB) continues to pose a significant public health challenge worldwide, with high morbidity and mortality, particularly in low- and middle-income countries. Rapid and accurate detection of Mycobacterium tuberculosis (MTB) and drug resistance is essential for early treatment and control. This study aimed to evaluate the diagnostic utility of GeneXpert in comparison with Ziehl–Neelsen (ZN) smear microscopy and to assess rifampicin resistance in pulmonary and extrapulmonary samples.*

Aim and Objective: *Comparative study of GeneXpert MTB/RIF assay with conventional AFB smear for the diagnosis of suspected cases of Pulmonary and Extrapulmonary tuberculosis*

Materials and Methods: *A total of 1389 clinical samples were processed in the TB laboratory, of which 100 confirmed cases were included. Pulmonary (sputum, BAL) and extrapulmonary (pus, gastric aspirate, pleural fluid, CSF, ascitic fluid, tissue) specimens were examined. Preliminary identification was performed by ZN staining, and confirmation with rifampicin resistance detection was carried out using the GeneXpert MTB/RIF assay.*

Results: *Among the 100 patients, 60% were males and 40% females, with the majority (32%) in the 21–30 year age group. Pulmonary specimens accounted for 74% of cases, predominantly sputum (67%). Smear positivity was 92%, while GeneXpert detected MTB in 100% of cases, including 8 smear-negative samples. Rifampicin resistance was detected in 19% of cases–17.6% in pulmonary and 23.1% in extrapulmonary TB. A strong correlation was observed between smear grading and bacterial load by GeneXpert.*

Conclusion: *GeneXpert demonstrated superior sensitivity over smear microscopy and enabled rapid detection of rifampicin resistance. Its integration into routine diagnostics can enhance early case detection, guide timely treatment, and reduce TB transmission in high-burden settings.*

Keywords: *Tuberculosis ,GeneXpert MTB/RIF,Molecular diagnosis, Extrapulmonary tuberculosis*

INTRODUCTION

Tuberculosis (TB) remains one of the key infectious diseases that has plagued humankind for thousands of years and continues to claim an enormous death toll in many vulnerable populations, even in the 21st century. According to estimates, Mycobacterium tuberculosis (MTB) kills over 1.4 million people each year and infects 8.7 million new people according to the most recent WHO TB report (World Health Organisation, 2023).(1) Rapid MTB diagnosis is difficult, particularly in low- and middle-income nations, where more than 90% of TB cases occur. (2)Drug resistance has also spread widely as a result of the continuous spread of tuberculosis (3) TB is frequently a death sentence because there is no cure. By inhaling droplets containing bacteria, a vulnerable person can contract tuberculosis (TB) through the air. As a result, TB has long been thought to be inherited. (4)

Tuberculosis in lung tissue, trachea, bronchus, and pleura is referred to as pulmonary tuberculosis.**(5)** TB is known as extrapulmonary TB (EPTB) when it affects any area of the body other than the lungs.**(6)** Standard techniques for tuberculosis detection comprise microscopic smear analysis, culture, drug susceptibility testing, and chest radiography. The culture and drug sensitivity test for tuberculosis bacteria

are the gold standard for detecting tuberculosis, whereas microscopic analysis of sputum smear is an essential diagnostic technique. While a positive result in sputum smear indicates high infectiousness, its sensitivity is low.(7)

Gene Xpert is a WHO-recommended fast diagnosis tool for Mycobacterium tuberculosis that relies on molecular technologies and real-time PCR detection. High sensitivity, precision, and speed are features of this approach (results can be obtained in as little as two hours). Additionally, it is capable of identifying Mycobacterium tuberculosis even in cases where the results of a sputum smear are negative and the rifampicin resistance.(8)

Therefore, this study aims to evaluate rifampicin resistance along with Genexpert's usefulness for the quick molecular diagnosis of M.tuberculosis from clinical specimens.

MATERIALS AND METHODS

All the Pulmonary (sputum, bronchoalveolar lavage) and extrapulmonary (pus, ascitic fluid, synovial fluid, pleural fluid, cerebrospinal fluid, gastric aspirate, pus and tissue) samples were processed in the TB laboratory using standard microbiological techniques. Prelimnary Identification was done by ZN staining and confirmed by GeneXpert assay along with Rifampicin resistance detection

RESULTS

Out of 1389 Clinical samples received from various wards, a total of 100 patients were included in this study, of which 40 (40%) were females and 60 (60%) were males. Most of the patients were in 21-30 years of age.

