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Original Research Article

Diagnostic Performance of Genexpert MTB/RIF for Early Detection of Pulmonary Tuberculosis

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Abstract: *Tuberculosis (TB) is one of the key health challenges in the world, particularly in low- and middle-income countries. Conventional diagnostic tools, such as microscopy of smears, are low in sensitivity, whereas culture tools, though sensitive, are slow. The GeneXpert MTB/RIF assay is a rapid method of detection; however, its local diagnostic capabilities require evaluation.* ***Objective:*** *To determine the diagnostic quality of the GeneXpert MTB/RIF test versus sputum smear microscopy for early Diagnosis of pulmonary tuberculosis in DG Khan.* ***Methodology:*** *The study was a cross-sectional descriptive study conducted over a six-month period (January 1, 2025, to June 30, 2025) at the Medicine and Pulmonology departments of Allama Iqbal Teaching Hospital, Dera Ghazi Khan. One hundred ninety clinically suspected pulmonary TB patients were recruited. Sputum samples were analyzed using both smear microscopy and GeneXpert MTB/RIF. Diagnostic performance indices, including sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and overall accuracy, were calculated using culture results as the reference standard. Data were analyzed with SPSS version 25.* ***Results:*** *Of the 190 patients, the mean age was 37.2 ± 16.8 years, with 51.6% males and 48.4% females. The majority were from rural areas (72.6%) and farmers by occupation (62.1%). GeneXpert detected 156 positive cases compared with 108 by smear microscopy, including 54 additional smear-negative but GeneXpert-positive cases. Overall, the assay's accuracy was 71.6%, with a sensitivity of 71.2%, a specificity of 72.0%, a PPV of 91.8%, and an NPV of 35.5%. A moderate correlation was observed between smear microscopy results and the study's findings ($r = 0.48$, $p < 0.001$).* ***Conclusion:*** *GeneXpert MTB/RIF demonstrated a superior diagnostic yield compared to smear microscopy, particularly in smear-negative cases, making it a valuable tool for enhancing TB detection in high-burden settings.*

Keywords: Tuberculosis, GeneXpert MTB/RIF, Pulmonary TB, Diagnostic accuracy, Pakistan, Smear microscopy

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Introduction

TB, a chronic infectious illness that continues to be a major worldwide health concern, is mostly caused by Mycobacterium tuberculosis. In low-income and developing nations, the incidence and mortality rates of TB are disproportionately high (1). TB, sometimes known as the "white plague," mostly affects the lungs (pulmonary TB), though it can also spread to other organs, leading to extrapulmonary TB (2). Frequent coughing, fever, nocturnal sweats, and inadvertent weight loss are some of the classic clinical symptoms (3). Since airborne droplets from infected people are the means of transmission, prompt and precise diagnosis is essential to limit the spread of the disease (4). In many low- and middle-income countries (LMICs), however, diagnostic limitations and weak healthcare systems sustain the heavy burden of TB (5).

The World Health Organization (WHO) reported that 27,000 cases of multidrug-resistant TB (MDR-TB) were reported in Pakistan in 2016, placing the country among high-burden nations (6). Several common diagnostic methods, including polymerase chain reaction (PCR), culture, and sputum smear microscopy, have certain limitations (7). Although sputum smear microscopy is inexpensive and widely used, it has low sensitivity and requires a high bacillary load to detect Mycobacterium Tuberculosis (8). Culture-based methods, which are often viewed as the Gold standard, offer greater accuracy but are time-consuming and can take weeks to produce results (9). Such a diagnostic delay not only aggravates the outcomes of the patients but also contributes to the continued transmission within the community (10).

Molecular diagnostics, including the GeneXpert MTB/RIF assay, have been used to address these challenges. They were introduced as useful tools for detecting TB and testing for drug resistance (11). The GeneXpert system, an automated cartridge-based nucleic acid amplification test (NAAT), can quickly identify M. tuberculosis and rifampicin resistance (12).

