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Original Research Article

Evaluation of GeneXpert MTB/RIF Diagnostic Performance for Extrapulmonary Tuberculosis

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Abstract: ***Introduction:*** Extrapulmonary tuberculosis (EPTB) represents a major diagnostic challenge in Morocco, accounting for 49% of tuberculosis cases. Conventional methods have limitations in sensitivity and turnaround time. This study evaluates the performance of the Xpert MTB/RIF system in diagnosing EPTB and detecting rifampicin resistance. ***Methods:*** A retrospective study was conducted on 413 samples (cerebrospinal fluid, urine, pus, biopsies) from 402 patients suspected of EPTB between January and December 2021. Compared techniques included microscopy (Ziehl-Neelsen staining), Löwenstein-Jensen culture, and the GeneXpert MTB/RIF test. The diagnostic performance of the latter was calculated against culture (gold standard). ***Results:*** Positivity rates were 9.5% for microscopy, 16.2% for GeneXpert MTB/RIF, and 11.3% for culture. GeneXpert showed a sensitivity of 82.6% and specificity of 92%. Among GeneXpert-positive cases, 41.7% were microscopy-negative, confirming its utility for paucibacillary forms. The highest detection rates were for pus (93%) and osteoarticular samples (100%), while cerebrospinal fluid had the lowest (60%). Rifampicin resistance detected by GeneXpert MTB/RIF was 2.98%. ***Conclusion:*** GeneXpert MTB/RIF significantly improves the diagnosis of EPTB, particularly for microscopy-negative samples. While a negative result does not rule out the diagnosis, its use as a first-line test enables more timely clinical management. Additional studies are needed to evaluate its performance in low-endemic settings.

Keywords: Extrapulmonary Tuberculosis, GeneXpert MTB/RIF, Mycobacterial Culture, Rifampicin Resistance, Morocco.

INTRODUCTION

Tuberculosis remains a major public health challenge in Morocco, ranking among the leading causes of infectious mortality worldwide according to the World Health Organization (WHO). In 2021, Morocco recorded an estimated incidence of 73 cases per 100,000 inhabitants, corresponding to approximately 27,000 new cases, with a mortality rate of 4.5 deaths per 100,000 inhabitants. Extrapulmonary tuberculosis (EPTB), which accounts for 49% of tuberculosis cases in the country, presents particular diagnostic challenges due to its varied clinical presentation and low bacterial load in samples. It affects diverse anatomical sites such as lymph nodes, pleura, bones and joints, the genitourinary system, meninges, and peritoneum [1, 2].

The diagnosis of tuberculosis traditionally relies on smear microscopy, a simple but poorly sensitive method for EPTB. Although culture remains the gold standard, its prolonged turnaround time delays clinical management. Faced with these limitations, WHO endorsed the Xpert MTB/RIF test in 2010,a rapid molecular tool (results in less than two hours) that accurately detects *Mycobacterium tuberculosis* and rifampicin resistance. Since 2014, Morocco has integrated this test into its National Tuberculosis Control Program (PNLAT), and as of 2020, it has replaced microscopy as the first-line method [1].

The objective of this study is to evaluate the diagnostic performance of the Xpert MTB/RIF test for extrapulmonary tuberculosis, compared to conventional methods, to determine its contribution in the moroccan context, where EPTB represents a significant portion of the tuberculosis burden.

MATERIALS AND METHODS

This was a retrospective and analytical study conducted over a 12-month period, from January 1 to December 31, 2021, at the Central Bacteriology

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Laboratory of Ibn Sina Hospital, aiming to evaluate the contribution and performance of the Xpert MTB/RIF system in the diagnosis of EPTB compared to conventional methods. The study included 413 biological samples (pleural, cerebrospinal, peritoneal, and synovial fluids; lymph node biopsies; and other tissues) from 402 patients suspected of EPTB, according to clinical and biological suspicion criteria. Samples were analyzed by direct examination (Ziehl-Neelsen staining for acid-fast bacilli: AFB detection), solid culture (Löwenstein-Jensen), and molecular testing GeneXpert MTB/RIF (detection of M. tuberculosis Deoxyribonucleic Acid: DNA and rifampicin resistance by real-time *polymerase chain reaction: PCR*). A tuberculosis case was considered bacteriologically confirmed when any of the tests (direct examination, culture, or GeneXpert MTB/RIF) was positive, in accordance with the recommendations of the National Tuberculosis Control Program (PNLAT) in Morocco and WHO guidelines.