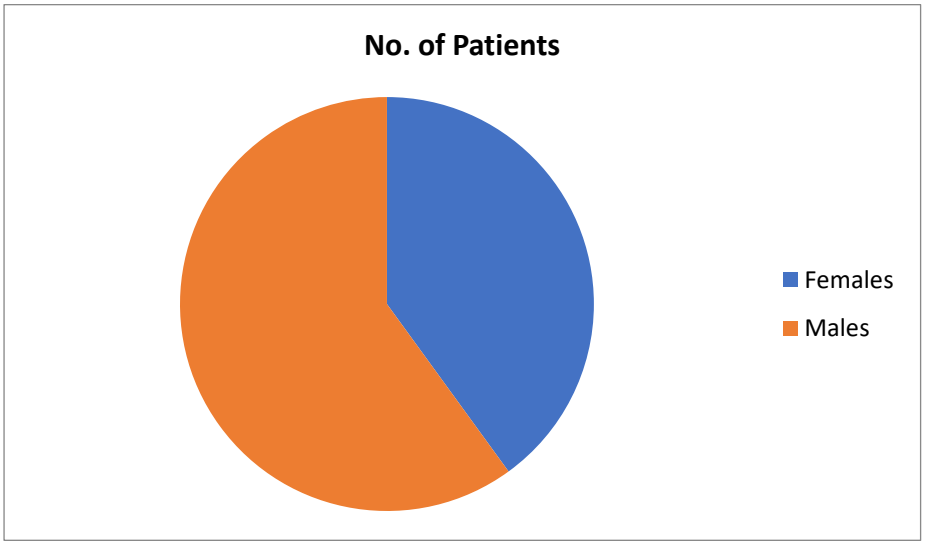
Distribution of age & gender wise are shown in table 1 and 2.

Table 1- Age wise distribution of Patients

Age	Number of patients
5-10 years	2 (2%)
11-20 years	18 (18%)
21-30 years	32(32%)
31-40 years	9 (9%)
41-50 years	12 (12%)
51-60 years	9 (3.65%)
61-70 years	7 (7%)
71-80 years	8 (8%)
81-90 years	2(2%)

Table 2- Gender wise distribution of Patients

Gender	Number of patients
Females	40(40%)
Males	60(60%)



Graph No.1: Gender wise distribution of Patients

Both pulmonary and extra-pulmonary specimens were included in this study out of which Pulmonary specimens were 74 and extrapulmonary specimens were 26 (table-3). Highest proportion was constitued by pulmonary specimensi.e of sputum 67 (67%) samples (table-4). Smear positivity was found to be 92% (table-5) .GeneXpert positivity for pulmonary remained 100% (table-6). In the case of rifampicin resistance out of 100 samples 19 (19%) were rifampicin detected . and 81 (81%) samples were not detected (table 7) . Out of 74 pulmonary TB samples, rifampicin resistance was detected in 13 cases (17.6%), while among 26 extrapulmonary TB samples, 6 cases (23.1%) were resistant(table 8)

The result of comparison between ZN staining and GeneXpert shown in table number 9. Among 8 ZN smear negative subjects were found to be positive for MTB by GeneXpert.

Table-3 Nature of the specimen distribution of Patients

Sample	Number of positive sample
Pulmonary	74 (74%)
Extrapulmonary	26 (26%)

Table -4 Sample wise distribution of Patients

Sample type	Number of positive samples
Sputum	67 (67%)
BAL	7(7%)
Pus	11 (11%)
Gastric aspiration	5(5%)
Pleural fluid	4 (4%)
CSF	2 (2%)
Tissue	2(2%)
Ascitic fluid	1 (0.73%)

Table -5 Results of Specimens on ZN smear

Positive grading	Number of positive samples
Scanty	18 (18%)
(1+)	38 (38%)
(2+)	22 (22%)
(3+)	14 (14%)

Table- 6 Sample positivity by GeneXpert

Detected rate	Number of positive samples
Very low	7 (7%)
Low	31 (31%)
Medium	22(22%)
High	40 (40%)

Table- 7 Detection of Rifampicin resistance in samples

Rifampicin resistance	Number of samples
Detected	19 (19%)
Not detected	81 (81%)

Table -8 Detection of Rifampicin resistance in pulmonary and extrapulmonary samples

Rifampicin resistance	Pulmonary samples	Extrapulmonary samples
Detected	13	6
Not detected	61	20

Table- 9 Comparison between ZN staining and GeneXpert

ZN Smear Result	GeneXpert	Total

	VL	L	M	H	
Scanty	3	13	2		18 (18%)
1+	1	13	17	7	38 (38%)
2+			2	20	22 (22%)
3+			1	13	14 (14%)
Negative	3	5			8 (8%)
Total	7	31	22	40	100

VL= Very Low, L= Low, M= Medium, H= High, P=Positive, N=Negative

DISCUSSION

To break the chain of disease transmission, TB must be diagnosed early. While most doctors focus on ZN smear-positive patients because they are thought to be extremely contagious, smear-negative patients are also known to be responsible for roughly 17% of transmission and its effects on public health.(9).The results of routine TB diagnoses using microscopy and XpertMTB were compared in this study.