Its quick turnaround time and great sensitivity, particularly in smear-negative and HIV co-infected patients, make it ideal for application in high-burden areas (13).

Several studies have shown the effectiveness of GeneXpert in strengthening TB diagnostic capacity worldwide (14). In fact, the WHO recommends GeneXpert as the initial diagnostic tool for suspected MDR-TB and TB/HIV cases (15). Despite the associated challenges of cost, infrastructure, and technical support, many low-income countries, including Pakistan, have integrated GeneXpert into their national TB control strategies (16). Nonetheless, there remains a need to further evaluate its diagnostic performance in local clinical settings, particularly in resource-constrained environments. Therefore, this study aims to assess the diagnostic accuracy of GeneXpert MTB/RIF for early detection of pulmonary TB in DG Khan, highlighting its role in bridging diagnostic gaps and supporting timely disease management.

Methodology

Study Design and Setting: This was a descriptive cross-sectional study conducted over six months from January 1, 2025, to June 30, 2025, at the Medicine and Pulmonology departments of Allama Iqbal Teaching Hospital, Dera Ghazi Khan. Patients presenting with respiratory disorders suggestive of pulmonary tuberculosis were automatically evaluated and examined.

Inclusion and Exclusion Criteria: Participants were all male and female patients of various ages who were clinically suspected of having pulmonary tuberculosis and who received GeneXpert MTB/RIF testing in addition to sputum smear microscopy. Patients already receiving anti-tuberculosis therapy, those diagnosed with extrapulmonary TB, individuals with incomplete laboratory data, and those who refused to

give their informed consent were not allowed to participate in the research.

Sample Size: The sample size was calculated using Buderer's formula for diagnostic accuracy studies. With an assumed sensitivity of 90% for the GeneXpert MTB/RIF assay, a 95% confidence level (Z=1.96Z = 1.96Z=1.96), a desired precision of 7%, and an estimated prevalence of 41.02% among suspected TB patients in DG Khan (17), the minimum required sample size was 172 patients. To account for potential losses due to inadequate or contaminated specimens, a 10% increase in sample size resulted in a final target of 190 individuals.

Data Collection: Eligible patients were requested to provide sputum specimens in accordance with standard biosafety procedures. Two samples were collected whenever possible (a spot sample and an early-morning sample). In the Allama Iqbal Teaching Hospital microbiology lab, every specimen was prepared for both the GeneXpert MTB/RIF assay and direct smear microscopy. Sputum cultures were also performed according to standard protocol. To guarantee consistent documentation, test findings, clinical information, and demographic traits were entered into a structured proforma. The study was conducted with strict confidentiality in place.

Statistical Analysis: SPSS version 25 was used to analyze the data. Categorical variables were displayed as frequencies and percentages, whilst continuous variables were represented as mean ± standard deviation. Using culture data as the Gold standard, the diagnostic accuracy of GeneXpert was assessed by computing sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and overall accuracy. To gauge diagnostic agreement, kappa statistics and likelihood ratios were also calculated. Statistical significance was defined as a p-value of less than 0.05.

Ethical Approval: The hospital's Institutional Review Board (IRB) gave its approval to the study. All participants, or their legal guardians, provided written informed consent, guaranteeing adherence to ethical guidelines for human research.

Results

The study included 190 patients who were suspected of having pulmonary tuberculosis. The participants' average age was 37.2 ± 16.8 years. In terms of gender, there were 92 (48.4%) females and 98 (51.6%) males. Regarding place of residence, the majority (n = 138; 72.6%) resided in rural areas, while 52 (27.4%) were from urban settings. In terms of marital background, 133 (70.0%) of the patients were married and 57 (30.0%) were single. Analysis of employment status revealed that 118 (62.1%) were farmers, 54 (28.4%) were unemployed, and 18 (9.5%) were engaged in other professions.

Legend: illustrates the sociodemographic profile of the 190 study participants. The majority lived in rural areas, were married, and most were farmers by occupation.

