

Early Diagnostic Utility of GeneXpert for the Detection of *Mycobacterium tuberculosis*. A descriptive cross-sectional study.

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ABSTRACT

Background: TB continues to be one of the commonest causes of morbidity and mortality globally, making the need for accurate diagnosis essential in order to improve patient outcomes.

Objective: To determine the accuracy of the GeneXpert MTB/RIF test in comparison with sputum smear microscopy among suspected cases of pulmonary TB.

Methods: It was a descriptive, cross-sectional study that took place within six months (January to June 2024) in THQ Hospital Jand, Attock District. The cross-sectional study was performed among 150 patients suffering from suspected pulmonary tuberculosis. Data about their demographic and clinical characteristics was taken, and all subjects had sputum smear microscopy and GeneXpert MTB/RIF test performed. Sputum smear microscopy served as the gold standard against which the results were compared in terms of their sensitivity, specificity, positive/negative predictive values, likelihood ratios, and diagnostic accuracy. All statistical analyses were carried out using the SPSS software, version 20. Correlation between continuous variables was assessed through the Pearson correlation coefficient.

Results: Participants had an average age of 36.40 ± 17.12 years, of which 74 (49.33%) were male while

76 (50.67%) were female. The majority of participants lived in rural settings (111; 74.0%) and were married (107; 71.33%), with a farming occupation being the most common (94; 62.67%). Some common signs and symptoms observed were fatigue (140; 93.33%), chest pain (139; 92.67%), unintended weight loss (137; 91.33%), and coughing out mucus or blood (117; 78.0%). The assay showed moderate diagnostic performance with strong agreement with smear microscopy.

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Conclusion: GeneXpert MTB/RIF assay offers a rapid and more sensitive alternative to smear microscopy, particularly beneficial for detecting smear-negative TB cases, thereby improving case detection in high-burden settings.

Keywords: Tuberculosis, GeneXpert MTB/RIF, smear microscopy, diagnostic accuracy

INTRODUCTION

Mycobacterium tuberculosis is still the cause of tuberculosis (TB), a chronic and dangerous public health hazard, with low-income and developing nations having the greatest rates of infection and death [1]. Formerly referred to as the "white plague," tuberculosis (TB) most often attacks the lungs (pulmonary TB), however it may also spread to other organs, resulting in extrapulmonary TB [2]. Night sweats, fever, persistent cough, and unexpected weight loss are the disease's hallmarks [3]. Since it is carried by airborne particles from infected people, early and precise diagnosis is crucial to stopping its spread [4]. In low- and middle-income countries (LMICs), where diagnostic and healthcare infrastructure is often insufficient, the worldwide burden of tuberculosis (TB) is still high [5].

Pakistan has one of the highest rates of multidrug-resistant tuberculosis (MDR-TB) in the world, with 27,000 cases reported by the WHO in 2016 [6]. Pakistan has one of the highest incidences of tuberculosis in the world. There are significant drawbacks to conventional diagnostic

methods such polymerase chain reaction (PCR), culture methods, and sputum smear microscopy [7]. Although sputum smear microscopy is commonly used and reasonably priced, it has low sensitivity and requires a large bacterial load for detection [8]. Because culture-based approaches, the gold standard, are more sensitive but require weeks to produce results, the start of therapy is postponed [9]. This delay not only negatively impacts the outcomes for certain individuals but also promotes the ongoing spread of tuberculosis.

In about two hours, *M. tuberculosis* and rifampicin resistance can be found using the GeneXpert system, an automated, cartridge-based nucleic acid amplification test (NAAT) [12]. Its excellent sensitivity in smear-negative and HIV co-infected individuals makes it a useful tool in high-burden circumstances, and its quick return time enables the early administration of appropriate therapy [13].

The effectiveness of GeneXpert in improving TB diagnosis, especially in high-burden settings, has been demonstrated in numerous investigations [14]. The WHO advises using GeneXpert as the



initial diagnostic test for individuals with suspected MDR-TB and HIV-associated TB [15]. Low-income countries like Pakistan have incorporated it into their national TB control programs, despite ongoing challenges related to cost, infrastructure, and operational constraints [16]. Despite these challenges, when utilized in specialized and remote healthcare facilities, GeneXpert may significantly enhance early TB detection and treatment. Previously, no such study on Tuberculosis has been conducted at THQ Hospital Jand, District Attock. This hospital previously lacked Gene Xpert equipment, which has been installed 2 years back by Health and Population Department, Government of Punjab. This study would help to evaluate Tuberculosis burden in Teshil Jand, District Attock.