Our study demonstrated a higher TB positivity rate in males (60%) compared to females (40%), with most cases recorded in individuals aged between 21-30 years (32%). These findings are consistent with the study conducted by Humayun M et al in Harare, Zimbabwe in 2022,found that males had a 53% higher risk of developing TB compared to females (41.5%) (risk ratio [RR] = 1.53; 95% confidence interval [CI], 1.12–2.09 . with most cases occurring between 25-44 years of age (64.5%), followed by the 45–64 age group (19.7%).(1

Among the 100 positive samples obtained in this study, 74% were pulmonary samplesand 24% were samples. These results are similar to the findings of Sivaraman et al , who reported in Kerala, India, the EPTB burden increased from 26% in 2010 to 40% in 2021,(11) while a U.S. study conducted by Jim E. Banta et al reported during 2014–2016 showed PTB at 71.4% and EPTB at 17.4% indicating lower proportions of EPTB compared to the results.(12)

In the current study sputum samples accounted for 67% of positive diagnostic results. The prevalence of positive results was lower for other specimen categories, including tissue (2%), BAL (7%), stomach aspiration (5%), pleural fluid (4%), CSF (2%), pus (11%), and ascitic fluid (0.73%). In contrast a study conducted by Mousa .J et al found sample positivity was more in EPTB -Tissue (35%), Pleural fluid (19%) , Pus(17%) , CSF (5%) as compared tp PTB(13)

In the present study, out of 74 pulmonary TB samples, rifampicin resistance was detected in 13 cases (17.6%), while among 26 extrapulmonary TB samples, 6 cases (23.1%) were Rifampicin resistant and 81% rifampicin-sensitive. In study conducted by Misra R et al from northern India, rifampicin resistance was notably higher in pulmonary tuberculosis cases (23.5%) compared to extrapulmonary cases (17.4%)—underscoring the differential resistance burden across TB manifestations.(14)

Our analysis reveals a clear relationship between smear grading and Mycobacterium tuberculosis bacterial load as measured by GeneXpert. Specifically, cases with 2+ and 3+ ZN smear grades align overwhelmingly with the “High” GeneXpert bacterial category (20 of 22 and 13 of 14 cases, respectively), whereas lower grades (Scanty, 1+) are distributed across lower load categories. A recent 2024 study conducted by Gota BVA et al at Kasturba Hospital, Manipal, found a strong positive correlation (polychoric $r = 0.8681$, $p < 0.05$) between GeneXpert Ultra cycle threshold (Ct) values and smear grades, reinforcing their finding that smear positivity increases in tandem with bacterial burden as detected by molecular diagnostics.(15)

Recent evidence in 2024 and 2025 continues to strengthen the role of GeneXpert in tuberculosis (TB) diagnostics. A 2024 study by Orgeur et al. highlighted the evolutionary adaptability of Mycobacterium tuberculosis, underscoring the need for rapid molecular diagnostics to combat emerging resistant strains. Similarly, Asare et al. (2024) demonstrated that GeneXpert significantly outperformed microscopy in detecting smear-negative TB, supporting its integration into routine diagnostic protocols. Most recently, Sivaraman et al. (2025) reported a rising proportion of microbiologically confirmed extrapulmonary TB

cases in India, where GeneXpert played a crucial role in accurate detection. Together, these findings from 2024 and 2025 affirm that GeneXpert not only enhances early case detection but also strengthens global TB control strategies by bridging diagnostic gaps in both pulmonary and extrapulmonary disease (16-18).

CONCLUSION

The study establishes GeneXpert as a highly sensitive and rapid diagnostic tool for Mycobacterium tuberculosis compared to conventional ZN smear microscopy. Its ability to detect MTB in smear-negative cases and simultaneously identify rifampicin resistance makes it invaluable for both pulmonary and extrapulmonary TB. With a rifampicin resistance rate of 19% observed, early detection through molecular diagnostics is critical for guiding appropriate treatment and controlling the spread of resistant strains. Integrating GeneXpert into national TB control programs can improve case detection, enable timely interventions, and ultimately contribute to reducing the TB burden in vulnerable populations**.**

DECLARATIONS

Conflicts of interest: There is no any conflict of interest associated with this study
Consent to participate: There is consent to participate.
Consent for publication: There is consent for the publication of this paper.
Authors' contributions: Author equally contributed the work.

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