Figure 1 presents the clinical manifestations observed among the study participants. A productive cough with sputum or blood was noted in 148 patients (77.9%), while chest discomfort or pain during breathing or coughing was reported by 176 patients (92.6%). Involuntary weight reduction was documented in 174 individuals (91.6%), and persistent tiredness was the most frequently reported symptom, affecting 178 patients (93.7%).

Legend: Figure 1 highlights the key clinical manifestations among participants, with fatigue and chest discomfort being the most commonly reported symptoms.

Figure 2 demonstrates the comparative findings of sputum smear microscopy and the GeneXpert assay. Out of 108 smear-positive cases, 102 were also confirmed positive by GeneXpert, while 6 yielded negative results. Conversely, among 82 smear-negative cases, 54 were identified as positive by GeneXpert, and 28 were confirmed negative. Overall, GeneXpert detected 156 positive cases and 34 negative cases, highlighting its greater diagnostic yield compared to smear microscopy.

Legend: Table 3 highlights the cross-tabulation between smear microscopy and GeneXpert, showing the enhanced detection capability of GeneXpert, particularly in smear-negative cases.

The diagnostic indices of the GeneXpert MTB/RIF assay are compiled in Table 2 and compared with those of sputum smear microscopy. The assay's capacity to accurately classify both positive and negative cases was proved by its true positive rate of 71.2% and true negative rate of 72.0%. The relative diagnostic strength of GeneXpert was demonstrated by the likelihood ratio for a positive test, which was 2.54, and the likelihood ratio for a negative test, which was 0.40. The proportion of disease cases within the study cohort was 83.7%. Furthermore, GeneXpert achieved a predictive value of 91.8% for positive results, while the predictive value for negative results was lower at 35.5%. The overall correctness of the assay reached 71.6%. A moderately positive correlation was observed between GeneXpert and smear microscopy outcomes, as indicated by Pearson's r ($r = 0.48$, $p < 0.001$).

Legend: Table 2 presents the performance of GeneXpert in diagnostic terms, demonstrating equal sensitivity and specificity, high predictive power in positive cases, and a moderately high correlation with traditional smear outcomes.