Statistical Analysis

Patient data, including demographic variables (sex, age) and diagnostic results obtained by different methods (direct examination, culture, and GeneXpert MTB/RIF), were entered and analyzed using Microsoft Excel 2017 software. The diagnostic performance of the Xpert MTB/RIF test was evaluated in comparison with culture, considered as the gold standard, by calculating sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV).

RESULTS

In this study, 413 samples from 402 patients were analyzed. The mean age was 58.3 ± 15.2 years, with a female predominance of 57.3%. The age distribution showed that 13.9% of patients were under 15 years, 50.8% were aged 15 to 65 years, and 35.2% were over 65 years. The sample types reflected the diversity of suspected extrapulmonary tuberculosis cases in our context: 150 cerebrospinal fluids (CSF), followed by 70 diverse biological fluids (ascites, dialysate, peritoneal), 60 tissue biopsies, 50 urine specimens, 40 pus collections, 30 lymph nodes, and 13 osteoarticular samples.

Among the 413 samples tested by GeneXpert MTB/RIF, the *Mycobacterium tuberculosis* complex (MTBC) was detected in 67 specimens, representing a positivity rate of 16.2%, while 346 samples were negative (83.7%). The comparative results of microscopy, GeneXpert MTB/RIF, and culture for all samples are presented in Figure 1. The positivity rates were 9.5% for microscopy, 16.2% for GeneXpert MTB/RIF, and 11.3% for culture.

