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Frequency of Sputum Acid-Fast Bacilli and GeneXpert MTB/RIF Positivity in Suspected Pulmonary Tuberculosis Patients with Radiological Evidence in a Tertiary Care Hospital

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ABSTRACT

Background: Pulmonary tuberculosis, a serious threat to the world's health, especially in areas with limited resources, remains among the most challenging infectious diseases of our century. Radiological signs often point into the direction of pulmonary tuberculosis. However, bacteriological confirmation through Sputum Acid Fast Bacilli (AFB) smear and the Gene-Expert MTB/RIF tests is essential for diagnosis, determining therapy, and ascertaining drug resistance. Evaluation of the efficacy of such diagnostic methods is necessary. **Objectives:** To determine the frequency of sputum AFB and Gene-Expert MTB/RIF positivity among patients who present to a tertiary care center with radiologically suspected pulmonary tuberculosis. <|system|>It is often very easy to get an answer to a question on the Internet. **Study design:** A cross-sectional study. **Place and duration of study.** From 01 November 2023 to 30 April 2024, Medicine Department, Sandeman Provincial Hospital / Bolan Medical Complex Hospital Quetta. **Methods:** A cross-sectional study was done at a tertiary care hospital over six months. From a total of 166 patients with radiologically suspected PTB, all patients were included in the study. Sputum samples were collected from all contributors for both AFB smear microscopy and Gene-Expert MTB/RIF testing. Demographic data collected were analyzed statistically using SPSS, where any p- value of less than 0.05 was considered significant. **Results:** Overall, the mean age of the 166 patients was 42.6 years with a standard deviation of 13.5 years. From the 92 patients with positive AFB smears, Mycobacterium tuberculosis was found in 108 by Gene-Expert MTB/RIF (65.1%). Rifampicin resistance was found in 14 patients (8.4%). Genotype MTB/RIF detection ratios exceeded AFB ratios (p = 0.003), indicating increased diagnostic sensitivity, especially for smear-negative subjects. **Conclusion:** Gene-Expert MTB/RIF performs better than the ordinary AFB smear microscopy in diagnosing PTB in patients who present with the radiological evidence, and also it facilitates the identification of rifampicin resistance. It is recommended to implement the rapid adoption as a key diagnostic method for PTB cases in high-burden settings to increase the rate of treatment initiation and reduce contagion risks.

INTRODUCTION

Tuberculosis (TB) is one of the top 10 causes of death in the world and its leading cause for death from a single infectious agent is moving ahead of HIV/AIDS. According to the World Health Organization (WHO), 10.6 million individuals were diagnosed with TB internationally in 2021, and almost 1.6 million TB patients died [1]. The most common form of pulmonary tuberculosis (PTB) primarily affects the lungs and currently is one of the major public health concerns especially in areas with high incidence of PTB and little resources. Timely diagnosis and treatment are key to containing the spread of the disease, reducing complications, and limiting drug resistance [2]. PTB diagnosis usually involves a synergistic approach of clinical assessment, imaging studies and laboratory appraisal to affirm the infection. Chest X-rays and CTs are widely used for the purposes of initial diagnosis because they can demonstrate features such as caviar lesions, infiltrates, etc. which are usual indicators for TB. These radiological signs, although suggestive, are not conclusive, and may overlap with bacterial pneumonia, lung cancer, or mycolic infections [3]. Therefore, a conclusive diagnosis entails a combination of radiographic findings and laboratory tests. A sputum smear microscopy for acid-fast bacilli (AFB) is a classic method of diagnosis of TB because of its user-friendliness and low cost [4]. However, AFB microscopy has a low sensitivity especially in cases of low bacillary load or in cases where patients are HIV co-

infected [5]. In recent years the Gene-Expert MTB/RIF assay has become a revolutionary diagnostic tool that is recommended by the World Health Organization. This molecular test is capable of detecting Mycobacterium tuberculosis DNA and Rifampicin resistance concomitantly rendering the strain to be identified as Rifampicin-resistant in a period of up to two hours Gene-Expert assay performs better than conventional smear microscopy in terms of sensitivity, particularly in smear The interpretation of PTB is very high in the gene-expert assay, which have always resulted in improved treatment results when followed [6]. Although it has some benefits, problems with accessibility and affordability can deter the widespread use of Gene- Expert in resource-limited settings, emphasizing the continued relevance of AFB microscopy. This study aims to evaluate the sputum AFB smear and the Gene-Expert MTB/RIF assay performance in screening patients for radiographic features typical of Pt. at the tertiary care hospital [7]. Informed knowledge of the relative accuracy of these tests will aid clinicians in identifying the most promising testing approach, particularly in settings of limited resources or for when additional proof is required. Also, the ability of Gene-Expert to determine rifampicin resistance is crucial in designing a functional treatment plan and the prevention of the emergence of MDR-TB [8]. Taking into account the continued incidence of TB on a global scale and the challenges it presents for diagnosis; This study focuses on contributing the relevant insights into the diagnostics of TB. By addressing the incidence of promising results for AFB smear and Gene-Expert among a clinically and radiologically suspected PTB cohort, we aim to enhance the significance of the molecular diagnostics and facilitate the introduction of combination strategy in tertiary care settings [9].