Considering its sensitivity, specificity, and potential for reinforcing TB control efforts in endemic settings, the objective of this research is to determine the usefulness of GeneXpert in diagnosing *M. tuberculosis* among suspected pulmonary TB patients.

METHODS

Considering its sensitivity, specificity, and potential for reinforcing TB control efforts in endemic settings, the objective of this research is to determine the usefulness of GeneXpert in

diagnosing *M. tuberculosis* among suspected pulmonary TB patients.

Study Design and Setting

This was a descriptive, cross-sectional study conducted over a period of six months (January to June 2024) data was collected prospectively at THQ Hospital Jand, District Attock. The study location was pulmonology ward, where all patients admitted due to symptoms that indicated TB infection were assessed.

Inclusion and Exclusion Criteria

The inclusion criteria included adults, male or female, of any age, clinically diagnosed to have pulmonary TB infections and had been evaluated using GeneXpert MTB/RIF test and sputum smear microscopic examination. The exclusion criteria involved patients undergoing anti-TB treatment, patients without complete laboratory data, extrapulmonary TB cases, and those who had refused consent.

Sample Size

Convenience sampling technique was adopted whereby 150 people among the eligible pool of inpatient and outpatient were enrolled who had been presented with symptoms of pulmonary tuberculosis. The reasons for employing convenience sampling included conducting the research at a single center where all eligible patients within January to June 2024 had to be



recruited consecutively. Considering the nature of the research and its exploratory aspect, there could not have been any a priori statistical power and sample calculation. Nonetheless, the sample size is consistent with other previous studies carried out in the real world, assessing the diagnostic efficacy of GeneXpert for TB diagnosis [17].

Data Collection

For data collection, the first step included collecting sputum specimens from the selected subjects in accordance with the established guidelines on bio-safety. Lab results for both sputum smear microscopy and GeneXpert MTB/RIF analysis tests were documented. All the results obtained from the testing process were captured using a pre-prepared pro-forma sheet.

Statistical Analysis

SPSS version 20 software was utilized for analysis of data. For continuous variables, the mean $\pm$ standard deviation was calculated. Association between two continuous variables was established by the use of Pearson correlation coefficient. Statistical significance was assumed at $P < 0.05$. The following parameters were used to establish the diagnostic capability of GeneXpert: sensitivity, specificity, positive

likelihood ratio, negative likelihood ratio, prevalence of illness, positive predictive value, negative predictive value, and accuracy.

Ethical Approval

Approval of the study protocol was granted by the IRB of THQ Hospital Jand, District Attock. Informed consent forms were signed by each individual or his or her guardian if they were below age, in compliance with ethical guidelines regarding the conduct of human subjects research.

RESULTS

The mean age of the 150 subjects was 36.40 ± 17.12 years, with 74 (49.33%) being males and 76 (50.67%) females. Of all the subjects, the majority came from the countryside ($n=111$; 74.0%) compared to those who lived in the cities ($n=39$; 26.0%). Also, the majority were married ($n=107$; 71.33%), while the rest ($n=43$; 28.67%) were unmarried. In terms of occupation, the largest proportion were farmers ($n=94$; 62.67%).



Table 1: Demographic, clinical characteristics, and age distribution of study participants.

Variable	Category	Frequency	Percent
Gender	Male	74	49.33
	Female	76	50.67
Residence	Rural	111	74.00
	Urban	39	26.00
Marital Status	Married	107	71.33
	Single	43	28.67
Occupation	Unemployed	46	30.67
	Employed	10	6.67
	Farmer	94	62.67

Figure 1 shows the distribution of clinical signs and symptoms among the 150 study participants. Coughing up mucus or blood was reported by 117 patients (78.0%), chest pain or pain with

breathing/coughing by 139 patients (92.67%), unintentional weight loss by 137 patients (91.33%), and fatigue by 140 patients (93.33%).