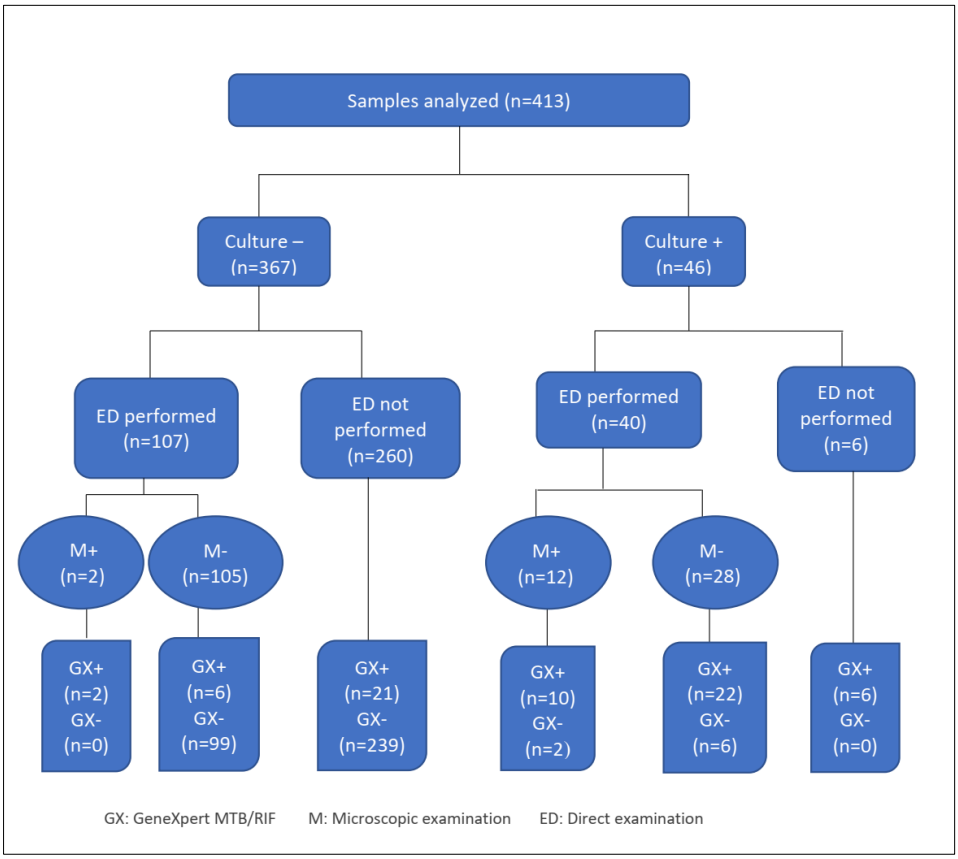

Figure 1: Characteristics of study sample results according to the techniques used

Compared to culture (gold standard), GeneXpert MTB/RIF demonstrated a sensitivity of 82.6%, specificity of 92%, PPV of 56.7%, and NPV of 97.9%.

Among the 367 culture-negative samples, 29 patients (8%) were found positive by GeneXpert MTB/RIF, among which 12 samples (41.4%) corresponded to biopsies.

GeneXpert MTB/RIF identified 28 patients with negative direct examination, representing 41.7% of GeneXpert-positive cases (28/67). Furthermore, culture corrected the diagnosis for 6 patients who were negative by both GeneXpert MTB/RIF and direct examination.

GeneXpert MTB/RIF showed variable detection rates depending on sample type. The highest rates were observed for osteoarticular samples (100%) and pus samples (93%), followed by ascitic and pleural fluids with respective rates of 89% and 81%, while urine and lymph node biopsies showed rates of 80% and 72%. CSF had the lowest detection rate (60%).

Two cases of rifampicin resistance were detected among the 67 specimens confirmed positive by GeneXpert MTB/RIF, corresponding to a resistance rate of 2.98%.

DISCUSSION

The management of EPTB, particularly in developing countries, is hindered by the lack of rapid and sensitive diagnostic tools. Although microscopic examination is widely used as a first-line test, its poor sensitivity (9.5% in our study versus 48% in the literature) limits its effectiveness for early case detection. These findings underscore the importance of adopting more sensitive methods, such as GeneXpert MTB/RIF, to improve tuberculosis diagnosis in resource-limited settings [3].

The GeneXpert MTB/RIF is a rapid molecular assay that simultaneously diagnoses tuberculosis and assesses rifampicin resistance in under two hours. Its use provides significant added value in detecting microscopy-negative cases. In our study, among 413 tested specimens, the overall GeneXpert MTB/RIF positivity rate was 16.2% (67/413). Notably, in patients with initially negative microscopic examination, the GeneXpert MTB/RIF positivity rate reached 41.7%, demonstrating its capacity to identify additional tuberculosis cases. These superior performance metrics compared to conventional methods are supported by multiple studies, including those by Zeka *et al*., (2011) and Opota *et al*., (2016), which similarly highlighted GeneXpert MTB/RIF's efficacy in detecting microscopy-missed cases [4, 5]. However, culture corrected the diagnosis in 6 patients who were negative by both direct examination and GeneXpert MTB/RIF. These findings emphasize the importance of a combined diagnostic approach integrating molecular methods and culture to maximize EPTB case detection.

The performance differences between GeneXpert MTB/RIF, microscopy, and culture can be partially explained by their respective detection limits. Indeed, culture can detect bacterial loads as low as 10 to 100 colony-forming units per milliliter (CFU/ml), while GeneXpert MTB/RIF has a detection limit of approximately 131 CFU/ml. In contrast, microscopy requires a much higher bacterial load, between 5,000 and 10,000 CFU/ml, to yield positive results [6]. Among culture-negative samples, 8% (29/367) were positive by GeneXpert MTB/RIF. Comparable results were reported by Pandey *et al*., (11%) and Iram *et al*., (15%), confirming the ability of GeneXpert MTB/RIF to identify tuberculosis cases missed by culture [7, 8].