MATERIALS AND METHODS

This cross-sectional study was conducted in the pulmonology department of a tertiary hospital for six months. The individuals studied included those with clinical symptoms and radiographic manifestations of pulmonary tuberculosis. After informed consent, we collected data on the patients' demographics and clinical symptoms. Sputum samples were obtained and cultured for acid-fast bacilli (AFB) using Diehl- Nielsen staining, as well as for Mycobacterium tuberculosis using Gene-Expert MTB/RIF assay. Outcome data were recorded and analyzed to estimate the prevalence of positive AFB smears and Gene-Expert results. In case of Gene-Expert MTB/RIF positive individuals, rifampicin resistance status was recorded. Institutional review board approval was obtained prior to the inception of the study.

Inclusion Criteria

Patients aged 18 years and above who had radiological signs suggestive of pulmonary tuberculosis and symptoms such as coughing, fever, or weight loss were consented to be admitted into the study.

Exclusion Criteria

Cases of patients that were currently under treatment for Tuberculosis, patients with extra-pulmonary TB or those that refused to give sputum samples were excluded.

Data Collection

Demographic patient data, previous medical history, radiological evidence and laboratory test outcomes used standard data forms to collect information. Sputum specimens were treated using AFB smear microscopy and Gene-Expert testing at the microbiology laboratory. Clinical and laboratory results were converted into a secure electronic database for further analysis, while ensuring the privacy of the patients and adherence to ethical principles. <<

Statistical Analysis

The SPSS, version 24.0, was used to do statistical analysis on the dataset. Continuous data were presented in a form of mean plus or minus standard deviations. Categorical variables were presented in terms of their frequency and percentage distribution. We used the Chi-square test as a measure of assessing differences in proportions across groups. Statistically significant p-value was set at $p < 0.05$ for all the statistical tests used.

RESULTS

A total of 166 patients with radiological signs of pulmonary tuberculosis were enrolled into the study. The average age was 42.6 (SD = 13.5), and the study had 98 males (59%) and 68 females (41%). In the participants, 92 patients (55.4%) were sputum AFB-positive/cases that were Gene-expert MTB/RIF positive were 108 (65.1%). From the Gene-expert positive results, 14 (8.4%) had rifampicin resistance. The Gene-expert test revealed a more than two-fold increased detection rate compared with AFB smear ($p = 0.003$), reflecting its better performance. Importantly, the number of smear-negative patients, who were positively identified as Mycobacterium tuberculosis, exceeded 22 out of the patients under investigation by Gene-expert, highlighting its efficacy in identifying cases that are not visible on AFB smears. On the other hand, only 6 patients showed AFB positivity but Gene-expert negative and these might be due to non-viable or degraded DNA. These results highlight the increased sensitivity of Gene-expert MTB/RIF compared to the traditional smear microscopy, particularly in early or paucibacillary infections. Further, the ability of Gene-expert MTB/RIF in detecting rifampicin resistance highlights its critical role in drug-resistant TB. The age, gender, and positivity rates of the tests had no statistically significant relationship.

Figure 1

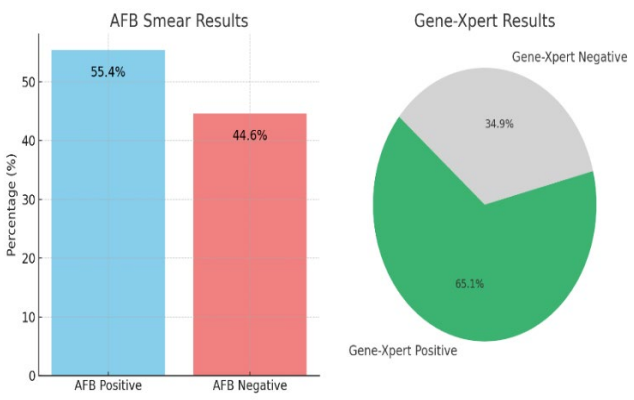

Table 1
Demographic Characteristics of Study Participants (n = 166)

Variable	Frequency (n)	Percentage (%)
Age (years)		
Mean ± SD	42.6 ± 13.5	-
Gender		
Male	98	59.0%
Female	68	41.0%

Table 2
Diagnostic Results of AFB Smear and Gene-Expert MTB/RIF

Diagnostic Test	Positive (n)	Percentage (%)	Negative (n)	Percentage (%)
AFB Smear	92	55.4%	74	44.6%
Gene-Expert MTB/RIF	108	65.1%	58	34.9%
Rifampicin Resistance (within Gene-Expert Positives)	14	8.4% of total	-	-