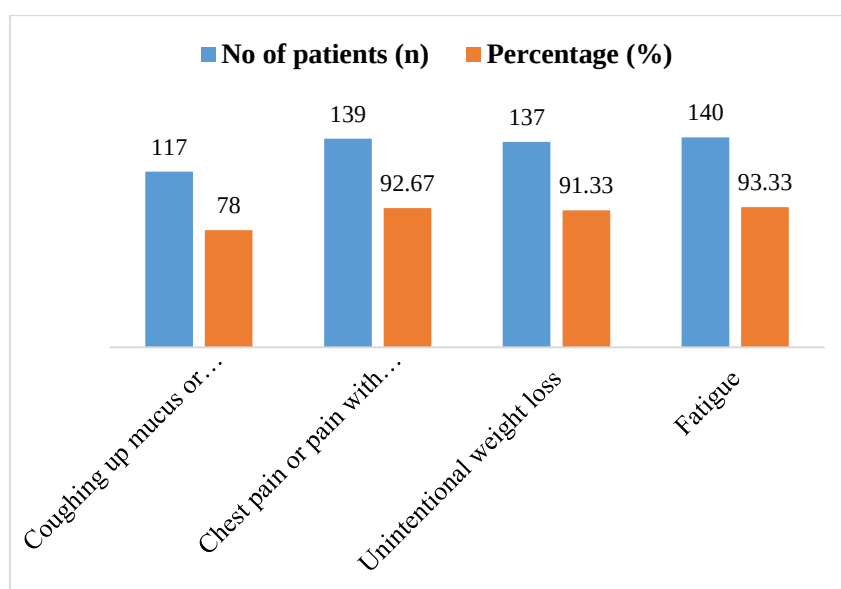
**Figure 1: Distribution of clinical signs and symptoms among study participants (n = 150)**

Figure 2 presents the cross-tabulation of smear microscopy and GeneXpert results among the

150 study participants. Of the 85 smear-positive cases, 80 were also GeneXpert positive, while 5

were GeneXpert negative. Among the 65 smear-negative cases, 43 were GeneXpert positive and 22 were GeneXpert negative. Overall,

GeneXpert detected 123 positive cases and 27 negative cases.

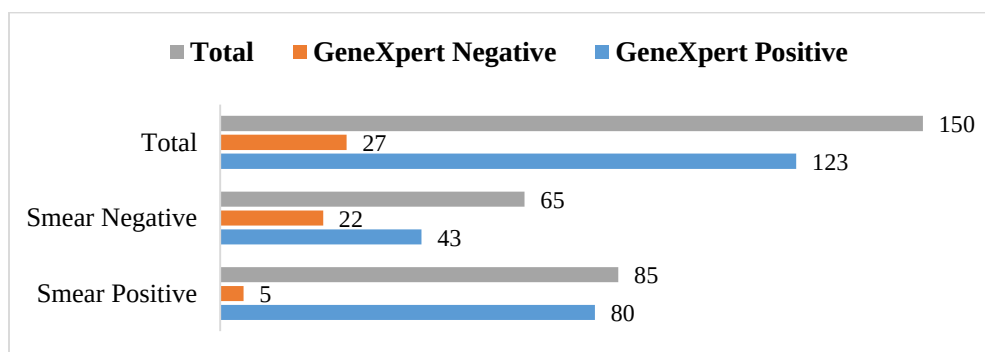


Figure 2: Cross-tabulation of smear microscopy vs GeneXpert results

The GeneXpert MTB/RIF assay's diagnostic performance is compiled in Table 2. The test showed a 70.31% (95% CI: 49.82–86.25) specificity and a 68.29% (95% CI: 59.29–76.39) sensitivity. 2.30 (95% CI: 1.27–4.17) was the positive likelihood ratio, while 0.45 (95% CI: 0.32–0.64) was the negative likelihood ratio. The study population had an 82.0% disease

prevalence (95% CI: 74.90–87.79). 91.30% (95% CI: 85.29–95.0) was the positive predictive value, while 32.76% (95% CI: 25.43–41.04) was the negative predictive value. With a Pearson correlation value of 0.46 ($p < 0.001$) and an overall accuracy of 68.67% (95% CI: 60.50–75.98), GeneXpert and smear microscopy data showed a moderately favorable association.

Table 2: Diagnostic performance of the GeneXpert MTB/RIF assay compared to sputum smear microscopy

Variables	Value	95% CI
Sensitivity	68.29%	59.29 – 76.39
Specificity	70.31%	49.82 – 86.25
Positive likelihood ratio	2.30	1.27 – 4.17
Negative likelihood ratio	0.45	0.32 – 0.64
Disease prevalence	82.0%	74.90 – 87.79
Positive predictive value	91.30%	85.29 – 95.0
Negative predictive value	32.76%	25.43 – 41.04
Accuracy	68.67%	60.50 – 75.98
Pearson correlation coefficient	0.46	$p < 0.001$