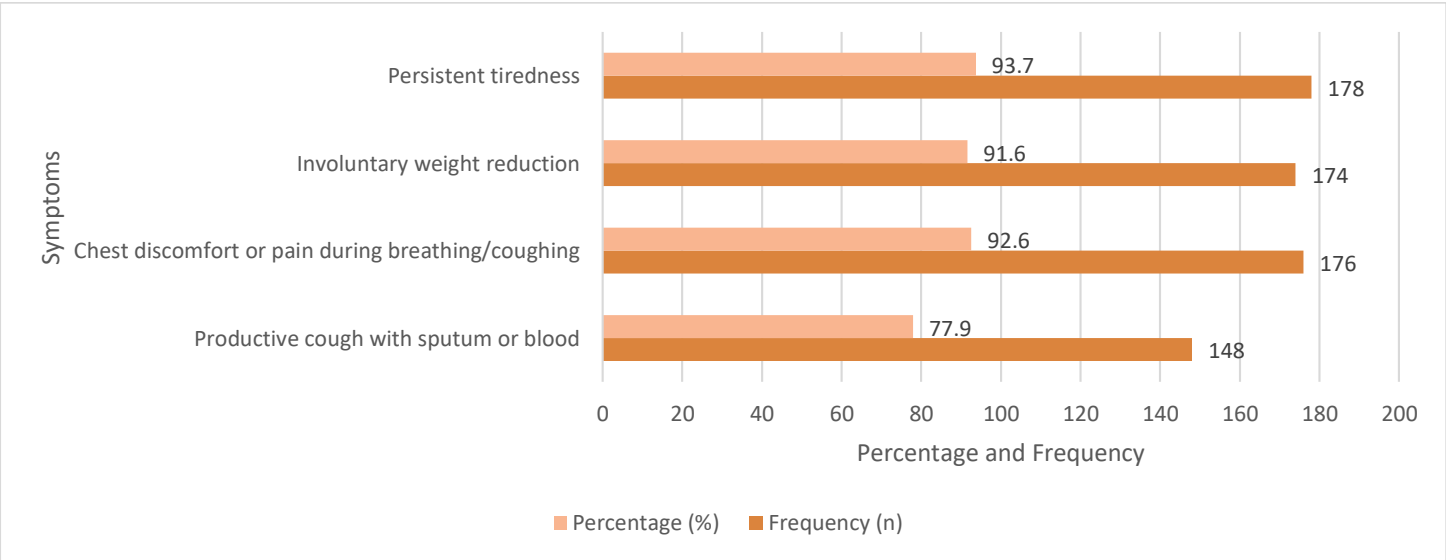

Figure 1: Clinical manifestations among study participants (n = 190)

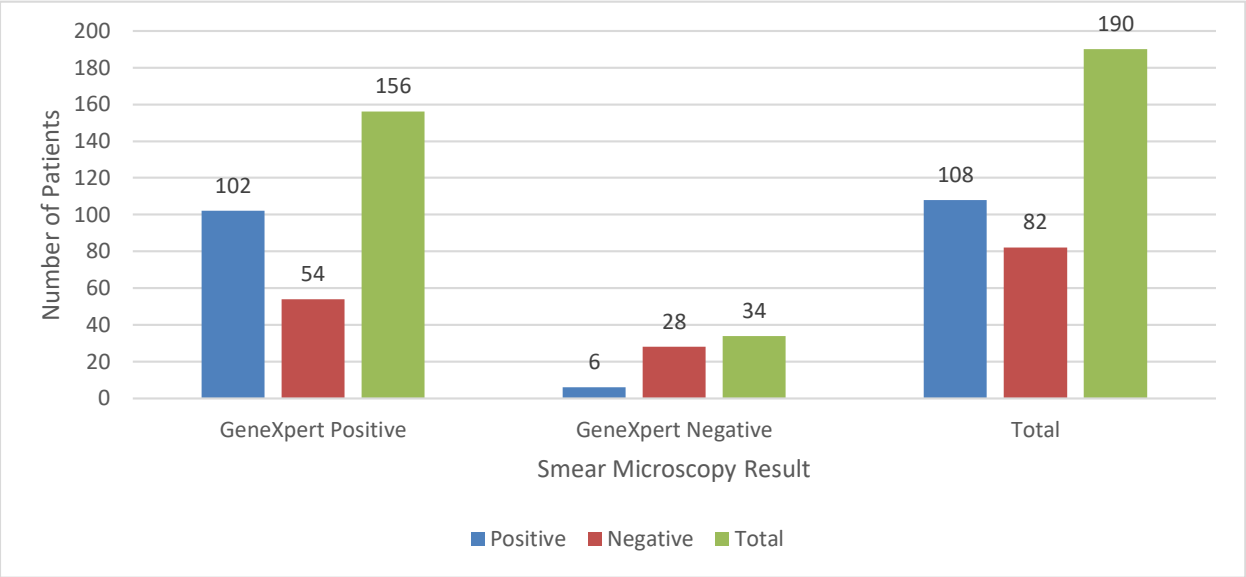

Figure 2: Comparison of sputum smears, microscopy, and GeneXpert assay outcomes (n = 190)

Table 1: Sociodemographic and clinical profile of study participants (n = 190)

Variable	Category	Number (n)	Percentage (%)
Gender	Male	98	51.6
	Female	92	48.4
Place of Living	Rural	138	72.6
	Urban	52	27.4
Marital Background	Married	133	70.0
	Single	57	30.0
Employment Status	Unemployed	54	28.4
	Employed	18	9.5
	Farmer	118	62.1