Table 3
Cross-tabulation of AFB and Gene-Expert Results

	Gene-Expert Positive	Gene-Expert Negative	Total
AFB Positive	86	6	92
AFB Negative			

DISCUSSION

This study involved sputum acid-fast bacillus (AFB) smear microscopy and Gene-Expert Mycobacterium tuberculosis (MTB) and rifampicin (RIF) testing to determine the diagnostic outcome of patients presenting with radiological signs suggestive of pulmonary tuberculosis (PTB) in a tertiary care facility Our findings indicate that Gene-Expert method reported higher positivity rate (65.1%) compared to AFB smear microscopy (55.4%) which highlights the enhanced sensitivity of molecular-based tests especially the ability to detect smear-negative cases. Our findings are consistent with other studies which emphasize the increased sensitivity and rapid results offered by the Gene-Expert assay. Boehme et al presentation showed that Gene-Expert detected 98.2% of smear positive and 72.5% smear negative TB cases, proving that its sensitivity has exceeded traditional microscopy particularly in the case of individuals with low bacterial load [10]. Pandey et al had also reported that among the clinically suspected cases of PTB, 61.2% were Gene-Expert positive, while only 43.8% were smear-positive, corroborating our findings [11]. In addition, our findings indicated that Gene-Expert was able to identify 22 smear-negative cases as positive a fact that demonstrates its utility in identifying early or lightly bacillary TB cases that could be missed by smear microscopy. This is in line with the work of Lawn et al. who stated that Gene-Expert is excellent at detecting the smear-negative and HIV-positive patients an area that the conventional methods are not so good [12]. In our study, 8.4% of the Gene-Expert positive cases were rifampicin resistant, which emphasizes the necessity of early detection to provide proper second-line treatment. Zeta et al. and Theron et al. have reported similar resistance rates as well, thereby lending credence to the importance of the test to contain drug-resistant tuberculosis [13,14]. Smear microscopy remains popular due to its low cost and easy manipulation though its reduced sensitivity in early or extra pulmonary TB necessitates further testing's. In the case of limited resources, it is possible to achieve greater diagnostic accuracy, as per the WHO guidelines [15], by applying both approaches simultaneously. However, WHO now recommends the Gene-Expert test as the first line for diagnosing patients reported to have MDR-TB or HIV related TB due to its high sensitivity and specificity [16]. Gender and age did not significantly influence test positivity in our study, albeit similar to the finding of Sharma et al., who emphasized that clinical and microbiological variables prevail in TB diagnosis compared to demographic factors [17]. Our results show that 59% of the subjects is male, which corresponds to global patterns where men suffer from TB more often compared to women due to their increased exposure and delay in seeking health care [18]. One of the major limitations of this study is that it was conducted in a single center with no HIV-status information, which may affect generalizability of these findings to other centers. Our findings are in line with the growing evidence that supports the use of Gene-Expert MTB/RIF as the top diagnostic-choice in suspected pulmonary tuberculosis cases whereby timely and accurate diagnosis is a necessity. The prospect of future multicenter study should embrace HIV co-infection assessment, culture-based confirmation, and cost-effectiveness studies to refine the diagnostic algorithms in high-burden areas [19,20].

CONCLUSION

The Gene-Expert MTB/RIF assay has a higher level of diagnostic sensitivity in comparison to conventional AFB smear microscopy for TB patients with radiologic evidence of PTB. Rifampicin resistance detection ability substantially enhances the clinical value of Gene-Expert MTB/RIF. Utilization of Gene-Expert as the first diagnosis tool can help to achieve earlier diagnosis, early treatment initiation, and reduction in the spread of drug-resistant tuberculosis in high prevalence areas.

Limitations

The study's use of a single center and lack of HIV status data could have influenced test sensitivity. Additionally, regular culture-based diagnostics, the gold standard, were excluded from the study. Such limitations diminish the opportunity for generalization of findings and complete assessment of diagnostic accuracy in different patient populations.

Future Directions

Further work needs to explore multi-center designs in the use of a variety of patient groups especially co-infected HIV. Integrating culture-based diagnostics and examining cost-effectiveness are crucial steps in improving integration protocols. Examination of the success of Gene-Expert in detecting extra pulmonary and pediatric TB may facilitate its usage within the clinical setting.

Abbreviations

PTB: Pulmonary Tuberculosis
AFB: Acid-Fast Bacilli
MTB/RIF: Mycobacterium Tuberculosis / Rifampicin Resistance

TB: Tuberculosis
WHO: World Health Organization
HIV: Human Immunodeficiency Virus
MDR-TB: Multidrug-Resistant Tuberculosis
CT: Computed Tomography
SPSS: Statistical Package for the Social Sciences

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