Culture negativity despite GeneXpert MTB/RIF positivity may be explained by several factors. First, antibiotic use (fluoroquinolones or anti-tuberculosis drugs) prior to sampling may reduce bacillary viability while leaving DNA detectable by GeneXpert MTB/RIF. Second, aggressive sample decontamination with 4% NaOH may damage or destroy tubercle bacilli, yielding negative culture results while allowing DNA detection by molecular testing. Finally, the paucibacillary nature of extrapulmonary samples, where Mycobacterium tuberculosis tends to form clumps, results in uneven bacillary distribution within the specimen [9].

The negativity of GeneXpert MTB/RIF despite culture positivity can be explained by three principal factors: the presence of PCR reaction inhibitors (such as blood, pus, human DNA, tobacco, or certain medications like corticosteroids), the detection by culture of nontuberculous mycobacteria (which the GeneXpert test does not specifically target), or a bacillary load below the test's sensitivity threshold (131 CFU/ml). These factors highlight the importance of using complementary methods to maximize diagnostic precision [10].

GeneXpert MTB/RIF performance varied significantly across sample types, with a positivity rate of 60% for cerebrospinal fluids compared to 72-100% for other specimens. Specifically, detection rates were highest for osteoarticular samples (100%), pus samples (93%), and ascitic fluid (89%), followed by pleural fluid (81%), urine samples (80%), and lymph node biopsies (72%). This variability primarily reflects differences in mycobacterial load, which tends to be higher in pus-containing or osteoarticular specimens and lower in cerebrospinal fluid and lymph node biopsies. Our results were lower than those reported in the literature, where some studies describe 100% sensitivity for certain sample types. These differences could be related to variations in sampling protocols, sample quality, or population characteristics. These findings highlight the importance of considering sample type and using

GeneXpert MTB/RIF in combination with other methods to improve tuberculosis diagnosis [3-12].

Our results regarding GeneXpert MTB/RIF align with those reported in countries with comparable tuberculosis endemicity. Indeed, diagnostic performances vary significantly depending on tuberculosis endemicity level and the nature of anatomical sites sampled. In low-endemicity countries, the test's sensitivity varies between 82.9% and 95%, while its specificity ranges from 96% to 99%. In contrast, in high-endemicity countries, these performances are slightly reduced, with sensitivity between 80% and 88% and specificity from 95% to 98%. These observations highlight the necessity of interpreting GeneXpert MTB/RIF results in light of local epidemiological context along with sample-specific clinical and microbiological characteristics [13-15].

The NPV obtained for our samples reached 97.6%, confirming the utility of GeneXpert MTB/RIF in ruling out tuberculosis diagnosis. This performance demonstrates the test's ability to minimize false negatives, helping exclude tuberculosis diagnosis particularly in settings where rapid confirmation of disease absence is crucial. These results align with WHO recommendations emphasizing the importance of rapid, accurate molecular tests to optimize tuberculosis diagnosis and clinical management [16].

Our study found a 2.98% rifampicin resistance rate, higher than both the 0.84% reported by Mechal *et al*., (2019) in Rabat Military Hospital and WHO's 1% estimate for Morocco (based on only 4% of notified cases), highlighting the need to improve national resistance surveillance [17, 2].

This study provides relevant insights into GeneXpert MTB/RIF's performance for extrapulmonary tuberculosis diagnosis but has several limitations. First, direct examination was only performed on 147 out of 413 included samples, potentially affecting result representativeness. Second, the study's retrospective design prevented access to patients clinical data and medical history (comorbidities, HIV status, prior treatments), thereby limiting interpretation of discrepancies between diagnostic methods. Furthermore, culture was restricted to solid media without using liquid culture (MGIT), which are more sensitive and faster, potentially underestimating culture's performance as the gold standard. Finally, rifampicin resistance detected by GeneXpert MTB/RIF was not confirmed by phenotypic methods, which would have strengthened result validity.