DISCUSSION

This research assessed the GeneXpert MTB/RIF assay's early diagnostic value for detecting *Mycobacterium tuberculosis* in probable pulmonary tuberculosis patients by comparing it to sputum smear microscopy. Comparing GeneXpert to smear microscopy, we found that it had an overall accuracy of 68.67%, with a sensitivity of 68.29% and a specificity of 70.31%. These findings are in line with earlier research that shown that GeneXpert had better detection rates in smear-negative individuals with a high sensitivity (88.6%) and specificity (93.6%) when compared to culture [18]. For instance, GeneXpert had a pooled sensitivity of approximately 67% and specificity of nearly 98% for smear-negative pulmonary tuberculosis, according to a meta-analysis. Similar performance in different circumstances was shown by the variability observed across different patient demographics and regional settings [19]. 43 (66.15%) of the 65 smear-negative patients in our cohort were positive for GeneXpert, proving its value in identifying additional cases that conventional microscopy would miss. This aligns with GeneXpert's WHO-endorsed role of improving TB case identification, especially in smear-negative people [18]. This type of enhancement is

particularly important in high-burden, resource-constrained settings where timely diagnosis may have a significant influence on transmission management.

Our study's excellent positive predictive value (91.30%) indicates how reliable GeneXpert is in confirming tuberculosis when the test result is positive.. Additionally, our demographic information reveals epidemiological patterns that are pertinent to TB prevention. Similar findings have been made in previous studies, thereby reinforcing the powerful positive predictive value that GeneXpert has and making it an appropriate test tool to confirm TB infection in different scenarios [20]. It must be noted, however, that a negative result in GeneXpert testing is not conclusive evidence that TB is not present in this population because of the extremely low negative predictive value of only 32.76% in this study. This limitation has been highlighted by previous research conducted, particularly in areas where there is a high prevalence rate of the disease [21].The bulk of the cases involved people engaged in farming (62.67%) and those who lived in rural settings (74%). This was in accordance with several studies that found that there are higher rates of occurrence of TB in rural areas owing to limited access to medical care and exposure to risk factors such as malnutrition and poor ventilation.



Weight loss (91.33%), chest pain (92.67%), and exhaustion (93.33%) characterized the clinical manifestation, which matched that found in other studies on TB [22].

These results largely confirm previous research which indicates that the GeneXpert technique is a valuable method for early diagnosis, particularly in cases where people are smear-negative; however, given the poor NPV and relatively low sensitivity, additional diagnostic methods should be utilized.

Strengths and Limitations

The most notable strength of this research is the practical evaluation of the performance of the GeneXpert MTB/RIF assay in the real-world setting of limited resources and a high prevalence of TB, and thus proving that the assay performs better than the gold standard of sputum smear microscopy in terms of diagnostic accuracy. Another strength includes the diversity of the study sample population. Moreover, a proper assessment of the validity of the tests is done through the study's comparison between sensitivity, specificity, predictive values, and likelihood ratios. The limitations of using a single center and small sample size ($n=150$), however, are apparent. The absence of gold standards for testing makes it difficult to validate the tests' efficiency. Similar findings have been

made in previous studies, thereby reinforcing the powerful positive predictive value that GeneXpert has and making it an appropriate test tool to confirm TB infection in different scenarios [20]. It must be noted, however, that a negative result in GeneXpert testing is not conclusive evidence that TB is not present in this population because of the extremely low negative predictive value of only 32.76% in this study. This limitation has been highlighted by previous research conducted, particularly in areas where there is a high prevalence rate of the disease [21].

CONCLUSION

The importance of this study lies in the fact that GeneXpert MTB/RIF has certain special features compared to the sputum smear test, especially the capability of detecting individuals who cannot be detected by sputum smear test. It could prove extremely helpful for diagnosing patients and providing treatment simultaneously. The introduction of GeneXpert in the management of tuberculosis, particularly those smear negatives, would make great strides in improving diagnosis and preventing disease transmission, despite its inability to completely replace other forms of diagnosis due to some limitations.



DECLARATIONS

Funding: No funding was received for this study.

Competing Interests: The authors declare that they have no conflicts of interest related to this work.

Ethical Approval and Consent to Participate: Ethical approval was obtained from the Institutional Review Board of THQ Jand , Attock, Pakistan, vide letter No. 958 dated 16/04/2024. Informed consent was obtained from all participants. Patient anonymity and confidentiality were maintained.

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Data Availability Statement: The datasets generated and analyzed during the current study are available from the corresponding author on reasonable request.

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AUTHOR CONTRIBUTIONS

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Critical revision of the manuscript for important intellectual content: Muhammad Shams ul Haq, Aleena Batool.

Final approval of the version to be published: Usman Aslam, Muhammad Yousuf, Muhammad Shams ul Haq, Aleena Batool, Syed Muhammad Asif Shah, Fatima Bashir.

All authors reviewed and approved the final manuscript and agreed to be accountable for all aspects of the work.



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