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

Variable	Value	95% CI
Specificity	72.0%	52.4 – 86.6
Sensitivity	71.2%	62.8 – 78.5
Likelihood ratio (positive)	2.54	1.41 – 4.56
likelihood ratio (Negative)	0.40	0.28 – 0.58
Disease prevalence	83.7%	77.6 – 88.6
predictive value (Positive)	91.8%	86.3 – 95.3
predictive value (Negative)	35.5%	26.2 – 46.0
Accuracy	71.6%	64.7 – 77.6

Discussion

This study has shown that GeneXpert MTB/RIF assay had a moderate sensitivity compared to conventional smear microscopy (65.38%), and specificity (82.35%) was observed, representing its capacity to make correct decisions in relation to true-positive and true-negative cases. The overall diagnostic accuracy was 68.42, indicating that the test is better than the smear. By itself, microscopy is not completely reliable. Notably, the assay revealed a strong positive predictive value (91.84%), indicating that patients with a positive result using GeneXpert are highly likely to be infected with tuberculosis. Conversely, the negative predictive value was quite small (34.15%), highlighting the threat of false negatives and cases being missed. These results underscore the fact that GeneXpert is more effective when used as a confirmatory test for pulmonary tuberculosis, particularly in patients who are smear-negative and where conventional testing can be incomplete. Nevertheless, its shortcomings reveal the need to combine GeneXpert with other diagnostic modalities, including either culture-based modalities or clinical assessments, to guarantee a confirmed Diagnosis.

The diagnostic performance observed in this research was found to be similar to that of GeneXpert in various international and regional studies that have evaluated the tool in pulmonary tuberculosis (18). Similar research has consistently shown GeneXpert to have higher sensitivity than smear microscopy, particularly in cases with low bacillary load (19). Reported sensitivity values in comparable populations often range between 60% and 80%, with specificity exceeding 80%, which aligns with this study's findings (20).

Several studies in South Asian settings with high TB prevalence also confirm that GeneXpert is more reliable than smear microscopy, especially in detecting smear-negative but GeneXpert-positive cases, a trend also noted in the present study (21). In low-resource and high-burden countries, GeneXpert has been reported to significantly enhance early detection rates, improve case confirmation, and contribute to the faster initiation of treatment, as reported by Geatun DA, Layland LE et al. in 2023 (22).

However, other reports have highlighted that while GeneXpert improves diagnostic accuracy, it cannot fully replace culture methods, which remain the Gold standard (23). The relatively low negative predictive value in this study echoes similar concerns raised in global evaluations, indicating that a negative GeneXpert result should not exclude the Diagnosis of TB, particularly in highly suspicious clinical cases (24).

Limitations and Future Directions: Due to the study's six-month time frame and single tertiary care facility setting, the results may not be as broadly applicable as they could be in a multicenter study. It also relied on smear microscopy as the comparator, rather than mycobacterial culture, which could have provided a more robust reference standard. Additionally, resource constraints prevented stratification of results by HIV status or drug resistance patterns, factors that may influence test performance.

Future studies should be designed as multicenter trials with larger sample sizes to enhance external validity. More accurate assessments of diagnostic accuracy will be possible with the inclusion of mycobacterial culture as a gold standard reference. Furthermore, evaluating the cost-effectiveness, turnaround time, and clinical impact of GeneXpert in the local context will provide policymakers with deeper insights into these aspects. To better appreciate its wider therapeutic relevance, research should also examine its role in extrapulmonary TB and in communities with a high incidence of HIV co-infection.

Conclusion

The outcomes of this study demonstrate that the GeneXpert MTB/RIF assay provides superior diagnostic value compared to smear microscopy in detecting pulmonary tuberculosis, particularly in cases that are smear-negative. While its high positive predictive value makes it a reliable tool for confirming TB, the relatively lower negative predictive value indicates that it should be used in conjunction with other diagnostic approaches. Overall, GeneXpert has the potential to enhance early case detection and TB control efforts in high-burden settings, such as Pakistan.

Declarations

Data Availability statement
All data generated or analysed during the study are included in the manuscript.

Ethics approval and consent to participate
Approved by the department concerned. (IRBEC--24)

Consent for publication
Approved

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Conflict of interest

The authors declared the absence of a conflict of interest.