CONCLUSION

The GeneXpert MTB/RIF test reduces diagnostic delays, improves tuberculosis detection especially for microscopy-negative extrapulmonary forms, and enables early identification of rifampicin resistance. While a negative result does not definitively exclude tuberculosis, its systematic use combined with culture significantly enhances diagnostic sensitivity and specificity, facilitating rapid patient management in line with WHO recommendations. Its integration into tuberculosis control programs, with particular importance in endemic regions, is therefore crucial to optimize disease control and reduce transmission.

REFERENCES

1. National Strategic Plan for TB 2024-2030 [Internet]. [cited 2025 Mar 24]. Available from: https://www.sante.gov.ma/Documents/2023/11/Plan%20strate%C3%ACgique%20National%20TB%202024-2030.pdf
2. World Health Organization. *Global Tuberculosis Report 2021* [Internet]. [cited 2025 Mar 24]. Available from: https://www.who.int/publications/digital/global-tuberculosis-report-2021
3. Tortoli E, Russo C, Piersimoni C, et al. Clinical validation of Xpert MTB/RIF for the diagnosis of extrapulmonary tuberculosis. *Eur Respir J.* 2012;40(2):442-447.
4. Opota O, Senn L, Prod'hom G, et al. Added value of Xpert MTB/RIF compared to sputum smear microscopy to assess tuberculosis transmission risk in low-prevalence settings. *Clin Microbiol Infect.* 2016;22(7):613-619.
5. Zeka AN, Tasbakan S, Cavusoglu C. Evaluation of GeneXpert MTB/RIF for rapid tuberculosis diagnosis and rifampin resistance detection in pulmonary/extrapulmonary specimens. *J Clin Microbiol.* 2011;49(12):4138-4141.
6. Diallo AB, Kollo AI, Camara M, et al. Performance of GeneXpert MTB/RIF® in diagnosing extrapulmonary tuberculosis in Dakar (2010-2015). *Pan Afr Med J.* 2016;25:129.
7. Iram S, Zeenat A, Hussain S, et al. Rapid tuberculosis diagnosis using Xpert MTB/RIF in a developing country. *Pak J Med Sci.* 2015;31(1):105-110.
8. El Baghdadi J, Lazraq R, Ibrahimy S, et al. Primary drug resistance of *Mycobacterium tuberculosis* in Casablanca, Morocco. *Int J Tuberc Lung Dis.* 1997;1(4):309-313.
9. Kohli M, Schiller I, Dendukuri N, et al. Xpert® MTB/RIF for extrapulmonary tuberculosis and rifampicin resistance. *Cochrane Database Syst Rev.* 2018;8:CD012768.
10. Lawn SD, Nicol MP. Development and implementation of Xpert® MTB/RIF for tuberculosis and rifampicin resistance. *Future Microbiol.* 2011;6(9):1067-1082.
11. Theron G, Peter J, Calligaro G, et al. Factors affecting Xpert MTB/RIF performance across different specimen types. *Sci Rep.* 2014;4:5658.
12. Diop SA, Massaly A, Ka D, et al. Use of GeneXpert for tuberculosis diagnosis at Fann University Hospital Infectious Diseases Department. *Pan Afr Med J.* 2016;23:244.

13. Rice JP, Seifert M, Moser KS, et al. Xpert MTB/RIF performance in low-incidence, high-resource settings. *PLoS One.* 2017;12(10):e0186139.
14. Boehme CC, Nabeta P, Hillemann D, et al. Rapid molecular detection of tuberculosis and rifampin resistance. *N Engl J Med.* 2010;363(11):1005-1015.
15. Li S, Liu B, Peng M, et al. Xpert MTB/RIF accuracy varies across tuberculosis prevalence regions: A meta-analysis. *PLoS One.* 2017;12(7):e0180725.
16. Steingart KR, Schiller I, Horne DJ, et al. Xpert® MTB/RIF for pulmonary tuberculosis and rifampicin resistance in adults. *Cochrane Database Syst Rev.* 2014;1:CD009593.
17. Mechal Y, Benaissa E, El Mrimar N, et al. GeneXpert MTB/RIF performance for extrapulmonary tuberculosis diagnosis. *BMC Infect Dis.* 2019;19:1069.

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