Author Contribution

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All authors reviewed the results and approved the final version of the manuscript. They are also accountable for the integrity of the study.

References

1. Silva S, Arinaminpathy N, Atun R, Goosby E, Reid M. Economic impact of tuberculosis mortality in 120 countries and the cost of not achieving the Sustainable Development Goals tuberculosis targets: a full-income analysis. Lancet Glob Health. 2021 Oct;9(10):e1372–9. https://doi.org/10.1016/S2214-109X(21)00299-0
2. Pezzella AT. History of pulmonary tuberculosis. Thorac Surg Clin. 2019 Feb;29(1):1–7. https://doi.org/10.1016/j.thorsurg.2018.09.002
3. Verma A, Singh V, Singh Chouhan AP, Dutt PK. Tuberculosis: Symptoms, Diagnosis, and the Fight Against Drug Resistance. Biochem Cell Arch. 2024 Oct 2. https://doi.org/10.51470/bca.2024.24.1-S.3337
4. Swaminathan N, Perloff SR, Zuckerman JM. Prevention of Mycobacterium tuberculosis transmission in health care settings. Infect Dis Clin North Am. 2021 Dec;35(4):1013–25. https://doi.org/10.1016/j.idc.2021.07.003
5. Harries AD, Kumar AM, Satyanarayana S, Takarinda KC, Timire C, Dlodlo RA. Treatment for latent tuberculosis infection in low- and middle-income countries: progress and challenges with implementation and scale-up. Expert Rev Respir Med. 2020 Feb;14(2):195–208. https://doi.org/10.1080/17476348.2020.1694907
6. Arif S, Mehboob Q, Arif W. Prevalence and analysis of drug resistance pattern of MDR-TB in retreatment cases at Allied Hospital, Faisalabad, Pakistan. Ann Punjab Med Coll. 2020 Dec 31;14(4):331–5. https://doi.org/10.29054/apmc/2020.994
7. Garg SK, Tiwari RP, Tiwari D, Singh R, Malhotra D, Ramnani VK, et al. Diagnosis of tuberculosis: available technologies, limitations, and possibilities. J Clin Lab Anal. 2003;17(5):155–63. https://doi.org/10.1002/jcla.10086
8. Che-Engku-Chik CE, Yusof NA, Abdullah J, Othman SS, Said MH, Wasoh H. Detection of tuberculosis (TB) using gold standard method, direct sputum smears microscopy, PCR, qPCR, and electrochemical DNA sensor: a mini review. J Biochem Microbiol Biotechnol. 2016 Dec 30;4(2):16–21. https://doi.org/10.54987/jobimb.v4i2.305
9. Chopra KK, Singh S. Newer diagnostic tests for tuberculosis, their utility, and their limitations. Curr Med Res Pract. 2020 Jan;10(1):8–11. https://doi.org/10.1016/j.cmrp.2020.01.004
10. Shah HD, Nazli Khatib M, Syed ZQ, Gaidhane AM, Yasobant S, Narkhede K, et al. Gaps and interventions across the diagnostic care cascade of TB patients at the level of patient, community, and health system: a qualitative review of the literature. Trop Med Infect Dis. 2022 Jul 15;7(7):136. https://doi.org/10.3390/tropicalmed7070136
11. Yadav S, Rawal G, Jeyaraman M, Jeyaraman N. Advancements in tuberculosis diagnostics: a comprehensive review of the critical role and prospects of Xpert MTB/RIF Ultra technology. Cureus. 2024 Mar 31;16(3):e57311. https://doi.org/10.7759/cureus.57311
12. Sahana KS, Prabhu AS, Saldanha PR. Usage of cartridge-based nucleic acid amplification test (CB-NAAT/GeneXpert) as diagnostic modality for pediatric tuberculosis; case series from Mangalore, South India. J Clin Tuberc Other Mycobact Dis. 2018 May;11:7–9. https://doi.org/10.1016/j.jctube.2017.12.002
13. Mishra R, Pal R, Mandal RK. Emerging technologies in tuberculosis diagnosis: a comprehensive review. Int J Res Med Sci. 2025;7(1d):119. https://doi.org/10.33545/26646862.2025.v7.i1d.119
14. Williams V, Calnan M, Edem B, Onwuchekwa C, Okoro C, Candari C, et al. GeneXpert rollout in three high-burden tuberculosis countries in Africa: a review of pulmonary tuberculosis diagnosis and outcomes from 2001 to 2019. Afr J Lab Med. 2022 Aug 30;11(1):1811. https://hdl.handle.net/10520/ejc-ajlab_v11_i1_a1811
15. Sasikumar C, Utpat K, Desai U, Joshi J. Role of GeneXpert in

the diagnosis of Mycobacterium tuberculosis. Adv Respir Med. 2020;88(3):183–8. https://doi.org/10.5603/ARM.2020.0102
16. van den Boom M, Bennani K, Sismanidis C, Gunneberg C, Khawaja L, Safdar MA, et al. 2022 TB programme review in Pakistan: strengthening governance, with better patient diagnosis and treatment. IJTLD Open. 2024 Mar;1(3):136–43. https://doi.org/10.5588/ijtldopen.23.0587
17. Zeb S, Mehmood Z, Basit A, Khan MZ, Khan I, Ullah S. Identification of Mycobacterium tuberculosis in pulmonary and extrapulmonary specimens: a comparative analysis from suspected TB patients. Pak J Chest Med. 2023 Dec 3;29(4):477–85.
18. Arora D, Dhanashree B. Utility of smear microscopy and GeneXpert for the detection of Mycobacterium tuberculosis in clinical samples. Germs. 2020 Jun 2;10(2):81–5. https://doi.org/10.18683/germs.2020.1188
19. Walusimbi S, Bwanga F, De Costa A, Haile M, Joloba M, Hoffner S. Meta-analysis to compare the accuracy of GeneXpert, MODS, and the WHO 2007 algorithm for diagnosis of smear-negative pulmonary tuberculosis. BMC Infect Dis. 2013 Oct 30;13(1):507. https://doi.org/10.1186/1471-2334-13-507
20. Li H, Li Z, Cui X, Liu X, Zhao J, Zhang Y, et al. Mycobacterium tuberculosis detection in diverse clinical specimens by GeneXpert MTB/RIF: a large-scale retrospective study. Infect Drug Resist. 2025 Dec 31;18:2401–13. https://doi.org/10.2147/IDR.S514220
21. Jain J, Jadhao P, Banait S, Salunkhe P. Diagnostic accuracy of GeneXpert MTB/RIF assay for detection of tubercular pleural effusion. PLoS One. 2021 Jun 14;16(6):e0251618. https://doi.org/10.1371/journal.pone.0251618
22. Getahun DA, Layland LE, Hoerauf A, Wondale B. Impact of the use of GeneXpert on TB diagnosis and anti-TB treatment outcome at health facilities in Addis Ababa, Ethiopia, in the post-millennium development years. PLoS One. 2023 Aug 25;18(8):e0289917. https://doi.org/10.1371/journal.pone.0289917
23. Mugenyi N, Ssewante N, Baruch Baluku J, Bongomin F, Mukenya IM, Andama A, et al. Innovative laboratory methods for improved tuberculosis diagnosis and drug-susceptibility testing. Front Tuberc. 2024 Jan 5;1:1295979. https://doi.org/10.3389/ftubr.2023.1295979
24. Hernandez AV, de Laurentis L, Souza I, Pessanha M, Thota P, Roman YM, Barboza-Meca J, Boulware DR, Vidal JE. Diagnostic accuracy of Xpert MTB/RIF for tuberculous meningitis: systematic review and meta-analysis. Trop Med Int Health. 2021 Feb;26(2):122–32. https://doi.org/10.1111/tmi.13525